International APEC Symposium on APEC-ATCWG Biofuels Network Annual Symposium and Biotrade/Technical Training Workshop

Agricultural Technical Cooperation Working Group

May – June 2011

Symposium documents compiled for the
APEC Agricultural Technical Cooperation Working Group under ATC08/2010A by:

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The project overseers also greatly appreciate the time and valuable contributions of the following keynote speakers and active participants to the compilation of this symposium document:

1. **Australia:** Emeritus Professor Dr. **Peter L. Rogers**, School of Biotechnology and Biomolecular Science, University of New South Wales.
2. **Chile:** Professor Dr. **German Aroca**, School of Biochemical Engineering, P. Universidad Católica de Valparaíso General Cruz, Valparaíso, Chile.
3. **Chile:** Professor Dr. **Rodrigo Navia**, Department of Chemical Engineering, University of La Frontera, Temuco, Chile.
4. **China:** Professor Dr. **Dehua Liu**, Department of Chemical Engineering,Tsinghua University, Beijing, China.
5. **China:** Mr. **Jianbo Liu**, Chairman, Hunan Rivers Bioengineering Co. Ltd., Hunan Province, China.

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6. **China:** Professor Dr. **Tianzhong Liu**, Group leader of Microaglae Biofuels, Qingdao Institute of BioEnergy and Bioprocess Technology Chinese Academy of Sciences, Qingdao, China.
7. **Indonesia:** Dr. **Dadan Kusdiana**, Directorate General New Renewable Energy an Energy Conservation, Ministry of Energy and Mineral Resources, Jakarta, Indonesia.
8. **Indonesia:** Dr. **Oberlin Sidjabat**, Process Technology Division, R&D Centre for Oil and Gas Technology "LEMIGAS", Ministry of Energy and Mineral Resources, Indonesia.
9. **Italy:** Professor Dr. **Roberto De Philippis**, Department of Agricultural Biotechnology, Piazzale delle Cascine 24; I 50144 Firenze, Italy.
10. **Japan:** Professor Dr. **Takashi Watanabe**, Vice Director – Research Institute for Sustainable Humanosphere, Kyoto University**;** Professor – Laboratory of Biomass Conversion**,** Gokasho, Uji, Kyoto, Japan**.**
11. **Korea:** Professor Dr. **Choul-Gyun Lee**, Department of Biological Engineering, Inha University, Korea.
12. **Malaysia:** Associate Professor Dr. **Lee Keat Teong**, School of Chemical Engineering, Universiti Sains Malaysia, Pulau Pinang, Malaysia.
13. **Malaysia:** Dr. **Wan Hasamudin Wan Hassan**, Senior Research Officer / Group Leader, Malaysian Palm Oil Board, Kuala Lumpur, Malaysia.
14. **Mexico:** Mr. **José Luis Arvizu Fernández**, Manager of biomass project, Instituto de Investigaciones Eléctricas, Gerencia de Energías No Convencionales, Morelos, México.
15. **Mexico:** Mr. **Julian Vega Gregg**, Secretary General of the Mexican Network on Bioenergy (REMBIO), Mexico.
16. **Peru:** Mr. **Ari Loebl**, Peruvian Energy Committee, National Industry Society, Peru.
17. **Peru:** Mr. **Pedro Gamio Aita**, Regional Manager for Global Village Energy Partnership for Latin América, Peru.
18. **Philippines:** Professor Dr. **Carmelita Garcia-Hansel**, Director of Research/Professor, Mindanao State University- Main Campus, Marawi City, The Philippines.
19. **Philippines:** Prof. **Nicomedes D. Briones**, School of Environmental Science and Management University of the Philippines Los Baños College, Laguna, The Philippines.
20. **PNG:** Mr. **Andrew Puy**, Faculty of Applied Science, Papua New Guinea (PNG) University of Technology, PMB, Lae, PNG.
21. **PNG:** Mr. **Reilly Nigo**, Faculty of Applied Science, Papua New Guinea (PNG) University of Technology, PMB, Lae, PNG.
22. **Russia:** Mr. **Alex Ablaev**, President, Russian Biofuels Association, Moscow.
23. **Thailand:** Dr. **Gumpanart Bumroonggit**, EGCO Co., Ltd., Bangkok, Thailand.

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24. **Thailand:** Assistant Professor Dr. **Mallika Boonmee**, Department of Biotechnology, Faculty of Technology, Khon Kaen University, Khon Kaen, Thailand.

25. **Thailand:** Dr. **Natanee Vorayos**, Department of Mechanical Engineering, Faculty of Engineering, Chiang Mai University, Chiang Mai, Thailand.

26. **Thailand:** Assistant Prof. Dr. **Wongkot Wongsapai**, Department of Mechanical Engineering, Faculty of Engineering, Chiang Mai University, Chiang Mai, Thailand.

27. **USA:** Dr. **Kulinda Davis**, Director - Grain Processing Enzymes, Verenium Corporation, San Diego, U.S.A.

28. **Viet Nam:** Associate Professor Dr. **Doan Thai Hoa**, School of Chemical Engineering, Ha Noi University of Science and Technology, Ha Noi, Viet Nam.

29. **Viet Nam:** Mr. **Nguyen Duc Cuong**, Director of Center for Renewable Energy and CDM, Institute of Energy, Ha Noi, Viet Nam.

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Introduction

The fluctuating and soaring oil prices in recent years has inevitably affected and pressured APEC economies to seek other alternative approaches in order to satisfy the ever rising demand for energy. The proposed symposium project responds directly to support proposed 2010 work plan of ATCWG-APEC fora activities in the area of food and agriculture of which aim to counter high food prices, ensure food security with sustainable growth, promoting development of next generation sustainable biofuels, and enhance social stability in the Asia-Pacific region. Development of the second and third generation biofuels which utilize raw materials from agricultural sector play crucial roles in preventing and/or mitigating agricultural food crops from being transformed into the controversial first generation biofuels “cash crop”.

The APEC Ministerial meeting in 2008 and 2009 in Peru and Singapore respectively noted that enhancing human security by improving food security in the Asia-Pacific. Food issues are one of the APEC priorities. The SOM FOTC is working closely with relevant APEC fora on an Action Plan on Food Security which is expected to be one of the concrete deliverables for the Ministerial Meeting on Food Security which was held on 16 - 17 October 2010 in Niigata, Japan). The declaration of the following quote during the meeting: “Recognizing the opportunities provided by biofuels, we shared the need to cooperate on developing second-generation biofuels” clearly stated the relevance of the proposed activity of this project to the key priority work plan of APEC. In addition, the expanded agricultural trade directly reflects the interdependence of food among APEC economies. The wastes generated from such food production process could be used for second/third generation biofuels production. ABAC’s Strategic Framework for Food Security noted that there is room for improving biofuels policies so that any possibility of negative impact on food production and prices can be minimized. The proposed project also responds to the priorities and key themes of the APEC Energy Working Group (EWG) which encourages the use of clean fuels and energy technologies.

The goal of the symposium is to promote the awareness and understanding of non-food based biofuels potential of biotrade among the developed and developing APEC economies which accelerate the synergistic cooperation and networking towards the sustainable development and commercialization while mitigating the impact on food security by building on the completed work or underway by the Biofuels Task Force.

This symposium will provide a discussion platform to all APEC economies and link to EWG 01/2006A APEC project (The Future of Liquid Biofuels for APEC Economies Energy Working Group). The working group will invite representatives from APEC economies interested in the research and development of biofuels including lecturers, researchers, staff, trader, as well as policy makers from both public and private sectors as well as students and farmers that are involved in the fields to:

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- Strengthen and expand the established APEC biofuels network by setting up a biofuels symposium on an annual basis to stress the importance of the second and third generation biofuels research development. By the end of the training, participants/trainees will be able to gain the perspective of suitable trading of raw materials, technology, and products among APEC economies with general participants.

- Update the progress on technical, economics, and environmental information and experiences regarding the bioconversion of agricultural and agro-industrial wastes to biofuels among participants of APEC member economies by concentrating on biofuels regulatory cooperation.

- Establish training workshop on biotrade/technical aspects of biofuels by inviting experienced biotraders, academics, and private sectors to train active participants working in the field of biofuels to create initiative and make sure that participants/trainees realize the importance of food security, sustainable trade development, as well as rural community improvement with the accompanied gender-aggregated data by the end of the training. By concentrating on the second and third generation biofuels, the existing agricultural and agro-industrial wastes are used instead of expansion of cultivation area for food crop hence conservation assurance and biodiversity are also achieved.

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Symposium Relevancy to APEC Priorities

The project entitled "APEC - ATCWG Biofuels Network Annual Symposium and Biotrade/Technical Training Workshop" is the continuation of a successful APEC funded project (ATC 16/2009A) entitled "Biofuels from Agricultural and Agro-Industrial Wastes" organized during 24^{th} – 27^{th} May 2010 which resulted in the establishment of APEC biofuels expert/private sector network from Indonesia, Mexico, Australia, United States, Malaysia, Thailand, Viet Nam, Korea, and Philippines. The continuation support from APEC to the APEC biofuels network is a key process of capacity building.

The importance of food security was also outlined in the strategic plan during 2008 – 2011 of World Food Program (WFP) mentioning that millions of people on the global scale are exposed to desperation and hunger due to sky rocketing food and fuel prices which significantly undermine the purchasing power of households. The implementation of any initiative relating to biofuel should be assessed whether such activity make an impact to local staple food security (Final Report on Small - Scale Bioenergy Initiatives, Food and Agriculture Organisation of the United Nations, 2009). There exists sounded arguments from both sides who support and discourage the production of cash crops. Some studies from World Bank and FAO did not reveal significant evidence of correlation between bioenergy production and diminished food supply. However, the development of second/third generation biofuels can alleviate this controversial issue by finding the coexistence of food and biofuels production from non-edible parts of plant. A number of APEC developing economies have the capacity to produce raw materials for second/third generation biofuels production.

Biofuels also provide a significant advantage over the fossil fuels in term of environmental aspect as stated in the Draft Terms of Reference by APEC Biofuels Task Force, EWG31, Singapore, May 2006 and more recently at NREL Golden, CO, October 2008 where cellulosic biomass has been considered as a future resource for energy supply with the potential to replace up to 50% of gasoline. In term of air quality, the application of total ethanol fuel – a type of biofuels results in a drastic decrease of greenhouse gases emission such as CO_2, CH_4 and N_2O by 106% with the additional benefit of safeguarding the soil, water and biodiversity.

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Key Issues & Important Findings

Energy shortage is an undebatable realistic of the world. Biofuels, a substitute of energy, is mostly related with agriculture. For the APEC economies, agricultural sector is considered as one of the most important in term of economics point of view. This APEC funded symposium/training workshop had helped to promote the knowledge exchange both from institute and industries of APEC community, push the advancement of biofuels technology in combination with an existing know-how agricultural technics, and as the results, serve all the economies and people. Based on the distributed questionnaires to the participants of the symposium/training workshop before attending the activity, only 47.9% of them rated their biofuels understanding at high or highest level. This number has shifted to 89.6% after the events.

Biofuels has a very important role to all economics in the world. It is well known that biofuels is currently still too expensive as compared to conventional fuel, thus government support/funding is still required. The updated information obtained from this symposium/training workshop will offer APEC community the shared idea, research findings as well as policy implementation among each other to promote the use of biofuels. The regulatory cooperation can be imposed once the awareness on biofuels production in a sustainable way (environmental, financial and social) has been strengthened.

The current status, future trends, and sustainability of 1st, 2nd, and 3rd generations of biofuels have been discussed with the focus on bioethanol, biodiesel, as well as benefits for rural area people. The keynote speakers and active participants have learned numerous facts on the biofuels other than their expertise. The discussed topics included;

- Current status of biofuels development in respective APEC economies
- Global expansion and trend of first, second, and third generation biofuels
- Lignocellulosic ethanol production in certain APEC economies
- Electricity generation from renewable energy
- Possibilities and limitations of microalgal biofuels production
- Case study of biofuels R&D and regulatory policy in respective APEC economies
- Microbial hydrogen production: state of the art & challenges for industrial exploitation
- Microalgae as a superior feedstock for biodiesel production?
- Enzymatic biotransformation & biofermentation for biodiesel and 1,3-propanediol production
- Legislation analysis and evolution of regulatory system for biofuels
- Novel technology for thermostable & low pH enzymes for commercial biofuels producer.

After the organized training workshop on biotrade/technical aspects of biofuels by the invited experienced academics with insight discussions by biotraders and private sectors, 83.3% of

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the participants agree that education plays important roles in the decision of selecting biofuels while 77.1% realized the negative impact from fossil fuels to the environment by rating both at high to highest level. The importance of biofuels in the future had also come into focus with 91.7% of the supports from participants at high to highest level. Many participants were able to establish biofuels network as evident from 72.9% of them who rated this aspect at high or highest level. The sustainable understanding throughout the community could also be achieved at certain level as shown by 93.8% of the participants who intended to transfer the knowledge gained from the symposium/training workshop to community, friends, and/or students.

A joint international technical research proposal between interested APEC economies (Thailand) and acquire match funding from European Commission (EC) for Framework Programme 7 (FP7) under Professor Dr. Roberto De Philippis, who is currently the Project's European Coordinator, has been established under the project code of FP7-KBBE-2011-5 3.4-01 on BioWASTE – Novel biotechnological approaches for transforming industrial and/or municipal biowaste into bioproducts. The title of the proposed collaborating research project was Waste biovalorization: biofuels and high added value bioproducts from municipal and industrial wastes.

The project had been implemented successfully with attendance of keynote speakers (6 APEC funded) and active participants (2 from each of 11 eligible APEC funded economies) as proposed in the project proposal with the exception of Russia where only 1 active participant had not participated due to passport problem. The feedback from the participants was also quite positive with 97.9% of the participants who rated the overall satisfaction of symposium/training workshop at high or highest level.

Nearly 40% of female (36.1%) had participated in the symposium as keynote speakers as well as active and general participants which was compared to 43.4% from last year. This was not considered as a significant change in female participation within 5% margin of error (35 – 45%). The majority of respondents (89.6%) had education level equivalent to or higher than Master Degree with age range of 20 – 30 years old (37.5%). The respondents opined that the level of opportunities/roles of women in biofuels related agency/organization in their respective economies were relatively at high or highest levels (58.3%), an increase from 52.0% on the previous year, while only 6.3% (in comparison to 4.0% from last year) of respondents rated the level of women participation in biofuels at low level.

Nearly half of symposium participating respondents (47.9%) were awared that second/third generation biofuels development in their respective economies were not at sufficient level. This was compared to the last year level at 57.7%. The respondents realized the importance

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of biofuels in the future at high or highest level (91.7%). In addition, the majority of respondents (64.6%) were willing to select biofuels as an alternative to the normal biofuels at high or highest levels even though the prices of the former fuels were more expensive. This was compared to 61.5% on the previous year.

Suggestions on follow-up steps included the increased participation of government and biofuels trader representatives as well as the extended panel discussion in a more specific issue such as how the improve biofuels implementation in APEC member economies. The participants also wished to form or joining the existing biofuels network task force with specific theme of activity on an annual basis. The future conference topics could be segregated as following; research/technical (academic and research institute), government policy, business/financial aspects (private sector) and production/processing (farmers, manufacturers). Several biofuels network economies such as China, Peru, Chile, and Mexico also expressed their wishes to organize a biofuels network in their respective economies which will prove invaluable as distinct and broadened perspectives can be gained for the participants. If possible, the activity could be alternated between Asia-Pacific economy and Latin America every one or two years.

The keynote speakers/active participants provided the number of contact points or links within or outside their respective economy (not including those from Biofuels Network) as indicated in the Table below. Future activities on biofuels network could make use of these contact points.

Location	**Type of contact point/link (for each participant)**		
	Academic	**Biotraders/ Private sector**	**Policy Maker**
Within your economy	1-20	1-12	1-10
Outside your economy	5-50	1-20	1-10

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International Symposium Program on
APEC-ATCWG Biofuels Network Annual Symposium and
Biotrade/Technical Training Workshop
May 30 – June 4, 2011, Chiang Mai, Thailand

May 30, 2011

08.30 – 09.00	Registration
09.00 – 09.15	Introductory Remarks by the Dean of Faculty of Agro-Industry, Chiang Mai University (Assistant Professor Dr. Charin Techapun)
09.15 – 09.30	Welcoming Remarks by the Vice President of Chiang Mai University (Assistant Professor Dr. Nat Vorayos)
09.30 – 10.00	Opening Remarks by Secretary General of the Office of Agricultural Economics (Mr. Apichart Jongskul)
10.00 – 10.15	Group Picture Coffee Break

########################### Beginning of Session I ###########################

10.15 – 10.45	**Keynote Speaker 1:** Emeritus Professor Dr. Peter L. Rogers, School of Biotechnology and Biomolecular Science, The University of New South Wales, Sydney, Australia **(Topic: New Opportunities for Second Generation Biofuels)**
10.45 – 11.05	**Active Participant 1:** Assistant Professor Dr. Mallika Boonmee, Department of Biotechnology, Faculty of Technology, Khon Kaen University, Khon Kaen, Thailand **(Topic: Towards Lignocellulosic Ethanol Production: Current Development and Collaboration in Khon Kaen University)**
11.05 – 11.25	**Active Participant 2:** Dr. Gumpanart Bumroonggit, EGCO Co.,Ltd., Bangkok, Thailand **(Topic: Progress on Development of Electricity Generation from Renewable Energy in Thailand)**
11.25 – 12.00	**Panel Discussion (Topic: Bioconversion Technology of Agricultural and Agro-Industrial Wastes)** **Chairperson:** Emeritus Professor Dr. Peter L. Rogers **Panel Members:** Assistant Professor Dr. Mallika Boonmee Dr. Gumpanart Bumroonggit

############################ Ending of Session I ############################

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12.00 – 13.00	Lunch

########################## Beginning of Session II ##########################

13.00 – 13.30	**Keynote Speaker 2:** Professor Dr. Choul-Gyun Lee, Department of Biological Engineering, Inha University, Korea **(Topic: Possibilities and Limitations of Microalgal Biofuel Production)**
13.30 – 13.50	**Active Participants 3:** Dr. Dadan Kusdiana, Directorate General New Renewable Energy and Energy Conservation, Ministry of Energy and Mineral Resources, Jakarta, Indonesia **(Topic: Updates on Indonesian Biofuels Development Program)**
13.50 – 14.10	**Active Participants 4:** Dr. Oberlin Sidjabat, Process Technology Division, R&D Centre for Oil and Gas Technology "LEMIGAS", Ministry of Energy and Mineral Resources, Indonesia **(Topic: Role of Rice Straw Potential for Bioenergy in Indonesia as Substituted Gasoline and Reducing of Environmental Problem)**
14.10 – 14.30	Coffee Break
14.30 – 15.00	**Keynote Speaker 3:** Professor Dr. Roberto De Philippis, Department of Agricultural Biotechnology, Piazzale delle Cascine 24; I 50144 Firenze, Italy **(Topic: Microbial Hydrogen Production: State of the Art and Challenges for Its Industrial Exploitation)**
15.00 – 15.20	**Active Participant 5:** Associate Professor Dr. Lee Keat Teong, School of Chemical Engineering, Universiti Sains Malaysia, Pulau Pinang, Malaysia **(Topic: Potential of Using Organic Fertilizer to Culture *Chlorella vulgaris* for Biodiesel Production: A Greener Approach Towards Commercialization)**

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15.20 – 15.40 **Active Participant 6:** Dr. Wan Hasamudin Wan Hassan,
Senior Research Officer / Group Leader
Malaysian Palm Oil Board, Kuala Lumpur, Malaysia
(Topic: Biofuel Implementation in Malaysia: Challenges & Solutions)

15.40 – 16.30 **Panel Discussion (Topic: Policies and Programs in APEC Member Economies to Encourage the Development of the Second and Third Generation Biofuels)**

Chairpersons:
Professor Dr. Choul-Gyun Lee
Professor Dr. Roberto De Philippis

Panel Members:
Dr. Dadan Kusdiana
Dr. Oberlin Sidjabat
Associate Professor Dr. Lee Keat Teong
Dr. Wan Hasamudin Wan Hassan

############################ Ending of Session II ############################

18:30 – 21:00 Welcome Dinner for Keynote Speakers and Active Participants

Note: Biofuels Research Demonstration Workshop at Chiang Mai University is displayed at the back of conference room for interested participants.

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########################### Beginning of Session III ############################

08.30 – 09.00	**Keynote Speaker 4:** Mr. Jianbo Liu, Chairman, Hunan Rivers Bioengineering Co. Ltd., Hunan Province, China **(Topic: Integrated Production for Biodiesel and 1,3-Propanediol with Enzymatic Transformation and Biological Fermentation)**
09.00 – 09.20	**Active Participant 7:** Professor Dr. Dehua Liu, Department of Chemical Engineering,Tsinghua University, Beijing, China **(Topic: The Development Status and Perspective of Biofuels in China)**
09.20 – 09.40	**Active Participant 8:** Professor Dr. Tianzhong Liu, Group leader of Microaglae Biofuels, Qingdao Institute of BioEnergy and Bioprocess Technology Chinese Academy of Sciences, Qingdao, China **(Topic: A Way to Enhance the Biomass Productivity of *Microalgae scenedesmus* in a Novel Hybrid Cultivation system)**
09.40 – 10.20	**Active Participants 9 & 10:** Mr. Pedro Gamio Aita Regional Manager for Global Village Energy Partnership for Latin América, Peru Mr. Ari Loebl, Peruvian Energy Committee, National Industry Society, Peru **(Topic: Biofuels Current Status, Situation, Perspectives, and Development in Peru)**
10.20 – 10.35	Coffee Break
10.35 – 11.15	**Active Participants 11 & 12:** Mr. Andrew Puy & Mr. Reilly Nigo, Faculty of Applied Science, Papua New Guinea (PNG) University of Technology, PMB, Lae, PNG **(Topic: Status and Prospects of Biofuels Development in Papua New Guinea)**

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11.15 – 12.00	**Panel Discussion (Topic: Issues Regarding Future Generation Biofuels on Economic & Environmental Aspects)** **Chairpersons:** Professor Dr. Dehua Liu Mr. Pedro Gamio Aita Mr. Jianbo Liu **Panel Members:** Professor Dr. Tianzhong Liu Mr. Ari Loebl Mr. Andrew Puy Mr. Reilly Nigo

########################### Ending of Session III ###########################

12.00 – 13.30	Lunch

########################## Beginning of Session IV ##########################

13.30 – 14.00	**Keynote Speaker 5:** Professor Dr. Takashi Watanabe, Vice Director – Research Institute for Sustainable Humanosphere Kyoto University Professor – Laboratory of Biomass Conversion Gokasho, Uji, Kyoto, Japan **(Topic: Disintegration of lignocellulosic biomass for 2nd generation biofuels & biorefineries)**
14.00 – 14.20	**Active Participant 13:** Mr. Nguyen Duc Cuong, Director of Center for Renewable Energy and CDM, Institute of Energy Ha Noi, Viet Nam **(Topic: Current Status of Biofuels Development in Viet Nam)**
14.20 – 14.40	**Active Participant 14:** Associate Professor Dr. Doan Thai Hoa, School of Chemical Engineering, Ha Noi University of Science and Technology, Ha Noi, Viet Nam **(Topic: Study on Woody Waste Pretreatment for Cellulosic Ethanol Production)**
14.40 – 15.00	Coffee Break

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15.00 – 15.20	**Active Participant 15:** Professor Dr. German Aroca, School of Biochemical Engineering, P. Universidad Católica de Valparaíso General Cruz, Valparaíso, Chile **(Topic: Biofuels in Chile: Policy, Research and Development)**
15.20 – 15.40	**Active Participant 16:** Professor Dr. Rodrigo Navia, Department of Chemical Engineering, University of La Frontera, Temuco, Chile **(Topic: Advances in biodiesel production in Chile)**
15.40 – 16.00	**Active Participant 17:** Mr. Alex Ablaev, President, Russian Biofuels Association, Moscow **(Topic: Second Generation Fuels in Russia: Bioethanol & Biobutanol. History of the Industry, Experience of Large-Scale Production in Russia, Today's Technology Level)**
16.00 – 17.00	**Panel Discussion (Topic: Third Generation Biofuels Development in the Near Future)** **Chairpersons:** Professor Dr. Takashi Watanabe Professor Dr. German Aroca **Panel Members:** Professor Dr. Rodrigo Navia Associate Professor Dr. Doan Thai Hoa Mr. Nguyen Duc Cuong Mr. Alex Ablaev

############################ Ending of Session IV ############################

Note: Biofuels Research Demonstration Workshop at Chiang Mai University is displayed at the back of conference room for interested participants.

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June 1, 2011

The program listed here is reserved for keynote speakers, active participants, and pre-notified general participants (prior to 1st May 2011 only). For the other general participants, today is designated "As Pleasure" day.

Program A: Tak Field Trip to Ethanol Production Plant

07:00 – 10:30	Trip from Kantary Hill Hotel to Tak
10:30 – 11:20	Lunch Break at Tak
11:20 – 13:00	Trip from Tak to Mae Sod Clean Energy
13:00 – 15:30	Case study at Mae Sod Clean Energy
15:30 – 17:00	Trip from Mae Sod Clean Energy to Tak
17:00 – 19:00	Trip from Tak to Big C Lampang
19:00 – 20:00	Dinner Break at Big C Lampang
20:00 – 21:30	Trip from Big C Lampang to Kantary Hill Hotel

Program B: Chiang Mai Field Trip to Energy Research and Development Institute (ERDI) & Chiang Mai Biogas Landfill

08:00 – 08:30	Trip from Kantary Hill hotel to ERDI
08:30 – 10:00	Case study at ERDI
10:00 – 11:30	Trip from ERDI to Krua Feung Fa Restaurant
11:30 – 12:30	Lunch Break at Krua Feung Fa Restaurant
12:30 – 13:30	Trip from Krua Feung Fa Restaurant to Chiang Mai Biogas Landfill
13:30 – 15:30	Case study at Chiang Mai Biogas Landfill
15:30 – 18:00	Trip back from Chiang Mai Biogas Landfill to Kantary Hill Hotel

Snack & refreshment will be served to all participants in the vans.

The workshop session on 2nd June 2011 will be started in the early afternoon after lunch to allow the participants who participated in each field trip a good rest from previous day activity.

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June 2, 2011

08.30 – 12.00 As Pleasure

12.00 – 13.30 Lunch

##################### Beginning of Workshop Session I #####################

13.30 – 15.00 **Workshop 1:** Emeritus Professor Dr. Peter L. Rogers,
School of Biotechnology and Biomolecular Science,
The University of New South Wales, Sydney, Australia
(Topic: Key Economic Issues for Bioethanol Production)

15.00 – 15.20 Coffee Break

15.20 – 16.30 **Workshop 2:** Professor Dr. Roberto De Philippis,
Department of Agricultural Biotechnology, Piazzale delle Cascine 24;
I 50144 Firenze, Italy
(Topic: Funding opportunities for non-European Countries in the 7th Framework Programme of the European Commission)

16.30 – 18.00 **Workshop 3:** Professor Dr. Takashi Watanabe,
Vice Director – Research Institute for Sustainable Humanosphere

Kyoto University
Professor – Laboratory of Biomass Conversion
Gokasho, Uji, Kyoto, Japan
(Topic: Structural Characterization and Conversion Technologies of Lignocellulosic Biomass)

###################### End of Workshop Session I ####################

Note: Biofuels Research Demonstration Workshop at Chiang Mai University is displayed at the back of conference room for interested participants.

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June 3, 2011

#################### Beginning of Workshop Session II ####################

09.00 – 10.45	**Workshop 4:** Assistant Prof. Dr. Wongkot Wongsapai, Department of Mechanical Engineering, Faculty of Engineering, Chiang Mai University, Chiang Mai, Thailand **(Topic: CDM-Programme of Activities in Biogas Production from Swine Farms)**
10.45 – 11.00	Coffee Break
11.00 – 12.00	**Workshop 5:** Dr. Natanee Vorayos, Department of Mechanical Engineering, Faculty of Engineering, Chiang Mai University, Chiang Mai, Thailand **(Topic: Carbon Footprint of Biofuels in Thailand)**

#################### End of Workshop Session II ####################

12.00 – 13.30	Lunch

######################### Beginning of Session V #########################

13.30 – 14.00	**Keynote Speaker 6:** Dr. Kulinda Davis, Director – Grain Processing Enzymes, Verenium Corporation, San Diego, U.S.A. **(Topic: A Novel Thermostable Broad pH Alpha-Amylase for Ethanol Production)**
14.00 – 14.20	**Active Participant 18:** Mr. Julian Vega Gregg, Secretary General of the Mexican Network on Bioenergy (REMBIO) **(Topic: Current Status and Perspectives of Biofuels in Mexico)**
14.20 – 14.40	**Active Participant 19:** Mr. José Luis Arvizu Fernández, Manager of biomass project, Instituto de Investigaciones Eléctricas, Gerencia de Energías No Convencionales, Morelos, México **(Topic: Challenge and Opportunities of Biofuels Production in Mexico)**
14.40 – 15.00	Coffee Break

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15.00 – 15.20	**Active Participant 20:** Professor Dr. Carmelita Garcia-Hansel, Director of Research/Professor, Mindanao State University- Main Campus, Marawi City, The Philippines **(Topic: Assessing the Potential of Microalgal Isolates for Lipid Production)**
15.20 – 15.40	**Active Participant 21:** Prof. Nicomedes D. Briones, School of Environmental Science and Management University of the Philippines Los Baños College, Laguna, The Philippines **(Topic: Biofuel Development in the Philippines: Socioeconomic and Environmental Challenges)**
15.40 – 16.30	**Panel Discussion (Topic: Commercialization of Second Generation Biofuels)** **Chairpersons:** Professor Dr. Nicomedes D. Briones Dr. Kulinda Davis **Panel Members:** Professor Dr. Carmelita Garcia-Hansel Mr. Julian Vega Gregg Mr. José Luis Arvizu Fernández

############################ Ending of Session V ############################

###################### Beginning of Workshop Session III ######################

16.30 – 18.00	**Workshop 6:** Professor Dr. Choul-Gyun Lee, Department of Biological Engineering, Inha University, Korea **(Topic: Factors That Affects Microalgal Growth)**

###################### End of Workshop Session III ######################

Note: Biofuels Research Demonstration Workshop at Chiang Mai University is displayed at the back of conference room for interested participants.

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June 4th, 2011

09.00 – 10.30	APEC Biofuels Network Brainstorming for Future Collaboration and Activities
10.30 – 10.45	Coffee Break
10.45 – 12.00	APEC Biofuels Network Presentation
12.00 – 13.30	Lunch
13.30 – 14.00	Closing Ceremony

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Introductory Remark by Assistant Professor Dr. Charin Techapun
Dean, Faculty of Agro-Industry, Chiang Mai University
at the International Symposium Program on
APEC-ATCWG Biofuels Network Annual Symposium and
Biotrade/Technical Training Workshop
May 30 – June 4, 2011, Chiang Mai, Thailand

Secretary General of the Office of Agricultural Economics (Mr. Apichart Jongskul)
Vice President of Chiang Mai University (Assistant Professor Dr. Nat Vorayos)
Honorable guests, Ladies and Gentlemen

On behalf of Faculty of Agro-Industry, Chiang Mai University, I would like to thank Secretary General, Office of Agricultural Economics, Ministry of Agriculture and Cooperatives (Mr. Apichart Jongskul) and Assistant Professor Dr. Nat Vorayos, Vice President of Chiang Mai University to be here with us this morning.

This International APEC Symposium on "APEC-ATCWG Biofuels Network Annual Symposium and Biotrade/Technical Training Workshop" is held during 30th May until 4th June 2011 with the warm supports from APEC and Office of Agricultural Economics, Ministry of Agriculture and Cooperatives.

The project entitled "APEC - ATCWG Biofuels Network Annual Symposium and Biotrade/Technical Training Workshop" is the continuation of a successful APEC funded project (ATC 16/2009A) entitled "Biofuels from Agricultural and Agro-Industrial Wastes" organized during 24th – 27th May 2010 which resulted in the establishment of APEC biofuels expert/private sector network from Indonesia, Mexico, Australia, United States, Malaysia, Thailand, Viet Nam, Korea, and Philippines. The continuation support from APEC to the APEC biofuels network is a key process of capacity building.

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There are 3 key objectives of this project: Firstly , to strengthen and expand the established APEC biofuels network by setting up a biofuels symposium on an annual basis to stress the importance of the second and third generation biofuels research development. The participants will be able to gain the perspective of suitable raw materials, technology, and products to be used for biofuels production. Secondly, to update the progress on technical, economics, and environmental information and experiences regarding the bioconversion of agricultural and agro-industrial wastes to biofuels among participants of APEC member economies. Finally, the additional activity of a training workshop in this year will aid further understanding on biotrade or technical aspects of biofuels for active participants working in the field of biofuels to create initiative and realize the importance of food security, sustainable trade development, as well as rural community improvement.

We have invited keynote speakers and active participants who are either representatives from APEC or non-APEC economies namely Australia, China, Indonesia, Malaysia, Mexico, Philippines, Korea, Chile, Russia, Papua New Guinea, Peru, Italy, Japan, Thailand, Viet Nam and The United States of America. These included lecturers, researchers, traders, as well as policy makers from both public and private sectors who are in the field of biofuels research and development.

In this symposium, we will have an opportunity to share knowledge and cost reduction information regarding second generation biofuels. Secondly, to exchange technical, economics, and environmental information and experiences regarding the bioconversion of agricultural and agro-industrial wastes to biofuels. In addition, we hope to establish a strong network of biofuels researchers/scientists and industry leaders working the importance of shifting the research direction from the first generation towards the second generation of biofuels. The symposium activity will be carried out during the first two and a half days which will be followed by workshop training where the participants will have the chance of working out problems set by the invited speakers. There will also be field trips to biofuels production facilities in Chiang Mai and Tak Provinces on the first of June.

I wish to thank Office of Agricultural Economics, Ministry of Agriculture and Cooperatives and APEC for their supports in the preparation of the meeting. Thanks to members of the Organizing Committee, the staff of Faculty of Agro-Industry, Chiang Mai University, who have contributed their untiring efforts to make this symposium possible.

Assistant Professor Dr. Charin Techapun
Dean of Agro-Industry, Chiang Mai University
30th May 2011

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Welcoming Remark by Assistant Professor Dr. Nat Vorayos
Vice President of Chiang Mai University
At the International Symposium Program on
APEC-ATCWG Biofuels Network Annual Symposium and
Biotrade/Technical Training Workshop
May 30 – June 4, 2011, Chiang Mai, Thailand

Secretary General of the Office of Agricultural Economics (Mr. Apichart Jongskul)
Dean of Faculty of Agro-Industry, Chiang Mai University
Distinguished guests, Ladies and Gentlemen

It is my great honor, on behalf of Chiang Mai University, to welcome you all to the International APEC Symposium on "APEC-ATCWG Biofuels Network Annual Symposium and Biotrade/Technical Training Workshop" I thank you all for honoring us with your participation today.

I would especially like to thank Secretary General, Office of Agricultural Economics, Ministry of Agriculture and Cooperatives (Mr. Apichart Jongskul) and all keynote speakers and active participants who will be with us for the next six days.

The fluctuating and soaring oil prices in recent years has inevitably affected and pressured APEC economies to seek other alternative approaches in order to satisfy the ever rising demand for energy. One common strategy employed by several APEC economies is to enact the pro-biofuels policy which encourages the production biofuels from biomass materials such as bioethanol from maize and sugar or biodiesel from vegetable oils which in turn transform these starting raw materials into popular "cash crops". A number of multilateral organizations, including APEC, have voiced their supports for the production of biofuels from non-food feedstock to dissipate the closely tied food and fuel prices at the present time.

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The symposium on ""APEC-ATCWG Biofuels Network Annual Symposium and Biotrade/Technical Training Workshop" will promote the awareness and understanding of the biofuels impact on socio-economic development in urban and rural areas as well as the corresponding environmental aspect among APEC economies with the strong emphasis on bioconversion of agricultural and agro-industrial wastes as well as the forest residues. The commercialization of second generation biofuels will also be examined to realize the potential of its biotrade and respective technology risks and possible opportunities.

I believe APEC has a very strong role to play in facilitating such frameworks at international level and we applaud the current initiative to review biofuels policy within the APEC context.

We are delighted to have this opportunity to host the International APEC Symposium on ""APEC-ATCWG Biofuels Network Annual Symposium and Biotrade/Technical Training Workshop" and sincerely hope that the program we have prepared will offer some fresh perspectives on biofuels potential among APEC economies.

I wish to thank APEC and Office of Agricultural Economics, Ministry of Agriculture and Cooperatives for their support in the preparation of this meeting. Also thanks to members of Faculty of Agro-Industry, Chiang Mai University in organizing this symposium.

Assistant Professor Dr. Nat Vorayos
Vice President of Chiang Mai University
30th May 2011

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Opening Remark by Secretary General of
the Office of Agricultural Economics
(Mr. Apichart Jongskul)
at the International Symposium Program on
APEC-ATCWG Biofuels Network Annual Symposium and
Biotrade/Technical Training Workshop
May 30 – June 4, 2011, Chiang Mai, Thailand

Vice President of Chiang Mai University (Assistant Professor Dr. Nat Vorayos)
Dean of Faculty of Agro-Industry, Chiang Mai University
Distinguished guests, Ladies and Gentlemen

It is an honor and pleasure to be here today and be part of the International APEC Symposium on "APEC-ATCWG Biofuels Network Annual Symposium and Biotrade / Technical Training Workshop". This symposium aims to promote the awareness and understanding of non-food based biofuels potential for biotrade among the APEC economies which accelerate the synergistic cooperation and networking towards the sustainable development and commercialization to all participants.

As the President said before, the increasing of oil prices in recent years has strongly affected and pressured all APEC economies to seek other alternatives sources for energy production such as the second generation biofuels from agricultural and agro-industrial wastes. The raw materials which are of interest to some APEC economies include farm and forest residues, low value timber and energy crops specifically grown for conversion to fuel on marginal agricultural or saline affected land, as well as microalgae.

I am so pleased to see the continual activity set up by the Faculty of Agro-Industry, Chiang Mai University, APEC and the Office of Agricultural Economics, Ministry of Agriculture and Cooperatives. At this time, Thailand also would like to invite all member economies to join the

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APEC biofuels expert/private sector network initiated during the APEC Biofuels from Agricultural and Agro-Industrial Wastes last May and currently the network consisted of 9 economies which are Australia, Indonesia, Malaysia, Mexico, Philippines, South Korea, Thailand, United States and Viet Nam. Thailand expects to use the network as a supportive mechanism to enhance APEC cooperation on the next generation biofuels in order to strengthen both energy security and food security within the region.

Therefore, I wish to encourage every participant to make the most out of this opportunity to equip yourselves with new knowledge and perspectives that International APEC Symposium on "APEC-ATCWG Biofuels Network Annual Symposium and Biotrade / Technical Training Workshop" stores for you during the forthcoming six days.

Finally, I wish to officially declare the International APEC Symposium open.

Thank you.

Mr. Apichart Jongskul
Secretary General of the Office of Agricultural Economics
30th May 2011

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Full Paper

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International APEC Symposium on
"APEC-ATCWG Biofuels Network Annual Symposium and
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May 30th – June 4th, 2011, Chiang Mai, Thailand

ETHANOL AND HIGHER VALUE FERMENTATION PRODUCTS FROM LIGNOCELLULOSIC RAW MATERIALS

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ABSTRACT

The development of second generation processes for bioethanol production from lignocellulosic raw materials is reviewed together with associated international government and commercial support for this technology. Commercial viability is focused on achieving sustainable low cost and high yield biomass production together with reducing pre-treatment and fermentation costs by use of improved enzymes and recombinant yeast and bacteria. Current research initiatives by our group include the genetic characterization and manipulation of high productivity ethanol-producing bacteria (*Zymomonas mobilis*) as well as the use of recombinant *Z. mobilis* for ethanol production from lignocellulosic hydrolysates. Kinetic studies are also reported on a mutant strain of yeast (*Candida tropicalis*) for conversion of xylose to xylitol – a higher value product which could be associated with ethanol production in a typical biorefinery operation. The essential role of government programs to support such biofuels R&D in its early stages is emphasized.

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1 INTRODUCTION

The recent increases in the price of oil, security advantages of increased domestic production, environmental benefits of reduced greenhouse gas (GHG) emissions and the potential for regional development, have all contributed recently to a greatly increased interest in bioethanol. In the longer term, second generation processes based on lignocellulosic materials from agricultural/forestry residues and/or specific high yield biomass energy crops offer greater potential for increased production as they avoid the food vs. fuel conflict. Second generation processes for biodiesel are also under development using lipids produced from large-scale algal cultivation rather than using oils from canola, palm nuts etc. Research over the past decades at the University of NSW has focused on the development of high productivity continuous processes for ethanol production based on the Gram-negative bacterium *Zymomonas mobilis* which has higher specific rates and ethanol yields than the traditionally used yeasts. In addition, considerable flexibility in the genetic manipulation of *Z.mobilis* has been demonstrated with strains now capable of utilizing the C5 sugars xylose and arabinose, as well as the C6 glucose present in cellulosic hydrolysates. The potential also exists for the production of higher value fermentation products from the C5 sugars in the lignocellulosic hydrolysates and these opportunities are currently being explored in the development of new biorefineries.

2 GLOBAL EXPANSION OF BIOFUELS

Brazil has played a leading role in this development after the Government mandated use of ethanol blend fuels in the mid-1980s, and it continues to be a global leader along with the US where ethanol production levels from corn are now similar to Brazilian levels. A major difference between Brazil and the US however, is that the industry cost structure and the use of sugarcane in Brazil as raw material (instead of corn) result in major cost advantages.

It is estimated that ethanol currently has a 16% market share of the total liquid fuel market in Brazil, with more than 40% of all fuels being ethanol blends (usually E10 or E85). Typically, about half of the sugar crop is converted to fuel ethanol. By comparison, it is estimated that ethanol accounts for only 2.6% of the total US liquid fuel market, and utilises about 20% of the current corn crop. Production costs in Brazil are estimated at $US 0.15–0.20 per litre by using large-scale fermentation technology and lower cost raw materials, while in the US production costs are estimated at $US 0.25-30 per litre for similar scale processes.

There has been rapid growth in US ethanol production over the past two decades. With production in the US at 28,000 mL in 2008, this is projected to increase to close to 32,000 mL/a by 2012 with the passing of the *US Energy Policy Act* in 2005. By then biofuels will account for some 5–6% of total fuels in the US, close to the percentage projected by the European Commission, although the latter will involve a much higher proportion of biodiesel with the increasing popularity of diesel-powered vehicles. By comparison, the previous Australian Government has set a low target for biofuels of 350 mL/a by 2010 which is only 1% total liquid fuels. The Australian target includes both biodiesel and bioethanol.

3 FIRST GENERATION ETHANOL PRODUCTION

Ethanol is traditionally considered as a very low cost, high volume fermentation product produced from a range of sugar-based (eg. sugarcane, sugar beet) or starch-based (eg corn, grains, cassava) raw materials. Some pre-treatment of the raw materials may be necessary viz. desalting of molasses or enzymatic hydrolysis of starch, prior to fermentation by yeasts developed for high ethanol and high osmotic pressure tolerance. With such low cost, high volume products, the operating costs of the process are strongly influenced by raw material costs which can be up to 50-60% of these operating costs. This compares with very high value, much smaller volume biopharmaceuticals (therapeutics) – where raw material costs are relatively minor compared to the much higher product recovery and purification costs.

Production technology can vary from large batch fermentations using high yeast concentrations (as in Brazil) to more sophisticated continuous processes using cell recycling and vacuum operation to remove inhibitory ethanol, such as the molasses-based Biostil process at Sarina in Queensland (CSR Ltd). A continuous, multistage processes for conversion of starch waste to ethanol is also in operation in Australia (Shoalhaven Starches, Manildra Group). Processes are designed to achieve as great an economy of scale as possible, with those producing 50–150 mL/a (such as those in Australia) being relatively small scale by international standards. Production capacity is not the only determinant of market size, and can be limited also by the costs of raw material transportation to the plant site. To balance increased transport costs against the advantages of economies of scale, it is often considered that raw materials should only be transported (by road) from within a

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radius of 150 km from the plant. Capital costs for plants of capacities of 100 mL/a are likely to be of the order of $A 70–80 m with possible cost savings by using plant fabrication/construction in countries such as India.

4 SECOND GENERATION BIOFUELS

4.1 Global trends

Projections by the US Department of Energy are that by 2020, the volume of ethanol produced from the conversion of cellulosic materials (biomass) will be twice that produced from corn. However to achieve this goal, a number of technological hurdles will need to be overcome. These can be summarised as:

- The development of cost effective pre-treatment strategies for the various lignocellulosic materials. These include: size reduction, steam explosion and/or concentrated/dilute acid or alkali pre-treatment, as well as the enzymatic hydrolysis of cellulosic sugars to glucose, following the initial conversion of the hemicellulose components to C5 sugars (mainly xylose and arabinose)
- Significant reduction in the costs of producing cellulase enzymes. The outlook is promising with 20-fold cost reductions already reported (Genencor, Novozyme) using protein engineering and gene shuffling techniques to enhance enzyme activities
- The availability of robust recombinant microbes (either yeast or bacteria) for high ethanol yields from the C5 (xylose, arabinose) and C6 (glucose) sugars from the lignocellulosic hydrolysates. The presence of inhibitors such as acetic acid and furfural and its derivatives in the hydrolysates may require appreciable microbial strain conditioning and development
- Product and market development for the non-reactive lignin by-product (approx. 15% total) with potential for its use in paints and adhesives
- Higher value fermentation products (such as biopolymers and low calorie sweeteners) produced in association with ethanol will require the development of mutant and recombinant bacteria/yeast which are capable of high yields and productivities as well as being resistant to potential inhibitors in the hydrolysates.

Pilot-scale studies are being carried out in a number of countries in response to the need for technological 'breakthroughs' and for more accurate cost estimates of second generation processes. These include:

- A large-scale pilot plant which is in operation in Canada, developed by Iogen in association with Shell and PetroCanada. The plant is processing 6 t / day of wheat straw, with yields of up to 350 L /t resulting in about 700,000 L of ethanol per year. Iogen is now proposing to build a commercial scale plant (220 m L/a capacity) using 2,000 t/day wheat straw, with a projected capital cost of $US 200 m
- A Swedish company (Etek) has a pilot plant for processing 2 t/day of softwood, with a yield of 350 L/t . Production costs are estimated at $US 0.45–0.50 per litre for a plant using 600 tonnes of dry matter per day, with a plant capacity of approx. 70 m L/a based on current technology
- Dupont and Danisco have announced a three year investment of $US 140m in a process to convert corn stover and bagasse to ethanol using a recombinant bacterial strain (*Zymomonas mobilis*). Future targets include wheat straw and a variety of energy crops and other biomass sources
- A Dutch company (Nedalco) has announced the establishment of a plant to produce 200 m L/a bioethanol (2.5% European market) using a recombinant yeast and wheat bran as raw material. An investment of 150 m Euro is projected which includes support of 60 m Euro from the Dutch Government.

To stimulate development of appropriate new technologies, the US Department of Energy announced major financial support in 2007.The support included $US 385 m over 4 years to six companies for cost-shared bioethanol processes, $US 200 m for smaller scale biorefineries (10% scale) to evaluate new feedstocks, products and processing technologies for bioethanol and higher value products, and $US 125 m each over 5 years for 3 new Bioenergy Research Centres for basic biomass-related research (US Department of Energy website (2007)). In addition BP has provided $US 500m for biomass related R&D at the University of California, Berkeley.

A concept that is creating considerable interest now in the US and Brazil is that of an 'integrated biorefinery' which can produce a range of fermentation products (including ethanol) from various carbohydrate fractions in the raw material. The idea draws heavily upon the extensive fractionation and range of products produced from hydrocarbons in oil refineries. In the case of the biorefinery, it offers the potential for higher value products, as well as ethanol, resulting in improved overall process economics. Archer Daniel Midlands (ADM) has pioneered this concept in the US, with large-scale

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fuel ethanol production from corn, and associated fermentation processes for amino acids (eg L-lysine), biopolymers, organic acids and antibiotics used in the livestock and poultry industries. Dupont has established an integrated corn biorefinery (ICBR) in which the higher value starch is converted to a biopolymer precursor 1,3 – propan-diol using a GM strain of bacteria (*Escherichia coli*) with the residual lignocellulosic sugars being converted to ethanol using a genetically-engineered strain of *Z. mobilis*. Similar opportunities in Australia could well exist for high protein feeds (and possibly enzymes for use in local industries) in association with larger scale ethanol production.

4.2 Australian situation

The environmental and regional economic drivers behind the global expansion of biofuels are relevant also to Australia, together with the additional issue of increasing total oil imports and recent major oil price rises (Biofuels for Transport: A Roadmap for Development in Australia (2008)). Current production of bioethanol in Australia is in excess of 250 mL/a, sourced from two major producers: CSR at Sarina, Queensland from molasses, and Shoalhaven Starches P/L (Manildra Group) at Bomaderry in NSW from a starch waste stream. A new ethanol plant have been established recently at Dalby in southern Queensland, predo-minantly using sweet sorghum with capacity in the range 50-100 mL/a. New plants have been proposed also in NSW associated with a large-scale dairy operation and it is planned to make direct use of the valuable high protein dried distillers grain (DDG) as a feed supplement.

Support for increased bioethanol production has been provided by mandates proposed by the various Governments for blends up to E10 in 2010–12. (The NSW Government has recently mandated E2 blends increasing to E10 blends by 2011 and this has stimulated increased ethanol production and agreements with oil industry partners). However, the Federal Government's projected removal of the excise tax on imported ethanol in 2011 and reduction in the ethanol fuel tax concession (2011–15) pose an increased risk for investors, in view of the finance required for these new regional industries and the time to achieve cost reductions and establish production capacities and markets. The initial proposed introduction of a Carbon Emissions Reductions Scheme in Australia, and current Government plans to introduce a Carbon Tax on polluting industries (eg electricity production from coal-fired power stations) may counterbalance some of these investor uncertainties.

In addition, there is considerable potential for combining large-scale ethanol production with a strategy for land reclamation, particularly in low rainfall areas. For example, salt tolerant species of fast-growing cane (*Arundo donax*) and eucalypts have been developed which could be used as coppiced plantations for salinity control and as a source of biomass for fuel ethanol and/or electricity co-generation. However it is clear that much more R&D needs to be supported on these new sources of raw materials and their related second generation processes, including the use of flexible pre-treatment plant design and new strains of bacteria/yeast (for both high yield energy crops and agricultural/forestry residues).

4.3 Current Research

Biofuel research at the University of New South Wales over the past two decades has focused on the use of C5/C6 sugars for the development of high productivity continuous processes for fuel ethanol production as well as the use of a mutant yeast strain for conversion of xylose to xylitol.

Recombinant strains of *Z. mobilis* have been used to produce ethanol from C5/C6 sugars (xylose/glucose) typical of those found in lignocellulosic hydrolysates (Davis *et al*., 2005; Rogers *et al*., 2007, 2008). High specific rates have been achieved with yields of 90-95% theoretical and ethanol concentrations in excess of 65 g/L in 48 h from equi-concentration mixtures of glucose and xylose. Stable flocculent strains of *Z.mobilis* have been isolated which are capable of achieving high density, high productivity fermentations.

Various cellulosic raw materials have been evaluated for ethanol production and concentrations of specific inhibitors determined in their hydrolysates .The conclusions of such studies are that relatively high ethanol yields and productivities can be achieved with wheat straw and sugar-cane bagasse. Use of sweet sorghum straw, sugar cane tops and *Arundo donax* (a fast growing tropical bamboo) resulted in some decrease in yields and productivities, while appreciable inhibition was evident in fermentation studies on woody hydroly-sates (Jeon *et al*., 2010). Recent research by our group has focused on the genetic characterization of improved strains of *Z.mobilis*, particularly floccu-lent strains, following publication of the genome of *Z.mobilis* ZM4 (Seo *et al*., 2005) and its refinement (Yang *et al*., 2009).

Microarray analysis has been used by our group to determine whether or not any genetic changes have occurred in improved strains such as flocculent *Z.mobilis* ZM401. As a result it has been found that genes involved in cellulose production (which would enhance cell/cell attachment) have been over-expressed compared to the parent ZM4, while there is reduced expression of those involved in production of flagella proteins (which would influence cell motility)(unpublished results).

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Additional studies by others have also been using metabolic modelling and network analysis to develop strategies for the redirection of the metabolism of *Z.mobilis* towards higher value fermentation products such as succinic acid (Lee et al., 2010).

The production of xylitol from xylose has been studied using a mutant strain of *Candida tropicalis* in which the metabolic step from xylitol to D-xylulose (and subsequent metabolism) has been blocked by disruption of the xylitol dehydrogenase gene (XDH) (EC 1.1.1.9). Following a series of controlled kinetic studies, it was determined that both the parent and mutant strains had similar growth characteristics and biomass yields on glucose medium. However when xylose was provided as the carbon source, good growth occurred for the parent strain ($Yx/s = 0.5$ g/g) while the mutant strain showed evidence of only minimal growth, thereby confirming the negative effect on growth of XDH disruption.

When both glucose and xylose were provided in the medium, rapid growth and uptake of glucose occurred for both strains. Growth continued on xylose for the parent strain with low level xylitol production ($Yp/s = 0.1$ g/g) while close to stoichiometric conversion of xylose to xylitol occurred for the mutant strain ($Yp/s = 0.95$ g/g) with little if any further growth.

Fed-batch studies with the mutant strain of *C.tropicalis* have demonstrated the potential of this fermentation process with a xylitol yield of 0.97 g/g, a productivity of 3.5g/l/h and a final xylitol concentration of 230 g/l. Further evaluations by Dr H-S Shin (LPBio Co., Seoul, Korea) have confirmed that excellent xylitol fermentation characteristics can be achieved with the mutant strain using sugar cane bagasse hydrolysates. The production of xylitol has attracted considerable recent interest as a result of its potential as a low calorie sweetener with anti-cariogenic effects (Granstrom et al., 2007a,b; Akinterinwa et al., 2008). It is currently produced by a high energy process involving the catalytic conversion of xylose to xylitol at elevated temperature and pressures.

Additional biofuels research by our group in collaboration with the Scientific Research Organization of Samoa (SROS) has involved the kinetic characterization of the transesterification /esterification reactions of coconut oil using both enzyme and alkali/acid- based reactions. The processed coconut oil is subsequently blended with diesel - and the glycerol by-product from these conversions may provide a low cost carbon source for further fermentation. Several vehicles have been successfully run in Samoa over the past year on a 50:50 transesterified coconut oil:diesel blend.

Collaborative R&D has been carried out also on these and related biofuel projects with US, German, Korean and Australian commercial partners.

4.4 NCRIS Program

Within the Australian Government's Program of support for a National Collaborative Research Infrastructure Strategy (NCRIS), a Biofuels Sub-Program has been identified with total NCRIS funding of $A 7.7 m over five years (2007–11), and similar matching funds from State Governments, industries and universities. Within the Sub-Program there are three components:

- A micro-algae photobioreactor facility for production of lipids for biodiesel, with total funds of $A 5 m from NCRIS and the South Australian Government being provided to the South Australian Research and Development Institute (SARDI)
- A pilot plant for ethanol production from bagasse with total funds of $A 6.5 m from NCRIS and the Queensland Government to the Queensland University of Technology and Mackay Sugar Pty Ltd
- An integrated R&D program on new second generation technology for conversion of biomass to ethanol, involving NSW, Sydney and Macquarie Universities, with total funds of $A 2.15 million from NCRIS, the NSW Government and the Universities.

NCRIS funding has been provided for infrastructure support and the challenge is now to build on this and develop industry-related projects and international research linkages.

5 CONCLUSIONS

Global ethanol and biofuel production over the last few years has been stimulated by the current high price of oil, fuel security and environmental concerns, as well as the potential to enhance regional development. The increased ethanol production has come largely from sugar and starch-based crops traditionally used also in the food industry. Projected further increases in biofuel production over the coming decades involve the greater use of potentially lower cost biomass (lignocellulosics) as feedstock. Achieving this will require significant technological breakthroughs (pre-treatment, enzymes, recombinant micro-organisms), while opening up the opportunity for the use of agricultural/forestry residues, new high biomass yielding energy crops and associated higher value fermentation products.

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CURRENT STATUS OF BIO-FUELS DEVELOPMENT IN CHILE

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ABSTRACT

Chile imports about 70 % of its primary energy demand, therefore the country has a high dependency of energy supply and subjected to the oil price fluctuations. Five years ago the government defined an Energy Security Policy that is based on three main drivers: diversification of the primary energy matrix, the increase of energy autonomy, minimizing the impacts of international market disturbances, and the use the energy more efficiently. Due to the available land for agricultural activities and the climate conditions, it has been proposed that the future of biofuels in Chile is in the second generation. Today there is not industrial production of biofuels in Chile, except the biogas generated in landfills. A regulatory framework for promoting the development of a new industry based on second generation biofuels is being established, and the main actions taken at a national level are the generation of R&D consortiums that include private companies and universities. It is expected that these initiatives will create the capabilities for the biofuel industry at a national level taking advantage of the resources available in the country.

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1 INTRODUCTION

The Chilean energy matrix is based mainly on oil (40 %), natural gas (24 %) and biomass (16 %). 98 % of the oil and 90 % of the natural gas are imported, if the coal imported is considered, Chile imports about 70 % of its primary energy demand, therefore the country has a high dependency of energy supply and subjected to the oil price fluctuations. Five years ago the government defined an Energy Security Policy that is based on three main drivers: diversification of the primary energy matrix, the increase of energy autonomy, minimizing the impacts of international market disturbances, and the use the energy more efficiently.

The consumption of petrol and diesel in 2010 was 3.3 and 7.6 million cubic meters respectively, and it is expected that in the next few decades the main source of energy in the transport sector will remain being the liquid fossil fuels growing it at least 2 times in the next 15 years. This context has given rise to the assessment and promotion of new energy sources, taking the non-conventional renewable energies an important role. The possibilities of producing Bioethanol, Biodiesel and Biogas has been assessed and represent a great opportunity to develop a new industrial activity using natural resources available in Chile in a sustainable way. The technical and economic information available so far suggest that biofuels could contribute up to 10% of the energy used in the transport sector in 2020, bringing more diversity to the energy matrix.

In 2006, the government created a commission with participation of several ministers and an advisory commission with members from the public and private sector. The report issued by this commission, released in January 2007, contains a large number of recommendations, in summary:

- The promotion policy for the generation of biofuels should be complementary to the strategy of food security.
- The use of land and water for the production of biofuels should not affect its availability for the production of food for domestic consumption and export.
- To ensure the proper use of water. The prevention of water shortage in other productive activities must be a prerequisite for the development of biofuels.
- It is necessary to have an inventory of the available land, to determine the profitability of energy crops, and to estimate the competition for land.
- The strengths on research and development at universities, research institutes and private companies should be assessed.
- The participation of all stakeholders in the value chain, from peasants to consumers should be ensured.
- The participation of small farmers in producing raw materials should be stimulated, and schemes for long-term agreements, to ensure supply to producers, should be generated.
- It is necessary to do research and generate innovative solutions for producing biofuels adapted to national conditions.
- The social and environmental benefits of using biofuels should be promoted.
- All the projects related with biofules must meet the requirements of the system for Environmental Impact Assessment.
- The existing mechanisms for promotion of this new type of industry must be analyzed and adjusted.
- The sustainable management of natural forests should be promoted.
- Quality standards and a regulatory framework for sustainability should be established.
- The blending of biofuels with liquid fossil fuels should be regulated.
- Tax exemptions for biofuels, either pure or blended, should be introduced.

2 FIRST OR SECOND GENERATION

The climate conditions and the soils in the central part of the country makes that Chile has yields over the world average in the feasible crops to be used as a raw materials for first generation biofuels, such us: sugar beet, corn, white wheat and rapeseed. However the maximum available surfaces for these crops are 50.000 Ha, 150.000 Ha. 315.000 Ha, 200.000 Ha respectively. On the other hand in Chile there are 2.2 million Ha of cultivated forest, mainly *Radiate pinus* and *Eucalyptus globulus* and *nitens*, with a potential area of 2.0 million Ha, and there are 15 million Ha of native forest that with a sustainable management could generate important amounts of biomass. Also it must be considered that Chile has a long shoreline more than 7000 Km, with a rich variety of sea weeds or macroalgae that represents a relevant source of biomass. The agro and forest industry generate large amounts of lingocellulosic residues, a high propor- tion of these are use in co-generation facilities. In Chile also there are area with poor soil and good weather conditions that can be used for exotic crops such as jatropha and *higuerilla*. Then the future of biofuels in Chile seems to be in second generation.

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During the last few years the Chilean Government has undertaken a series of actions for the promotion of biofuels in the country, some of them involve new tasks for government agencies related with economy, energy and environment, to set up a regulatory framework, and promote the development of systems, processes and technical tools to deal with this new source/supply of energy.

3 BIOFUEL LEGISLATION IN CHILE

In March 2008, the Congress passed a law on renewable energy sources including biofuels. It required that in 2010 at least five percent of Chile's electricity must come from renewable sources, including biomass. From 2015 on, the proportion must increase by 0.5% annually to reach 10% in 2024.

In May 8, 2008, the Ministry of Economy issued a decree (DS 11/2008) with definitions and quality specifications for the production, import, transport, storage, distribution and marketing of bioethanol and biodiesel. It authorizes blends of ethanol with gasoline and biodiesel with petroleum diesel in a 2% or 5% in volume. It also states that natural persons and companies involved in the production, import, transport, storage, distribution, marketing and blending in biofuels and their facilities must be authorized and registered in the Superintendence of Electricity and Fuels who will audit the procedures and certification.

To promote the incorporation of biofuels into the market, a tax incentive to the use of biofuels was created by exempting them from the tax on fossil liquid fuels. According to the law, there are to two taxes to pay in fuels: the value added tax (19 %), and a specific tax for fuels, gasoline pays 6 UTM/m3 and diesel 1.5 UTM/m3, being UTM a monthly tax unit. In summary, the price paid by the consumer includes about 53% tax for gasoline and around 33 % tax for diesel.

There are two bills at the National Congress related to the promotion of the use of biofuels:

- The promotion of the use of renewable fuels and energies, proposes modifications to the regulation frame of the sector, looking for the fulfillment of norms related to the recognition of non conventional sources and their incorporation into the energetic national matrix.
- The creation of the National Fund for Research and Development of renewable energies, which aims to finance projects and programs.

4 RESEARCH AND DEVELOPMENT ON BIOFUELS PRODUCTION TECHNOLOGIES

In addition to the funds provided for research and development by the Commission for Science and Technology (CONICYT), the government has promoted through the Corporation for Production Development (CORFO), an Agency depending of the Minister of Economy, the creation of consortiums with the participation of private companies, universities and research centers. The aim was to enable the development of domestic production of biofuels in the long term, given the conditions and types of resources available in Chile.

The first call for proposals for the creation of technological consortia on liquid biofuels was done in 2007 and a second in 2009, supported by public funds of about 4 billion Chilean pesos (27 million US$), an unprecedented contribution to research and development activities. As a result, it supported in the first call two consortia involving the major forestry companies and some the major universities in the country, for developing biofuels derived from lingocellulosic materials (BIOENERCEL and BIOCOMSA). In a second call there had been approved funds for three consortiums related with the research and development of biofuels from micro and macro algae (ALGAFUELS, DESERT Bioenergy and BAL Biofuels).

BIOENERCEL S.A.. Its aims are develop, capture and adapt technologies, and to develop human resources, for implementing a national industry for producing biofuels based on the biochemical and thermochemical conversion of lignocellulose, generating mainly bioethanol. The research and development program considers two projects: biomass and bioprocesses. The biomass project is addressing the generation of raw materials including the quantification of forestry residues and the evaluation of new species; the bioprocesses project will be focused on the production of bioethanol from biomass and their value-added byproducts.

The total funds for this consortium are 9,110,000 dollars, and it includes the participation of Universidad de Concepción, Pontificia Universidad Católica de Valparaíso, Fundación Chile, Arauco, Paper and Board Manufacturing Company (CMPC) and Wood Panel Manufacturing Company (MASISA).

BIOCOMSA Inc. is intended to improve the country’s competitiveness developing the basis for second-generation diesel production through the implementation of 8 subprojects that include from characterization and quantification of potentially available raw materials; forest resource prospection;
Management and transport of raw materials, to evaluate processes that involve the treatment of

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lignocellulosic materials to get their optimal yields of syngas and biofuels as final products. The total funds for this consortium are 3,360,000 dollars, including participation of National Oil Company (ENAP), the Wood Consortium and the Universidad de Chile.

ALGAFUELS. Is a technological consortium whose main objective is the production of biofuels from microalgae in the northern regions of Chile. The proposal was submitted by the company BIOSCAN S.A., and has been granted funds up to 5,900,000 dollars on a total cost of 12,500,000 dollars. By the end of the project the expected results are: to have a collection of the best microalgae strains for biofuels production, to have the know-how on how to produce the algal biomass in large quantities, taking advantage of the natural conditions in the desert located in the northern part of Chile, and using CO2 generated by industrial operations, to produce enough amounts of oil to get liquid biodiesel and meals for food and feed, livestock and agricultural industries. The first microalgae production centre should cover 300 ha.

DESERT BIOENERGY Inc. It is a consortium for research and development of microalgae biofuels industry. It was submitted by the Universidad de Antofagasta, and was granted with US $ 4,500,000 dollars, on a total cost of US $ 6,500,000 dollars. The aim of this consortium is the biodiesel production by optimizing biomass production in terms of cultivation time and yield/surface unit, besides developing processes to reduce production costs.

BAL BIOFUELS whose aim is the research and development of biofuels production from the macroalgae *Macrocystis pyrifera*. The proposal was submitted by the company BAL CHILE S.A. and was granted with US $ 6,900,000 dollars, on a total cost of 10,850,000 dollars. The objective is to develop technologies for extensive cultivation of *Macrocystes pyrifera* a marine macroalgae that yields up to 50 % of fermentable carbohydrates, with a productivity of 250,000 kg of material/ha/year. In a five-year program it is expected to: (a) cultivate the raw material at an industrial level; (b) To produce biofuels and specialized chemicals; (c) To sell these products; and (d) To generate by-products from fermentation residues.

5 CONCLUSIONS

Due to the available land for agricultural activities and the climate conditions it has been proposed that the future of biofuels in Chile is in the second generation, it means those that do not compete with raw materials for food production. Today there is not industrial production of biofuels in Chile, except the biogas generated in landfills. A regulatory framework for promoting the development of a new industry based on second generation biofuels is being established, and the main actions taken at a national level are the generation of R&D consortiums that include private companies and universities. It is expected that these initiatives will create the capabilities for the biofuel industry at a national level taking advantage of the resources available in the country.

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ADVANCES IN BIODIESEL PRODUCTION IN CHILE

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ABSTRACT

In Chile, second generation biodiesel research is mainly focused on residual animal fats and waste frying oils. In addition some experiences with residual fish oils have been also carried out at pilot-scale for biodiesel production. However, currently strong research is being performed regarding microalgae as a source of oils for biodiesel production. This oil source seems to be a promising one as it uses no agricultural land (desert land in the case of Chile), it can fix CO_2 from power plants and simultaneously can produce other added-value by-products. As normally all this second generation raw materials do have high free fatty acids (FFA) content, it seems that esterification of these FFA may be promoted by using biocatalysts such as enzymes or whole cell catalysts, enhancing the total biodiesel production yield. In addition, refining processes involving ceramic membranes may be in a near future part of the biodiesel production system as already being tested in Chile a laboratory and pilot-scale

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1 INTRODUCTION

The rapid depletion of non-renewable fossil fuels has pushed the development in new environmental friendly energy sources (Barnwal and Sharma 2005). In addition, the massive use of fossil fuel has contributed to a global warming process, due to the incorporation of new carbon (mainly as carbon dioxide) in the natural cycle (Asif and Muneer 2007). In this context, biofuels are a real alternative as a renewable energy source. Biofuels are referred to as liquid or gaseous fuels mainly used in transport sector, and are predominantly produced from biomass. Biofuels offer advantages such as sustainability, reduction of greenhouse gas emissions, regional development, social structure and agriculture, as well as security of energy supply (Meher *et al*. 2006). In particular, biodiesel or fatty acid alkyl ester (FAAE) is an important biofuel which is produced by transesterification of oils. Historically, first generation biodiesel has been produced using vegetable oils such as rapeseed and soybean. However, agricultural soils are normally preferred to be used for food production. This situation has pushed researchers to seek for novel and alternative sources of oil.

Currently there are three main topics involved in biodiesel development in Chile: First, the oils source is moving from rapeseed oil (first generation) to second generation raw materials such as residual animal fats (AF), waste frying oils (WFO) and microalgae oils (MO). Secondly, novel catalysts involved in the transesterification reaction of oils rich in fatty acids are being researched, including biocatalysts and solid catalysts. Third, novel biodiesel refining processes are currently being evaluated at different development stages for obtaining high quality biodiesel to be used in different industrial niches.

2 NEW SOURCES OF OIL FOR SECOND GENERATION BIODIESEL PRODUCTION

Novel alternative oils for second generation biodiesel in Chile are related to the use of residual animal fats (AF), waste frying oils (WFO) and microalgae oils (MO). Particularly WFO have been successfully tested in a pilot plant constructed during year 2009 in Gorbea, a little town in the South of Chile about 700 km from the capital Santiago to the South. In this pilot plant, biodiesel has been produced classically using NaOH as alkaline catalyst using mixtures of WFO and rapeseed oil as raw material. The biodiesel quality accomplished with the European standard EN14214.

In addition, advances in microalgae biodiesel production in Chile are being strongly supported by the Chilean Government in two different consortia. One of them, Desert Bioenergy S.A. was formed with the main objective to develop a process for biodiesel and other products from microalgae following a global bio-refinery concept. This consortium consists of several Chilean research institutions and industry (Universidad de Antofagasta, Universidad de La Frontera, Electroandina S.A., Prodalmar Ltda., Molinera Gorbea Ltda.).

The advantages of using microalgae for biodiesel production include high growth rates, adaptability to saline conditions and nutrient up-take from treated wastewater, the use of CO_2 from energy plant flue gases, as well as the potential production of high added value co-products.

This consortium is working on selecting microalgae species widespread in fresh and brackish waters of all continents, known to produce higher amounts of hydrocarbons (20–80% on dry weight) and lipids (15-40%). The by-products of these microalgae groups may also include carotenoids, antioxidants, fatty acids, bioactive molecules, and exopoly-saccharides.

Within this consortium an industrial plant will be installed in the North of Chile building upon the advantages of high solar intensity (4.828 kcal/m^2 day), large availability of desert land unsuitable for other agricultural activities (126.049 km^2), and the availability of CO_2 from existing energy production plants.

3 NEW CATALYSTS FOR SECOND GENERATION BIODIESEL PRODUCTION

Biotechnological process in biodiesel production may present some advantages compared to classical chemical biodiesel production (Azócar *et al*., 2010a). As alternative to the conventional NaOH-catalyzed biodiesel production process, enzymes and natural biocatalysts at nanometric scale present the advantages of a high selectivity associated with a high production yield. These kinds of biocatalysts are useful when oils with high free fatty acids content are used as oils source (i.e, WFO and MO). Results regarding the use of enzymes as biocatalyst in biodiesel production using WFO as feedstock have shown high reaction rate and yield, as well as excellent biodiesel quality (Azócar *et al*., 2010b).

However, the enzymatic stability has been one of the major problems for an industrial application. For this reason, several studies have been oriented to find methods to minimize those effects and many studies have been carried out in the field of enzyme immobilization, enzyme modification and protein engineering (Moelans *et al*. 2005; Kim et al., 2006). The high cost associated with the production of enzymes has lead to the concept of "Whole-Cell biocatalyst", where micro-organisms such as bacteria, yeast and filamentous fungi with ability to produce the required enzymes are used in different processes. Here, interesting results have been

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obtained by using whole cell biocatalysts in biodiesel production (Ciudad et al. 2011).

Finally, another alternative is given by the use of heterogeneous catalysis. In this sence, heterogeneous catalysts (in particular metal oxides or carbonates) are one of the most attractive for the production of biodiesel, simplifying and making more economical the purification of the obtained products, avoiding the generation of alkaline and corrosive residues, simultaneously eliminating the wastewater treatment stage.

4 NEW BIODIESEL REFINING PROCESSES

The transesterification of raw oils is never complete. This it is because the transesterification reaction has three important limitations. First, the reaction it is developed in a two-phase system where the mass transference is limited by the insolubility of reactants (oil and alcohol). Second, the transesterification of tg is an equilibrium reaction. Therefore, is not possible to drive the reaction as close to complete conversion of the tg as possible (dubé et al. 2007). Third, the yield of the transesterification reaction depends of the water and ffa content in raw materials, especially when alkali-catalyzed method is used. A high water content and fatty acid in oil cause undesirable saponification reactions and catalyst consumption. The soaps formed retard the phases separation and additional washing steps to remove impurities are necessary include in the refining process (georgogianni et al. 2009). Consequently, the prevailing commercial process for biodiesel production (alkali-catalyzed batch configuration) involves long reaction and residence time, high alcohol/oil molar ratio and catalyst concentration. High energy consumption is required in the purification biodiesel steps and to recover excess amount of alcohol and catalysts during downstream processing (qiu et al., 2010). It is also necessary to employ multiple water washing steps of the product stream, which can give rise to a challenging waste treatment problem in the wastewater stream. This not only affects the yield and cost of production, but also its environmental friendliness.

An important aspect that depends directly on the process performance and associates the control of the reversibility of the reaction is the quality of biodiesel. In accordance with current international standards the free and bonded glycerin content (MG, DG and TG) indicates the quality of biodiesel. High total glycerin content is associated to injector fouling, and valves in motors, decantation in tank storage and reduction the engine's useful life in general. Furthermore, the combustion of these components can lead to the formation of acrolein, a constituent part of photochemical smog and - in addition to oxidation in air - is photolysed to form CO, CO_2 and H_2O as well as unsaturated hydrocarbons (Cao et al., 2008).

An option that grants mentioned advantages it is the technology of membrane processes. This technology is still in the phase of development and more and more applications are being found in raw materials processing. The general principle of a membrane refining system is based on carrying out a reaction while achieving simultaneously the separation of certain components from the reaction medium. In this way it is possible to get an increase the conversion of equilibrium-limited reactions by removing some products from the reactants stream via membranes (see Fig 1).

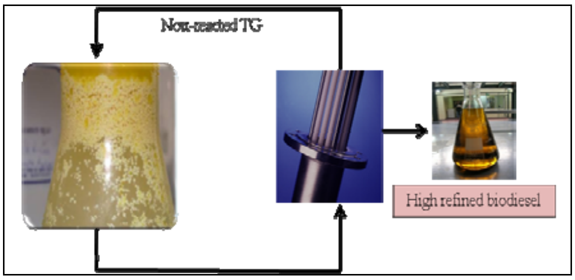

Fig 1: Coupled enzymatic biodiesel production with membrane refining process

The basic parameter governing the separation is molecular size of the interest material and degree of affinity for the membrane, however it is possible to separate free components from a matrix in the form of emulsion when a hydrophilic component form droplets of a size determined in hydrophilic environments. In this way we can use to advantage the immiscibility of the oil in alcohol and generate an emulsified system that allows separate fame from the other components bounded. Refining of biodiesel by a membrane reactor reports that it is possible to selectively permeate methyl esters, methanol and glycerol while it is possible to exclude bound glycerides (tg, dg and mg) from passing through. Hence, the no bound glyceridesc ontent in the permeate stream is minimum, this allows for a high yield of the reaction and a high purity biodiesel (cao et al., 2008).

The performance of membrane processes has been arelated to their resistance to temperature conditions and to chemical attack. Additionally, the easy application and control of these systems has been a special use in refining in the last time. In fact, in these last ten years the crude vegetable oils refining processes seem to be the main application of ceramic membranes. Specially, micro-porous ceramic membranes have found in removal of not wished substances in refined oil such as phospholipids, ffa, waxes and colorant pigments (coutinho et al., 2009).

5 CONCLUSIONS

In Chile, second generation biodiesel research is mainly focused on residual animal fats and waste

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frying oils. In addition some experiences with residual fish oils have been also carried out at pilot-scale for biodiesel production. However, currently strong research is being performed regarding microalgae as a source of oils for biodiesel production. This oil source seems to be a promising one as it uses no agricultural land (desert land in the case of Chile), it can fix CO_2 from power plants and simultaneously can produce other added-value by-products.

As normally all this second generation raw materials do have high free fatty acids (FFA) content, it seems that esterification of these FFA may be promoted by using biocatalysts such as enzymes or whole cell catalysts, enhancing the total biodiesel production yield. In addition, refining processes involving ceramic membranes may be in a near future part of the biodiesel production system as already being tested in Chile a laboratory and pilot-scale.

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THE DEVELOPMENT STATUS AND PERSPECTIVE OF BIOFUELS IN CHINA

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ABSTRACT

With rapid economic development, energy demand increases rapidly in China. China's total energy consumption already occupies the second place in the world. China's oil consumption and net imports increase annually. China national government pays more and more attentions to research and development of biofuels. "Mid and Long Term Development Plan of Renewable Energies" was issued in 2007, where it is indicated that by 2020, annual consumption of fuel ethanol shall reach 10 million tons, and that of biodiesel shall reach 2 million tons in China. Fuel ethanol (E10) project was started since 2001. There are incentives, regulations and financial support for E10. Now there are 5 plants to produce fuel ethanol with the official permission, and 10 provinces are mandatedly using E10 (totally or partly) in China. Non-grain based fuel ethanol is the hotspot in China now. There are dozens of biodiesel production companies in China. The standard of B5 was validated in Feb. 2011. Consumption fax is exempted for the biodiesel with more than 70% waste oil as feedstock. However, the resource of raw material is the bottleneck of the development of biodiesel industry. Central government has some encourage policy in raw material plantation.

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1 BACKGROUND

With rapid economic development, energy demand increases rapidly in China. China's total energy consumption already occupies the second place in the world. China’s oil consumption and net imports increase annually. It is forecasted that in 2020 the oil consumption and import in China will amount to 450 million tons and 250 million tons, respectively, with 55% dependence of petroleum upon petroleum import. (Sources: Xinhua Net, China Petrochemical News) Fig 1 shows the oil consumption in China in the past years. In 2007 China imported 196.8 million tons crude oil. It took 11 years from becoming net importer into importing 100 million tons, but it took only 3 years to increase importation from 100 to 200 million tons.

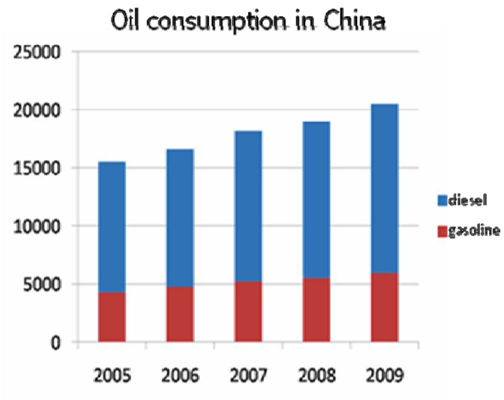

Fig 1: Oil consumption in China

The vehicular oil consumption is one of the main causes of continuous increase consumption of petroleum in China. Compared with the vehicular oil consumption in 2000 which occupied about 1/3 of the total petroleum consumption, it is forecasted that it will rise to 57% by 2020. In recent years, the number of motor vehicles has been growing rapidly in China (Fig 2). Annual auto sale was 1.83M in 1999 and 9.38 M in 2008, increased by 4.8 folds in 10 years.

Fig 2: Growth of automobile in china

The issues have driven Chinese government pay special attention to biofues, which is renewable.

2 DEVELOPMENT STATUS OF FUEL ETHANOL IN CHINA

China started fuel ethanol program in 2001 All the ethanol gasoline (gasohol) in China are E10 (10% fuel ethanol + 90% gasoline). The two state own petroleum companies, Petro China and Sinopec are the only blenders. There are policies for the utilization of fuel ethanol from the central government. The policies include: mandatory mixing of 10 percent fuel ethanol in gasoline in ten provinces to secure the biofuel market; the 5 percent consumption tax on fuel ethanol being waved and the 17 percent value added tax (VAT) being imposed first and then refunded to the fuel ethanol producers; and direct subsidy to fuel ethanol producers to ensure they can make an appropriate level of profit (Source: Law Concerning Testing for the Extensive Use of Ethanol Blended Gasoline for Automobiles and the Regulations Concerning the Conduct of Testing for the Extensive Use of Ethanol Blended Gasoline for Automobiles, 2004). At the very beginning, only four ethanol plants, Jilin Tianhe Ethanol Distillery, Henan Tianguan Fuel Ethanol Co. Ltd., China Resources (Heilongjiang) Co. Ltd. and China BBCA Group Corp. in Anhui province, who produce fuel ethanol mainly from corn or wheat, can receive these incentives. Considering the food safety, Chinese government adopted the principle of No competing for Grains with People and for Land with Grains in order to achieve sustainable development. In 2007, a new fuel ethanol plant was established in Beihai, Guangxi, using cassava as feedstock. Until now, E10 is used in 10 provinces and there are five fuel ethanol plants in China. Fig. 3 shows the fuel ethanol producers and its sales territory. In 2009, consumption was 1.76 million tons. China has become the third biggest fuel ethanol production country in the world.

Fig 3: Fuel ethanol producers and its sales territory

Cellulosic ethanol is one of the hottest topics in China now. Many universities and research centers are researching cellulosic ethanol technology, as well as the fuel ethanol producers, such as Tianguan, COFCO, etc.

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3 DEVELOPMENT STATUS OF BIODIESEL IN CHINA

There are more than 50 biodiesel plants now in China, with total capacity of more than 1,000,000 tons, with waste oil/fat as main feedstock. However, most of them are not on full running now, because of the market and shortage of feedstock. China imports much oil for cooking every year. Fig 4 shows the vegetable oil balance of China in the past years, where we can find 72% of the total consumption of oils are imported.

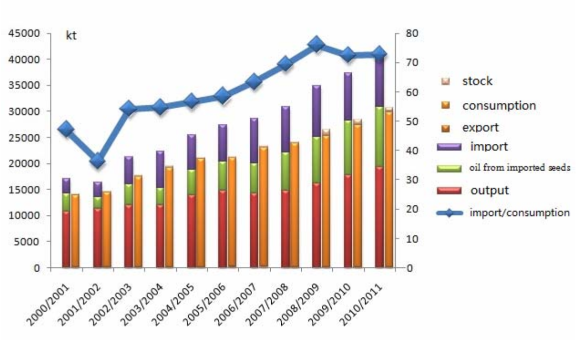

Fig 4: China's Vegetable Oil balance

Chinese government has launched incentives to encourage the plantation of woody oils for biodiesel as feedstock. According to "national bioenergy-directed forest construction program" and "Woody feedstock plantation plan for biodiesel during 11th Five-Year plan", 400 000 hectares of Jatropha curcas will be planted in Yunnan, Sichuan, Guizhou and Chongqing provinces; 250 000 hectares of Pistacia Chinensis will be planted in Hebei, Shanxi, Anhui and Henan Provinces; 50 000 hectares of Cornus Wilsonianawill be planted in Hunan, Hubei and Jiangxi Provinces; 133 333 hectares of Xanthoceras Sorbifolia in Inner Mongolia, Liaoning and Xinjiang provinces.

The development of biodiesel is not as broad as fuel ethanol. However, there are some encouraged policies now. <Biodiesel Blend Stock (BD100) for Diesel Engine Fuels> was issued in May 2007. Standard of B5 was valid in Feb. 2011. Consumption fax is exempted for the biodiesel with more than 70% waste oil as feedstock. 20kt/y biodiesel plant with enzymatic process was built in Yiyang city, Hunan province, China, which is the first biodiesel plant with enzymatic process in China and even in the world. The 60kt/a biodiesel plant of SRCA was constructed in Dongfang,Hainan.

4 DEVELOPMENT PLAN AND PERSPECTIVE FOR BIOFUELS IN CHINA

On September 4th 2007, "Mid and Long Term Development Plan of Renewable Energies" was officially issued in the press briefing of State Council. The general goals of the plan are to improve the proportion of renewable energy in energy structure in China, to resolve the electricity supply problem in remote areas, to improve the working and living conditions in the rural areas, to promote the use of the organic waste for energy utilization and to accelerate the industrialization of renewable energy.

However, the government will obey 3 principles while promoting the development of biofuel project: 1) to take the substitute for petroleum as the key point in the energy-substitute project, and give the priority to the vehicular fuel substitute; 2) to develop biofuel projects in accord with China's actual conditions and ensure the safety of national grain production because China is a country with many people and little land. 3) to insist on multiple feedstocks and non-grain resources and to exploit a special route for biofuel development in China. Therefore, R&D on technologies of non-grain fuel ethanol production is sped up and energy plantation is enlarged.

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A WAY TO ENHANCE THE BIOMASS PRODUCTIVITY OF MICROALGAE *SCENEDESMUS* IN A NOVEL HYBRID CULTIVATION SYSTEM

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ABSTRACT

Microalgae has been thought to the most potential resources for biofuels. R&D of the microalgae biofuel technology has been made in resent years. Though the principle of microalgae biofuels has been testified many years ago, there is a long way need to go for the industrial application as the challenge of economic feasibility. Among the technological steps of the microalgae biofuel production, the supply algal biomass in large quantity at low cost is the domination point. It is determined by the cultivation system. In algal mass culture, open pond and variety of artificial closed photobioreactors (PBRs) were conventionally used. Open pond has been widely chosen for the algal cultivation due to its advantages of cheap in capital investment, easy to build and scale up, but the biomass productivity is very low, around in 5-15g/m^2/d in large scale. Such low productivity would cause the huge land occupancy, Photobioreactors were of high efficiency of biomass productivity, around 15-30 g/m^2/d , but its disadvantages are its expensive for bioreactor building and difficulty to scale-up. So on the view of microalgae biofuel, both open pond and photobioreactors technology had not yet provided a simple solution to incorporate their positive aspects while eliminating most of their short-comings.

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This article described a novel design of hybrid system, in which microalgal broth was pumped to circulate between a pond and one or more PBRs. Such combination caused the improvement of major factors affecting growth, such as illumination; temperature control; CO_2 supply; O_2 remove; nutrient gradient. To evaluate the potential effect of this system, culture experiments both indoor and outdoor had been carried out. In the laboratory, the hybrid displayed a great enhancement of biomass production in terms of areal productivity, in which the maximum areal productivity could reach $32g·m^{-2}·d^{-1}$, with an average speed of $25g·m^{-2}·d^{-1}$ for a batch culture, significantly higher than that obtained from control. Similar effect was demonstrated in outdoor experiments, in which a hybrid was consisted of a 20cm deep open pond with a culture volume of 140L and a 3 cm thick panel reactor with a culture volume of 40L. An average real areal productivity of about $22g·m^{-2}·d^{-1}$ was achieved in outdoor hybrid system, which had increased over 140% to the control of pond and this value is comparable to the control of panel system. In addition, the maximum cell density approaching to $1.5g·L^{-1}$ largely surpassed the traditional pond ($0.4\ g·L^{-1}$).The results presented here indicate a good prospect for using hybrid system culture technology in algal cultivation for the biofuel industry.

1 INTRODUCTION

Microalgae can be used to produce numerous high-value products like carbohydrates, proteins, lipids (unsaturated fatty acids), and pigments (chlorophylls and carotenoids)[1-2] In recent decades microalgae are widely recognized as one of the most promising feedstocks for biofuels.[3-5] One major focus of interest is that algaes are believed to be efficient for biomass production and can be grown in a simple salts medium on a large scale[6]. Currently, two system that have been deployed are based on open pond and closed photobioreactor (PBRs) technologies are used for most algal cultivation[7].Open pond is the cheaper method of large-scale algal biomass production. They usually operate at water depths of 10–30 cm, as at these depths a cell concentration of about 0.5 $g\ L^{-1}$can be maintained, and a productivity of about $60\text{-}100\ mg·L^{-1}·d^{-1}$ (i.e.5-15 $g·m^{-2}·d^{-1}$) have been widely reported[8-9]. However, such productivities are not the rule and cannot be maintained on an annual average[10].Tredici *et al*. cultivated *A. maxima.* (a cyanobacterium) in seawater plus urea as nitrogen source for 1 year, attaining an average yield of 7.4 $g·m^{-2}·d^{-1}$ [11]. Jime´nez *et al*. attained a year-round productivity of 8.2 $g·m^{-2}·d^{-1}$ with *Spirulina* grown in a 450 m^2 and 30cm deep in Malaga, Spain[12].

There are several major limitations causing the low biomass productivity, including light limitation, inefficient mixing and CO_2 deficiencies[13]. Also, contamination risks, evaporative losses, large area of land required and limited to a few strains of algae can be recognized as the limiting factors of open pond system[3, 7-8, 14]. PBRs have higher efficiency and biomass concentration. As stated by Richmond [15], despite closed systems offer no advantage in terms of areal productivity, they largely surpass ponds in terms of volumetric productivity (8 times higher) and cell concentration (about 16 times higher). And the most important advantage of PBRs is that they permit cultivation of algal species that cannot be grown in open ponds[16]. These benefits can be attributed to their shape or design. PBRs such as tubular, flat, panel and column can apply larger illumination area and have higher surface-to-volume ratio (25-125/m)[17]. Moreover, PBRs offer closed systems better to control over culture conditions and growth parameters (pH, temperature, and mixing), prevent evaporation, and reduce CO_2 losses[13, 17]. However PBRs suffer from several drawbacks, including difficulty in controlling culture temperature, cell damage by hydrodynamic stress, bio-fouling and oxygen accumulation, which could slow down the growth of algal cells[14]. Most of important, the cost of biomass production in PBRs may be one order of magnitude higher than in ponds[17]. So, the high cost in capital investment and operating consumption make PBRs difficult to scale up. In conclusion, PBRs and open ponds should not be viewed as competing technologies; the combination of the two systems may get better results. For this reason, here we proposed a hybrid system which is that an open pond is associated with one or more PBRs. Parts of culture medium will be pumped to circulate between the pond and PBRs. This simple strategy can construct a novel cultivation system which collects the both advantages of former systems and provides the opportunity to easily enhance the output of cell mass. In this thesis, we studied the production effect of hybrid system through cultivation of microalgal *Scenedesmus dimorphus.* Finally, the significant enhancement of productivity could be obtained both in laboratory and outdoor experiments compared with the systems of separate open pond and PBRs.

2 MATERIALS AND METHODS

2.1 Algal strain and growth medium

Scenedesmus dimorphus was obtained from the Key Laboratory of Biofuels, (Qibebt,CAS). It was

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grown in conventional BG11 medium. BG11 nutrients (per liter): $NaNO_3$ 1.5 g, KH_2PO_4 0.04 g, $MgSO_4.7H_2O$ 0.075 g, Na_2CO_3 0.02 g, $CaCl_2.2H_2O$ 36 mg, Citric acid 6 mg, Ferric ammonium citrate 6 mg, Na_2.EDTA 1 mg, H_3BO_3 2.86 mg, $MnCl_2.4H_2O$ 1.81 mg, Na_2MoO_4 0.39mg, $CuSO_4.5H_2O$ 0.079 mg, $ZnSO_4$ 0.222 mg, $Co(NO_3)_2.6H_2O$ 0.049 mg[18]. (nys-m-3)The initial culture were inoculated with biomass concentration about 0.1g/L and 0.05g/L for experiments indoor and outdoor, respectively.

2.2 Indoor Hybrid System

To investigate the production effect of hybrid system at laboratory scale, the hybrid system consisted of one modified glass tank (playing the role of open pond) and two columns (playing the role of PBRs), as shown in figure 1. The height of the glass tank was 20cm and the total culture volume was 5.0L. Compressed air/CO_2 mixture (97/3, v/v) was bubbled at the bottom of the tank for mixing and maintaining pH at 7.5±0.5. The tank was wrapped with silver paper exposing only the upper surface to the illumination of 400μmol·m^{-2}·s^{-1}. The illuminated surface area was 0.025 m^2.The column with the diameter of 5cm, containing 0.7L of culture, were bubbled with a air/CO_2 (97/3, v/v) stream to support growth and maintain pH within the range of 7.5~8.5. The illumination of 400μmol·m^{-2}·s^{-1} was provided from one-side of the reactor. The illuminated surface area was thought to be the half of the column surface, 0.028 m^2. A peristaltic pump was used to circulate the microalgal broth from tank to column at a speed of 50 cycles per day. Moreover, experiments of circulated 1 cycle a day and the system without any circulation were also designed as control. All of the cultivation experiments were kept at room temperature of 25±3□.

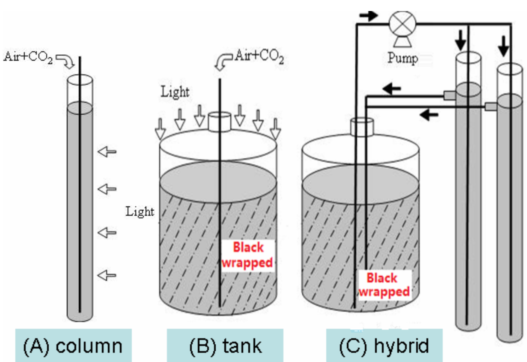

Fig. 1: Schematic graph for indoor cultivation

2.3 Outdoor hybrid system

To investigate the production effect of hybrid system outdoors, experiments were carried out during the summer 2010, at Qingdao (latitude: 36°57′N; longitude: 120°22′E), China. Three plexiplastic panels were arranged face south with the interval distance of 0.5m . Each panel has the length of 1.5m, height of 1.0m and width of 3cm, containing the volume of 40 liters. A perforated plastic tube was inserted in the bottom of panel for the air/CO_2 areation in daytime. heat exchanger was also installed in each panel to keep the temperature of medium automatically. Three ellipse ponds with 1.5m long and 0.5m width were also arranged. CO_2 was injected into the medium through a perforated plastic tube to supply carbon and regulate pH. Each pond has 0.7m^2 surface with running water depth of 20cm, so the total volume of medium in each pond is 140 liters. For the hybrid operation, a pump was connected between a panel and open pond to maintain the circulation.of about 60 cycles a day(Figure 2). Meanwhile, the individual pond and panel without circulation were adopted as control.

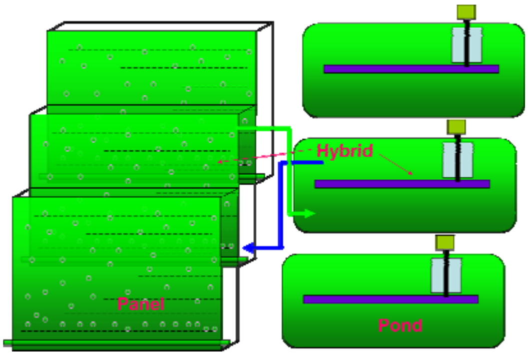

Fig. 2: Schematic graph for outdoor cultivation

In July, all the cultivation system were kept the temperature no more than 35℃, and in September, that was controlled below 30℃.

3 ANALYTICAL METHODS

The algal biomass density was determined by dry weight (g·L^{-1}) at 24h intervals according to method of Richmond *et al*[19].

The occupied land areal biomass density (g·m^{-2}) was calculated as following:

$$\text{Areal biomass density} = \frac{(C_t - C_0)\ V}{S} \qquad (1)$$

Where C_t is the dry weight at time t and C_0 is the dry weight of initial inoculums. V is the total volume of medium and S is the land area occupied of the each reactor(pond, panel). For the hybrid system, S is the total land area of the pond and panel which are incorporated in it. As the results, the areal biomass productivity (g·m^{-2}·d^{-1})of different systems was calculated according to the following equation:

$$\text{Productivity} = \frac{\text{areal biomass density}}{\text{culture time (days)}} \qquad (2)$$

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4 RESULTS

4.1 Comparison of growth in tank, column and hybrid systems indoors

The comparison the amounts of areal biomass output among the different systems in laboratory experiments showed in Fig.3. In contrast with tank, in which deep water layer made most culture broth lack of illumination, the areal biomass output obtained from column was great higher. However neither the column nor the tank could reach the level of hybrid in which per m^2 illuminated area could accumulate 175g algal biomass after 7 day' cultivation.

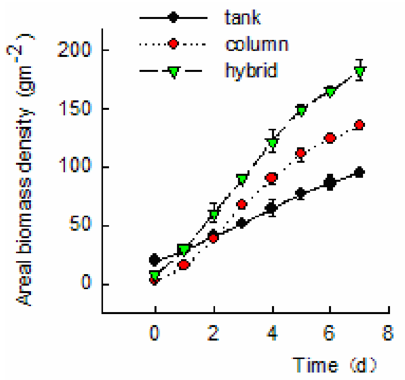


Fig.3: Comparison of areal biomass density in various cultivation systems (tank, column, and hybrid).

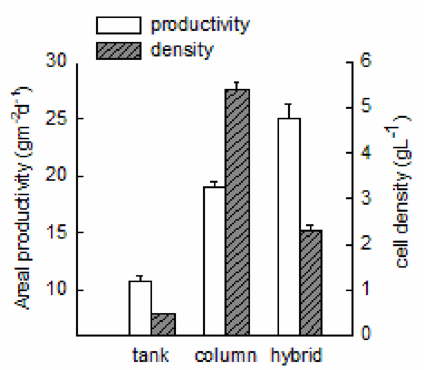
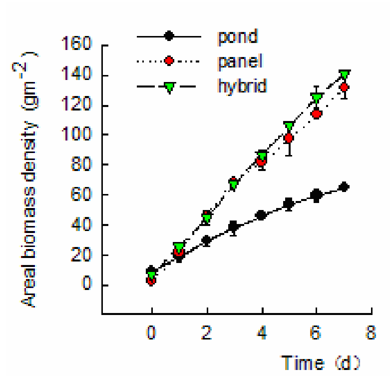


Fig.4: Maximum cell density and average areal productivity in various cultivation systems (tank, column, and hybrid).

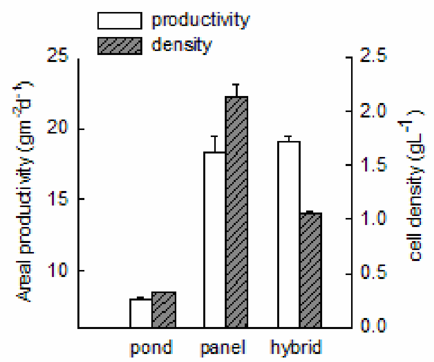
Consequently, the average areal productivity of hybrid system approaching 25 $g{\cdot}m^{-2}{\cdot}d^{-1}$ largely surpassed the individual system of tank and column in which the productivity were 10.8 $g{\cdot}m^{-2}{\cdot}d^{-1}$ and 18.9 $g{\cdot}m^{-2}{\cdot}d^{-1}$, respectively (Fig.4). In addition, the growth in column could obtain the maximum cell density (5.4 $g{\cdot}L^{-1}$) after 7 day' cultivation which attained a 10-fold greater than it of tank. Although the hybrid system had a lower maximum cell density relative to column, it also revealed a 5-fold increase to the value of tank (Fig.4).

4.2 Comparison of growth in pond, panel and hybrid systems outdoors

The similar findings were obtained in the outdoor experiments in July and September. As showed in Fig.5, the hybrid of pond and panel resulted in a significant increase in the areal biomass density. In July, after 7 day' cultivation, the areal biomass density obtained in hybrid system could reach 141 gm^{-2} which had raised more than 140% in relation to pond and this value is little greater than the control of panel system (131 gm^{-2}). In September, all of the culture systems made better performances due to the better temperature control. The areal biomass density could attain 74gm^{-2} and 162gm^{-2} in the individual systems of pond and panel. It was also found that the hybrid made a great enhancement of biomass output also reaching 160 gm^{-2} for the batch culture compared with the control.



(A)



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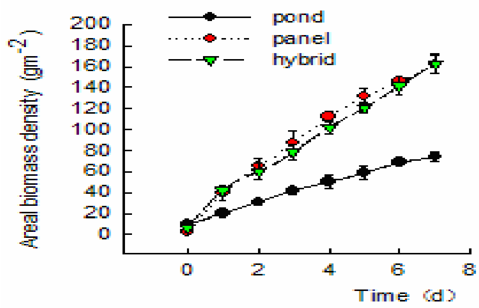

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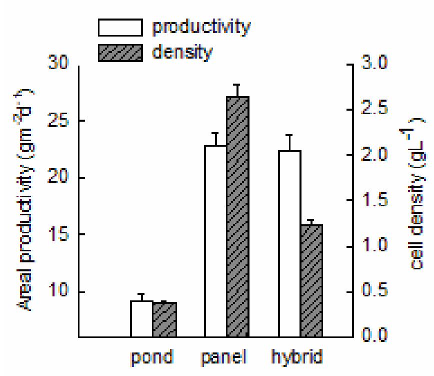

(D)

Fig.5: Areal biomass productivity, maximum cell density and average areal productivity of *Scenedesmus dimorphus* grown in various cultivation systems (pond, panel, and hybrid) outdoor. (A, B) operated in Jul; (C, D) operated in Sep.

Interestingly, although the maximum cell density in the panel reaching 2-3 g·L^{-1} largely surpassed pond (7 times higher) and hybrid (2 times higher), it offered no advantage in terms of areal productivity. The average areal biomass productivity in the pond and panel were 8-9 g·m^{-2}·d^{-1} and 18-22 g·m^{-2}·d^{-1}, respectively, which was less than that in hybrid system of 19-23 g·m^{-2}·d^{-1} in July and September.

4.3 The effect of circulation on biomass production

In order to investigate the effect of circulation on biomass production, three circulation rates of no circulation, 1 cycle per day and 50 cycles per day were adopted for the hybrid operation in laboratory. Figure 6 shown that areal productivity of the three circulations. it can be found that such circulation deadly enhance the growth of algal cells. The areal biomass density accumulated in the hybrid system with a circulation of once a day (121g·m^{-2}) only has a little increase to that of in "no-circulation" (108 g·m^{-2}) after 7days culture. While, the average areal biomass productivity and maximum cell density in the 50cycles/d experiment reached 25.1 g·m^{-2}·d^{-1} and 2.3 g·L^{-1}, which increased more than 60% of the value obtained in control experiments of "no-circulation".

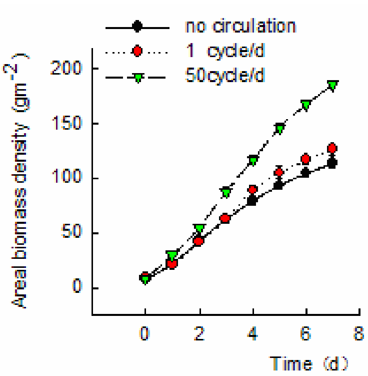

Fig.6: Effect of circulation rates on areal biomass density in a batch culture for hybrid system.

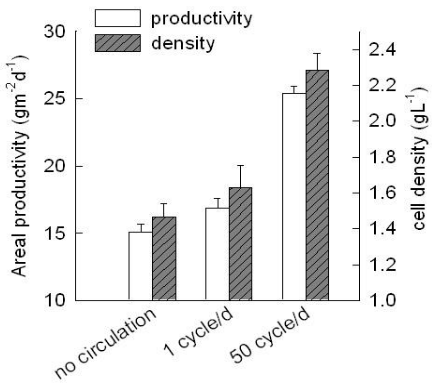

Fig.7: Effect of hybrid system on maximum biomass density and average areal productivity in a batch culture.

5 DISCUSSION

Generally, growth of microalgae was influenced by many factors, such as irradiance, culture temperature, mixing, nutrient supply, predator control, etc[20]. But conventional open pond and variety closed PBRs systems used separately could not optimize all of the factors at large scale[7, 14, 17, 21]. Consequently, a conceptual hybrid production system was proposed in previous literatures[13, 22].

Demirbas suggested that a combination of both systems is probably the most logical choice for cost-effective cultivation of algae for biofuels[22].

Brennan *et al.* illustrated a kind of hybrid system with two-stage cultivation[13]. The first stage is in PBRs where controllable conditions minimize contamination from other organisms and favour continuous cell division. The second phase for oil accumulation, which is aimed at exposing the cells to nutrient stresses, is ideally realized in open ponds[13].

Huntley *et al.*used such a two-stage system for cultivation of Haematococcus pluvialis successfully[23].

Rodolfi *et al.* also demonstrated such a hybrid system was valuable. According to their results, *Nannochloropsis* sp. F&MM24 had the potential for an annual oil production of 20 tons per hectare in the Mediterranean climate[4].

However, these kinds of hybrid systems as mentioned before could not incorporate the positive aspects of both open pond and PBRs at all times in a batch culture. Moreover, they merely divided the cultivation into two distinct stages in open ponds and PBRs leaving most of their shortcomings un-eliminated. This was the reason why, even in optimally operated pond or PBRs, it

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was still a hard work to breakthrough the bottleneck of algal production.

In our study, a novel hybrid system was proposed, in which pond and PBRs were incorporated into a system through the circulation. Both the results of laboratory (Fig.3, 4) and outdoor (Fig.5) had confirmed the significant impact of hybrid system on the yield of biomass. In the laboratory, the maximum areal productivity could reach $32g \cdot m^{-2} \cdot d^{-1}$(Fig.5C), with an average speed of $25g \cdot m^{-2} \cdot d^{-1}$ for a 7 day' cultivation. In outdoor experiments, hybrid also offered a great enhancement in which an average productivity of about $15g \cdot m^{-2} \cdot d^{-1}$ was achieved. Although this value was merely approaching the common level in relation to the published algal productivities[15], the biomass output obtained from our hybrid had increased over 60% and 40% to the control of pond and panel, respectively. The results implied that a more remarkable enhancement of productivity would be achieved under more favourable environment conditions.

Subsequently, the reason of hybrid promoting yield had been illustrated in Fig.3-7. It should be pointed out that the circulation was the major factor to make effect. In the hybrid system, the mixing and the mass transfer rate were improved by the circulation. Consequently, the major factors affecting growth (e.g. temperature control; CO_2 supply; O_2 remove; nutrient gradient) were ameliorated.

On the other hand, to reach maximum growth rates, algae need to be photo-saturated. However, due to the deep water layer of pond and the high cell concentration of PBRs, the mass cultures were always typically photo-limited. Moreover, in the beginning of PBRs' cultivation, the cell density would be so low that most of the impinging photons would pass through unused.

In the hybrid system, the circulation would provide a totally different flow pattern compared with pond or PBRs, which led to a certain movement of cells from zones of light-excess (PBRs) to zones of light-limited (pond). As a result, most cells were better exposed to light, it was anticipated that photosynthesis took place more significantly to attain higher biomass productivity.

Last but not least, in the hybrid system, pond provided about 75% volume of the total culture which causes it easy to scale-up and relative low-cost, comparing with the system of complete expensive PBRs. Additionally, to some algal species susceptive to the competitors, predators and pathogens, we could substitute the open pond with enclosed reservoir which would guarantee a relatively long and stable cultivation to achieve the comparable productivity.

In conclusion, the feasibility of a hybrid system for algal cultivation of *S. dimorphus* was demonstrated in our laboratory and outdoor experiments. The combination of pond and PBRs was probably the most logical choice for cost-effective cultivation of microalgae. The analysis of the culture ratio pond / PBRs and the circulation speed of the system were the two key factors need to be determined in further work.

6 ACKNOWLEDGEMENTS

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UPDATES ON INDONESIAN BIOFUEL DEVELOPMENT PROGRAM

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Since October 1st, 2008, the government of Indonesia has gradually mandated the utilization of biofuel in transportation, industry and power generation sectors. The objective of biofuel implementation program, in addition to enhance energy security, is reducing fossil fuel import, poverty alleviation, job creation and reducing CO2 emission. Annual production capacity for biodiesel production has reached more than 4 million ton while bioethanol is around 200 thousand ton.

In fact, biodiesel development in Indonesia is not just begun. It was dated back to the early 90th when PPPTMGB Lemigas, one of R&D Centre within Ministry of Energy and Mineral Resources, carried out research on biodiesel process and trial for its utilization. The activities were then expanded by other research institutes in Ministry of Agriculture, Ministry of Forestry, Ministry of Research and Technology and some leading universities.

The research and development can be categorized into feedstock, process and utilization. For feedstock, R&D mainly focused on how to investigate and develop feedstock available for biofuel. It is leaded by research centre from Ministry of Agriculture in which since 2004 has continued to develop Jatropha curcas. Agency of Technology Assessment and Implementation has been a pioneer in trying to develop biofuel engineering process started from size of 1000 L biodiesel per day and now they have capability to construct of 30,000 ton per year based on their own technology. Many R&D have also conducted in running test and engine test performance for biofuel conducted by government research institute, universities and companies.

In addition to large scale biofuel manufactures, initiatives to develop small scale biofuel than can be operated and utilized directly by rural people are now in the implementation across the country. The government through a program so called Energy Self-Sufficient Village constructs a small biofuel plant with capacity around

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400 litre per day. This program has been developed since 2007 and now around 20 plants have constructed. Cassava and sweet sorghum are considered as most interesting plantation since its multi function of the product. The plant is integrated with other productive economic activities, so the rural people can advantage not only from the fuel but also other economic product and services.

As a tropical country, Indonesia is rich and abundant with various oil producing plants such as coconut, palm oil, sunflower, kapok and jatropha. From a survey conducted by Bandung Institute of Technology, there are more than 50 plant species that can be developed for biodiesel feedstock. In addition to food-based feedstock, from a study carried out by APEC in 2008, as indicated by Table 2, at least 82% of gasoline consumption could be replaced by second generation bioethanol generated from cellulosic biomass.

As part of national energy policy, the utilization of biofuel will be continuously implemented and improved by combining efforts on providing incentives and diversification of feedstocks. Improving productivity of main feedstocks such as palm oil, jatropha, cassava and sugar cane are also of the concern from the government. Most research and development now are devoted to jatropha curcas and algae to bring this highly potential feedstocks to commercial implementation. In addition, development of second generation of biofuel that will create sustainable and low cost biofuel industry is also in the national pipeline. It is, therefore, necessary to seek cooperation on R&D on lignocellulosic based or woody-biomass bioethanol. Indonesia will continue to work with other countries and international community to develop sustainable biofuel industry and also global environmental issues.

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ROLE OF RICE STRAW POTENTIAL FOR BIOENERGY IN INDONESIA AS SUBSTITUTED GASOLINE AND REDUCING OF ENVIRONMENTAL PROBLEM

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ABSTRACT

Substituting fossil fuels with waste biomass-derived cellulosic ethanol is a promising strategy to simultaneously meet part of energy needs, mitigate greenhouse gas (GHG) emissions. The Indonesian abundant in varieties of plants or agricultural waste, such as rice straw, has created a greater possibility to develop bioethanol since the source of its main components can easily be found in Indonesia. Rice straw as second generation feedstock of bioethanol, does not competing with food production such as sugarcane, which is used for first generation feedstock.

Bioethanol production from rice straw is supported by the availability and development of conversion technology, thermochemical and biochemical process. A process can be economized to its maximum extent when the process steps are efficiently integrated. The both process integration is very important aspects that improve energy efficiency, reduce process costs, and enable both processes to be more sustainable.

Development of bioethanol intended to support the government policy to utilise of biofuel. Production of bioethanol is likely to open jobs in rural areas especially for the farmers that using agricultural waste (rice straw) as feedstock. There is a prospect to continue research towards the development of lignocellulose-based bioethanol.

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1 INTRODUCTION

Since the population, economic growth and the development of industrialization technology, the energy needs increase in most countries including Indonesia. The domestic demands of fuel fossil, particularly gasoline, increase around 7% of the total fossil fuel demand in year 2010. However, oil refinery production can not afford to supply the domestic needs. At the moment, the deficit of fuel demand must be fulfilled by imported fuel. Also Indonesia's crude oil reserves, in the past decade, has declined. In this case Indonesia government has a policy and provided some regulations as a part of solution in energy sector, such as Presidential Instruction No.1 year 2006 on supply and utilization of biofuels as alternative energy and Presidential Decree No.5 year 2006 on National Energy Policy, calling for 5% biofuels in the energy mix by 2025 (22.26 million kiloliters).

The utilization of biofuel particularly bioethanol, can be used as substituted gasoline without modification of vehicle engine. Bioethanol has received considerable attention in the transportation sector because of its utility as an octane booster, fuel additive, and even as neat fuel. Bioethanol can be produced from biomass such as agricultural waste (rice straw, forest residue) (Cheng *et al.*, 2007). With the oil crisis of the 1970s, ethanol became established as an alternative fuel.

Biofuel, as renewable energy, to be one of the key instruments to accelerating economic growth, alleviating poverty particularly for farmers, and creating employment opportunities. Increased use of biofuels, renewable sources of energy, involves a number of advantages. Firstly, renewable energy is all indigenous energy resources and can play a key role in reducing dependency on imported energy. Secondly, development of renewable energy is essential in order to reach our environmental goals, in particular as far as reduction of CO_2 emissions is concerned. Given that renewable sources of energy such as ethanol are CO_2 neutral, these energy sources are ideally placed to play a leading role in the CO_2 reduction strategy. Ethanol, unlike gasoline, is an oxygenated fuel that contains 35% oxygen, which reduces particulate and NO_X emissions from combustion.

2 POTENTIAL OF RICE STRAW FOR BIOETHANOL FUEL PRODUCTION

The use of alcohol (bioethanol) as an alternative transportation fuel has been steadily increasing around the world. Nowadays, alcohol fuels have been produced on industrial scales by fermentation of sugars derived from wheat, corn, sugar beets, sugar cane, cassava, etc.

In Indonesia, current ethanol production is based on either sugar (molasses) or starch crops with production volume is 52.5 million liters in 2009 (Dillon et.al., 2008). The government targeted that bioethanol will reach 6,280 million liter by 2025.

The Indonesian abundant in varieties of plants or agricultural waste, such as rice straw and forest residue, has created a greater possibility to develop biofuel (bioethanol) since the source of its main components can easily be found in Indonesia. Since bioethanol fuel production by using first generation feedstock (sugarcane, cassava)) competing with food production and pushing up food price, then bioethanol fuel production should be replaced by non-food feedstock (second generation) such as lignocellulosic biomass (rice straw, forest residue, etc). The availability of feedstock is a key factor in sustainable production of bioetanol fuel.

Rice is a staple food for the Indonesia people and is cultivated in rice fields in every region of the country. In the course of harvesting this rice, a large volume of rice straw must be reaped. The production of rice straws can reach 12 – 15 tons per hectare per harvest, varying with the location and the rice variety grown. Annual production of rice straw in Indonesia is about 67.31million ton [Departemen Pertanian/ Ministry of Agriculture, 2010; BPS, 2010]. Rice straw is so far known as side products of harvesting rice on fields. By some farmers, the straws are used as mulch when they plant secondary crops. Some farmers have used the straws as alternative feed for animals in the dry season because of the shortage of green feeds. On the other hand, as agricultural waste the straws often become a problem for farmers, so the straws are often burned for the solution. When rice straw is burnt then become problem for air pollution.

Biofuels (bioethanol) can provide a number of environmental advantages over conventional fossil fuels—most notably a reduction in greenhouse gas (GHG) emissions (Figure 1). By using of bioethanol from biomass can reduce emission until 85%. Since the transportation sector accounts for the main emissions of carbon dioxide (an abundant GHG), cleaner transportation fuels can play an important role in addressing climate change.

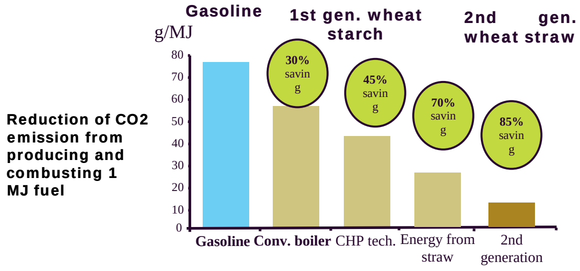

Fig 1: Comparison of GHG by using fossil fuel and bioethanol fuel. http://www1.eere.ener

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gy.gov/biomass/printable_versions/environ mental.html

The chemical composition of rice straws predomi- nantly contains cellulose 32-47%, hemicelluloses 19-27% and lignin 5-24%, ashes 18.8%. The pentoses are dominant in hemicelluloses which xylose is the most important sugar followed by arabinose and hexoses. The carbohydrate of rice straw involves glucose 41-43.4%, xylose 14.8-20.2%, arabinose 2.7-4.5%, man- nose 1.8% and galactose 0.4% (Roberto *et al*., 2003). The tipically general characteristic and constituents of rice straw is shown in Table 1.

Table 1: General characteristics and constituents of rice straw (Huang et.al, 2008)

Moisture (wt-%)	8.25
Proximate analysis[a] (wt-%):	
- Ash	13.36
- Volatiles	72.20
- Fixed carbon	14.44
Elemental analysis[b] (wt-%):	
- C	45.41
- H	6.28
- N	0.99
- S	0.21
- O[c]	47.11
Caloric value (MJ/kg)	15.26
Ash analysis[d] (wt-% of ash):	
- Na_2O	1.20
- K_2O	19.45
- CaO	2.60
- MgO	1.47
- Al_2O_3	0.36
- Fe_2O_3	0.40
- Mn_3O_4	0.26
- P_2O_5	4.74
- SiO_2[c]	69.52

[a] Dry basis. [c] Calculated by difference.
[b] Dry ash free basis. [d] Calculated from mineral elements.

According to reported research in Indonesia that rice straw can potentially produce 1 liter bioethanol with 80% content as ethanol by using 4-5 kg of rice straw (Dyono et.al.,2009). It means that from 67.31million ton of rice straw produce 13.46 million liters bioethanol per year, which is about 1% of total of consumption. It is the largest amount from a single biomass feedstock (excluding bioethanol from sugarcane and others). Meanwhile, production of 1 liter of ethanol by using 2.5 kg rice straw in Japan (www.companiesandmarkets.com /Market-Report/analyzying-biofuel-fro..). The diff- erentiation of this mate- rial amount to produce of ethanol content is due to the different of rice varieties and its rice straw compositions.

3 CONVERSION TECHNOLOGY FOR ETHANOL PRODUCTION

Lignocellulosic biomass is typically non-edible plant material composed primarily of the polysaccha- rides cellulose and hemicellulose. The third major component is lignin, a phenolic polymer that provides structural strength to the plant. Technology for producing biofuels (such as ethanol, butanol, or various hydrocarbons) and biobased chemicals from lignocellulosic material is experiencing significant advances in an effort to meet global energy and chemical needs (Himmel *et.al*, 2007; Ragauskas *et.al*. 2006). Examples of lignocellulosic biomass materials considered as feedstocks for bioethanol production include crop residues such as corn stover and wheat straw, woody residues from forest thinning and paper production.

3.1 Conversion Technologies

A number of different conversion technologies exist for the conversion of cellulosic biomass to biofuels (Farrell *et al*. 2006; Rammamorth *et al*. 2000; Huber *et al*. 2006). The selection of biomass conversion technology is govern by following factors: (a) The feedstock available, (b) The end application, and (c) The cost

In general, the conversion technology is grouped in three technology platforms as option that available for energy production from biomass (bioenergy pathways) such as rice straw as shown in Figure 2.

Generally, thermochemical and biochemical process are used to convert biomass for ethanol production. Ethanol can be made synthetically from petroleum or by microbial conversion of biomass materials through fermentation. In 1995, about 93% of the ethanol in the world was produced by the fermentation method and about 7% by the synthetic method. The fermentation method generally uses three steps: (1) the formation of a solution of fermentable sugars, (2) the fermentation of these sugars to ethanol, and (3) the separation and purification of the ethanol, usually by distillation.

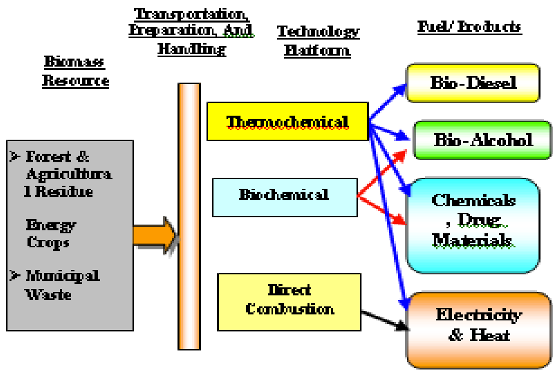

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Fig 2: A simplified illustration of the technology options available for energy production from biomass

Thermochemical conversion process that utilize pyrolysis/steam reforming processes (no oxygen or air) are currently capable of economically producing bioalcohols. Biochemical conversion processes have been available for nearly 100 years that utilize acid hydrolysis for the conversion of cellulose to sugars, followed by the fermentation of the sugars to bioethanol.

The predominant differentiation between the conversion options is the primary catalysis system (Foust *et al*. 2008). Biochemical conversion routes rely on biocatalysts, such as enzymes and microbial cells, in addition to heat and chemicals to convert biomass first to an intermediate mixed sugar stream and then to ethanol or other fermentation produced biofuel. However the lignin component is hard to break down into fermentable component, it remains as residual after fermentation. In contrast, thermochem- ical conversion technologies rely on heat and/or physical catalysts to convert biomass to an interme- diate gas or liquid, followed by a conversion step to convert that intermediate to a biofuel. Lignin can be converted for fuel production by thermochemical processing as syngas in a gasifier. Thermochemical conversion technologies tend to be grouped in two distinct categories for fuel production: gasification and pyrolysis (McKendry, 2002).

Gasification is a complete depolymerization of biomass with limited oxygen at high temperatures, typically >850 °C, to a gaseous intermediate (syngas), which consists of H_2 and CO. A good review of the different types of gasification techniques that exist and their relative advantages and disadvantages is provided by Spath and Dayton (Spath, 2003).

Pyrolysis on the other hand, is the milder depolymerization of biomass producing a liquid intermediate (pyrolysis oil or "bio-oil") in the absence of oxygen at lower temperatures, typically in the range of 400–650 °C. Good reviews of pyrolysis techniques and the current technical status of these techniques are provided by Bridgwater and Peacocke (Bridgwater, 2000) and Czernik and Bridgwater (Czernik, 2004).

3.2 Pretreatment

Pretreatment refers to a process that converts lignocellulosic biomass from its native form, in which it is recalcitrant to cellulase enzyme systems, into a form for which cellulose hydrolysis is much more effective. In general, pretreatment methods can be classified into three categories, including physical, chemical, and biological pretreatment.

The overall purpose of pretreatment is to break down the shield formed by lignin and hemicellulose, disrupt the crystalline structure and reduce the degree of polymerization of cellulose.

Pretreatment techniques have been developed for various end uses of biomass feedstocks. With the advancement of pretreatment technologies, the pretreatment is also believed to have great potential for the improvement of efficiency and reduction of cost (Mosier *et.al,* 2003). Pretreatment methods can be roughly divided into different categories: physical (milling and grinding), physicochemical (steam pretreatment/autohydrolysis, hydrothermolysis, and wet oxidation), chemical (alkali, dilute acid, oxidizing agents, and organic solvents), biological, electrical, or a combination of these (Zheng *et.al.*2009; Kumar *et.al.*, 2009).

The digestibility of cellulose present in lignocell- ulosic biomass is hindered by many physicochemical, structural, and compositional factors. In the conver- sion of lignocellulosic biomass to fuel, the biomass needs to be treated so that the cellulose in the plant fibers is exposed. Pretreatment uses various techniques, including ammonia fiber explosion, chem- ical treatment, biological treatment, and steam explosion, to alter the structure of cellulosic biomass to make cellulose more accessible (Hsu *et.al.* 1980) Therefore, acids or enzymes can be used to break down the cellulose into its constituent sugars. Enzyme hydrolysis is widely used to break down cellulose into its constituent sugars. The goal of pretreatment in biomass-to-biofuels conversion is depicted in Figure 3.

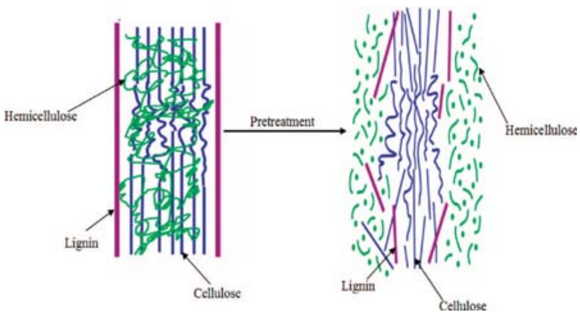

Fig 3: Schematic of the role of pretreatment in the conversion of biomass to fuel. (Hsu *et.al.*, 1980.)

Pretreatment is an important tool for biomass-to-biofuels conversion processes. The effect of pretreat- ment, can be exemplified by work of Kristensen et.al. as seen in Table 2. The main effect of the hydrothermal pretreatment on the composition of the biomass is the partial but substantial removal of hemicelluloses. All measurable arabinan is removed and the xylan content is reduced from 24.5% to 5.2%. Consequently, the overall cellulose content increases. After delignification of the pretreated material, no Klason lignin can be detected. The composition of the straw that has undergone SO_2-impregnated steam explosion is similar to that of

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the hydrothermally pretreated straw except for a slightly higher xylan content at 7.8%.

The rate and extent of enzymatic hydrolysis is affected by the pretreatment method, substrate concentration and accessibility, enzyme activity (loading), and reaction conditions such as pH, temperature, and mixing (Merino and Cherry, 2007; Taherzadeh and Karimi, 2007).

Table 2: Compositions of straw after hydrothermal pretreatment

	Cellulose	Xylan	Arabinan	Klason lignin	Ash
Straw, untreated	39.8	24.5	2.8	22.6	4.2
Pretreated straw	59.0	5.2	0.0	25.5	5.6
Delignified, pretreated straw	75.1	9.8	0.0	0.0	8.8
Steam-exploded straw	56.7	7.8	0.7	23.6	6.3

Contents expressed as percentages, based on dry matter. (Kristensen *et al. Biotechnology for Biofuels* 2008 **1**:5 doi:10.1186/1754-6834-1-5)

4 PROCESS INTEGRATION/INDUSTRIAL COMPLEXING

A process can be economized to its maximum extent when the process steps are efficiently integrated (Banerjee, 2010; Foust *et.al.*, 2009). There are two key design elements that have been applied to both the biochemical and thermochemical processes integra- tion: (1) energy self-sufficiency and integration, and (2) significant process recycle. These are very important aspects that improve energy efficiency, reduce process costs, and enable both processes to be more sustainable.

In the biochemical process, energy self-sufficiency comes from combusting lignin-rich residue to gener- ate steam and electricity for the biorefinery. Sufficient residue exists to fully meet the heating and electric demands of the plant. In the thermochemical process, a slipstream of unreformed syngas is combusted (along with the char from the gasifier) to provide the entire energy needs of the biorefinery. While this slightly lowers the overall ethanol yields, it does eliminate the need to purchase fossil-derived energy sources. Pinch analysis was used to develop an integrated system of heat exchangers that generate process steam and increase the overall energy efficiency of the process.

Forward-backward integration has been found to be very effective in cost reduction. To start with, the lignocellulose ethanol plant can be integrated with an operational ethanol plant; for instance, a sugarcane bagasse bioconversion facility can be integrated with an operational sugarcane (or cane molasses) ethanol plant. This saves on transportation costs considerably.
The CO_2 released during the fermentation process has commercial importance and can be harnessed. CO_2 generated during ethanolic fermentation can also be catalytically hydrogenated to methanol (Grassi, 1999).

The importance of integrating an ethanol production plant with a power generation plant to channel the high-calorific value lignin byproduct for energy generation has been well documented (Hahn *et.al.*, 2006; Laser *et.al.*, 2009). Anaerobic digestion of the waste water generated after distillation produces methane-rich biogas that can be captured and used to generate electricity. Lignin-rich residues can be gasified in a pressurized oxygen-blown gasifier to produce synthesis gas from which hydrogen can be separated via pressure swing adsorption (Laser *et.al.*, 2009). The energy thus obtained can fuel the power requirement of the plant, making it energy indepen- dent. A schematic representation of process integra- tion opportunities for lignocellulosic ethanol produc- tion is provided in Fig. 3.

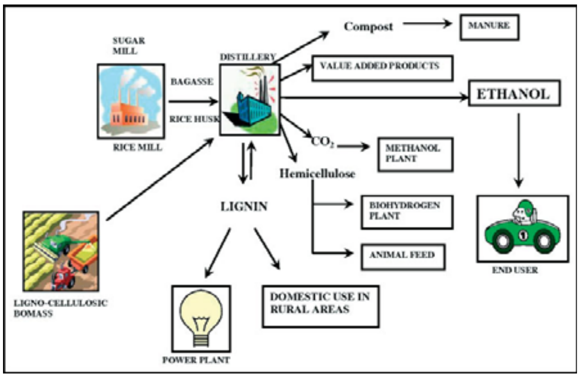

Fig 4: Process integration/industrial complexing opportunities for ethanol production from lignocellulosic biomass.(Banerjee, 2010)

5 DEVELOPMENT OF BIOFUEL IN INDONESIA

Indonesia has a good potential of lignocell-ulosic biomass residues available to energy production particularly agricultural waste (rice straw). Currently, ethanol production still using sugarcane, cassava, molasses as feedstock that

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competing with food production. Biomass may cover about 5% of total energy demand in Indonesia as government targeted in 2025. The technology of biomass conversion is now available to be utilized for bioethanol production.

Various ways are under development to convert biomass into bioethanol fuel:

- Biochemical conversion (Anaerobic digestion, Aerobic conversion, Fermentation)
- Thermochemical conversion (Gasification, Pyrolysis, Fischer-Tropsh)

The use of biomass (lignocellulosic) to provide energy has been fundamental to the development of civilisation. In recent times pressures on the global environment have led to calls for an increased use of renewable energy sources, in lieu of fossil fuels. Development of bioethanol intended to support the government policy to utilise of biofuel. Production of bioethanol is likely to open jobs in rural areas especially for the farmers that using agricultural waste (rice straw) as feedstock.

By looking at these benefits, there is a prospect to continue research towards the development of lignocellulose-based bioethanol. Research on the utilization of cellulosic material as raw material for production of ethanol has been started since 1950, but with the advancement technology then development of bioethanol becomes promising.

6 CONCLUSION

Based on the advancement of conversion techno- logy and the availability of rice straw in Indonesia, therefore bioethanol production from rice straw has a potential as substituted for gasoline fuel.

The selection of biomass conversion technology is govern by following factors.

- The feedstock available
- The end application
- The cost

Bioethanol was produced from various agricultural raw material using two conversion technologtes i.e. biochemical and thermochemical conversion.

Lignocellulosic raw materials (rice straw) will reduce the likelihood of conflict between land used for food production (and feed) and the land for the production of raw materials for energy supply. Price of this type of raw material cheaper than the first generation and not competing with food production

Since the bioethanol lignocellulose-based green- house gas (GHG) emissions are lower, reducing the environmental impact, especially climate change.

Bioethanol production from rice straw is likely to open jobs in rural areas particularly for farmers.

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MICROBIAL HYDROGEN PRODUCTION: STATE OF THE ART AND CHALLENGES FOR ITS INDUSTRIAL EXPLOITATION

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ABSTRACT

The biological processes leading to the production of hydrogen can be classified as follows: (i) biophotolysis of water, carried out by microalgae and cyanobacteria; (ii) fermentative hydrogen production (dark fermentation), carried out by chemoheterotrophic microorganisms; (iii) photofermentative hydrogen production (photofermentation), carried out by photoheterotrophic bacteria; (iv) integrated hydrogen producing systems, which combines dark and photofermentation. In this paper the most important issues of biological hydrogen production are reviewed, in particular focusing on the advantages and the drawbacks of the various processes under study. The main challenges for the development of economically and environmentally sustainable processes are also discussed.

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1 INTRODUCTION

One of the major challenges for the mankind in the near future is related with the resolution of the interconnected problems of the availability of clean and renewable Energy resources and the reduction in the emission of GHG (Greenhouse gases) into the atmosphere. In spite of a general agreement in considering the use of fossil fuels as the cause of serious environmental problems and in foreseeing a progressive decrease of their reserves, the amount of energy derived from these energy sources is very high, still reaching in 2008 about the 80% of the World energy consumption (Key World Energy Statistics, IEA, 2010). For these reasons, there is a pressing need of new, not polluting and renewable energy sources. In this connection, hydrogen, even if it is not an energy source, has been considered a promising alternative to fossil fuels. The two most interesting characteristics of H_2, which make its use in the production of energy quite promising, are its large presence in nature, even if usually linked with other atoms (e.g. with oxygen in water or with oxygen, carbon and other elements in organic compounds), and the possibility to use it without releasing pollutants or GHG in the atmosphere. However, as stated above, H_2 is not an energy source, i.e. an already existing source from which useful energy can be extracted or recovered, but an energy carrier. The main difference between an energy source and an energy carrier is that the former is already available in nature for being used in the production of energy while the latter has to be produced by using an energy source and can be used, like electricity, for the "transport" of energy from the production site to the sites of its use. The main consequence of this characteristic is that hydrogen can be produced only by consuming primary energy sources, which at the moment are mainly constituted by hydrocarbons (Lee *et al*., 2010).

However, hydrogen can also be produced by exploiting the metabolic features of several microorganisms in a carbon neutral process that has been called "biological hydrogen production".

Biological hydrogen production, which can be defined as the microbiological conversion of water, sunlight and/or organic substrates into H_2, is characterized by important advantages over the thermochemical and electrochemical techniques at present utilized or under study. Indeed, in the microbiological processes, hydrogen can be produced utilizing renewable resources, in carbon neutral processes operating at room temperature and pressure and characterized by a low environmental impact. A large number of microbial species, belonging to different taxonomic types, possesses the capability to produce hydrogen. However, free H_2 of biological origin is hardly found in nature because H_2-producing and H_2-consuming microorganisms live in the same natural environments.

2 ENZYMES INVOLVED IN BIOLOGICAL HYDROGEN PRODUCTION

The enzymes involved in biological hydrogen production are nitrogenases, in most cases a [FeMo] nitrogenase, and hydrogenases. Both groups of enzymes are sensitive to the negative effects of the presence of oxygen, even if at a different level of sensitivity.

2.1 Nitrogenase

Nitrogenase is a two-protein complex consisting of a dinitrogenase, which contains Fe and Mo as cofactors and has a molecular weight (MW) of 250 kDa, and of a dinitrogenase reductase, which contains Fe and has a MW of about 70 kDa. Alternative nitrogenases, a Vanadium and a Fe-only nitrogenase, have also been described, even if less frequently found (Larimer *et al*., 2004). In presence of N_2, the reactions of the three isozymes produce different ratios of hydrogen and ammonia (McKinlay and Harwood 2010), as shown in the following reactions:

[Mo-nitrogenase]

$N_2 + 8H^+ + 8e^- + 16ATP \rightarrow 2NH_3 + H_2 + 16ADP$

[V-nitrogenase]

$N_2 + 12H^+ + 12e^- + 24ATP \rightarrow 2NH_3 + 3H_2 + 24ADP$

[Fe-nitrogenase]

$N_2 + 24H^+ + 24e^- + 48ATP \rightarrow 2NH_3 + 9H_2 + 48ADP$

In the absence of N_2, nitrogenases only catalyze the reduction of protons with the consequent formation of H_2.

Nitrogenases catalyze a very expensive reaction in terms of energy, and thus they are very strictly regulated by the presence of dissolved ammonium ions (MacKinley and Harwood, 2010).

2.2 Hydrogenase

The hydrogenases are iron-sulfur proteins distributed into two main phylogenetically distinct classes, the [NiFe]-hydrogenases and the [FeFe]-hydrogenases, which respectively contain a Ni and a Fe atom or two Fe atoms at their active site.

All the hydrogenases are negatively affected by the presence of oxygen. [FeFe]-hydrogenases are the most sensitive and are denaturated in the presence of traces of O_2; [NiFe]-hydrogenases are less sensitive and the inactivation due to the

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presence of O_2 is reversible. Thus, even if they are less productive than [FeFe]-hydrogenases, [NiFe]-hydrogenases are considered most promising for the industrial exploitation.

The [NiFe]-hydrogenases, which are the most studied, have been divided into four groups, according to the function they have (Vignais 2008): (i) *uptake hydrogenases*: respiratory enzymes, involved in anaerobic respiration, which recover the electrons from H_2; (ii) *cytoplasmic H_2 sensors*: regulatory enzymes, activating the cascade that regulates the respiratory hydrogenases in presence of H_2; (iii) *bidirectional [NiFe]-hydrogenases*: enzymes able to bind NAD and NADP and to work in both directions, consuming or producing H_2; (iv) *H_2 evolving membrane-associated hydrogenases*: enzymes that couple anaerobic oxidation of one-carbon-atom organic compounds with the production of H_2.

In H_2 production processes, active *uptake-hydrogenases* are undesirable, as they reduce the productivity of H_2 producing bacteria. Indeed, it has been found that an inactivation of such enzymes usually leads to an enhanced hydrogen production (Ooshima *et al*. 1998; Franchi *et al*. 2004; Kim, Baek and Lee 2006; Öztürk *et al*. 2006; Kars *et al* 2008).

3 BIOLOGICAL HYDROGEN PRODUCTION PROCESSES

The biological processes leading to the production of hydrogen can be classified as follows: (i) biophotolysis of water, carried out by microalgae and cyanobacteria; (ii) fermentative hydrogen production (dark fermentation), carried out by chemoheterotrophic microorganisms; (iii) photofer- mentative hydrogen production (photofermentation), carried out by photoheterotrophic bacteria; (iv) integrated hydrogen producing systems, which combine dark and photofermentation.

3.1 Biophotolysis of water

The possibility of using oxygenic phototrophic microorganisms (microalgae and cyanobacteria) for the simultaneous production of hydrogen and oxygen from the light-driven photosynthetic water-splitting process is considered a very attractive option for biological hydrogen production. Indeed, this process utilizes largely available energy and hydrogen sources (sunlight and water, respectively) for obtaining H_2 in a clean and carbon neutral process. A simplified scheme of this process is reported in Figure 1: the photons deriving from sunlight cause the water-splitting reaction at photosystem II (PSII) of microalgal or cyanobacterial cells and the electrons deriving from this process are channeled, through Ferredoxin, to hydrogenase (a FeFe-H_2ase in microalgae or a NiFe-H_2ase in some cyanobacteria) or to nitrogenase (in most cyanobacteria), both enzymes capable of catalyzing the H_2-forming reaction:

$$2H^+ + 2e^- = H_2$$

The hydrogenases are reversible enzymes, i.e. they catalyze the reaction in both directions, depending on the environmental conditions, while nitrogenase is unidirectional, but requires ATP for being able to transfer the electrons to H^+.

In spite of the fascination of this clean and elegant way to obtain hydrogen from water and sun, there are three main constraints on the possibility to industrially exploit oxygenic phototrophic organisms for producing H_2 with this process: (i) both H_2ase and N_2ase are sensitive to O_2, which is produced in large amounts during the photosynthesis; (ii) potentially explosive hydrogen/oxygen mixtures are produced in the gas phase; (iii) this process shows low H_2 production rates in comparison with the other biological methods (Rupprecht *et al*., 2006).

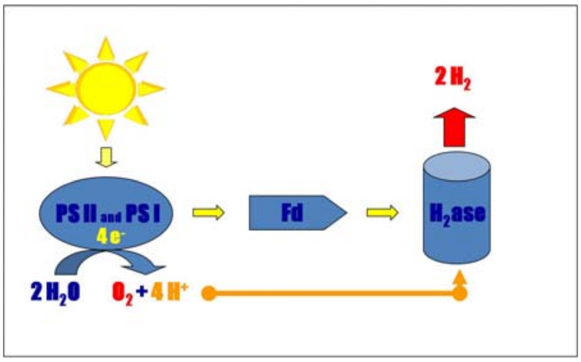

Figure 1: Scheme of H_2 production by a microalga by means of the biophotolysis of water. PSII = photosystem II; PSI = photosystem I; Fd = ferredoxin

3.2 Fermentative hydrogen production (dark fermentation)

Many chemoheterotrophic bacteria are capable to anaerobically produce hydrogen degrading organic matter in fermentative processes also called dark fermentation for distinguishing them from photofermentation, described in the following paragraph 3.3. The production of hydrogen in dark fermentation is due to the need of the cells to get rid of the excess of reducing power produced in the oxidation of organic substrates. The electrons are channelled to hydrogenase, the enzyme present in anaerobic dark-fermenting bacteria, which catalyze the formation of gaseous hydrogen. If the organic matter fermented is biomass (agricultural residues, wastes deriving from vegetable food products etc.) the biohydrogen obtained is considered renewable

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since the biomass was originally produced from photosynthesis.

However, fermentative bacteria partially use also other electron sinks for the excess reducing power, with the consequent release of soluble reduced compounds, such as acetate, butyrate, ethanol etc., depending on the fermentative pathway they use.

The fermentative pathways leading to the production of hydrogen always start with the degradation of carbohydrate-reach substrates. The carbohydrates are degraded, via the glycolytic or the pentose phosphate pathway, to pyruvate, that can be further oxidized following two different pathways catalyzed by pyruvate:ferredoxin oxidoreductase (PFOR) or by pyruvate:formate lyase (PFL).

In the first pathway, typical of *Clostridia*, pyruvate is degraded to acetyl-Coa, CO_2 and reduced Ferredoxin, in a reaction catalyzed by PFOR. Then, the reduced Ferredoxin donates the electrons to a [FeFe] hydrogenase with the consequent formation of hydrogen. In addition to this production, at low values of partial hydrogen pressure, the NADH produced during glycolysis is reoxidized, probably with the participation of a [FeFe] hydrogenase, producing additional amounts of hydrogen. However, when hydrogen partial pressures are moderate or high, the above described reaction becomes unfavourable and the reoxidation of NADH is obtained utilizing acetyl-CoA as substrate and is produced butyrate instead of hydrogen (Hallenbeck, 2009).

In the mixed-acid fermentation, typical of enteric bacteria, the pyruvate derived from the degradation of carbohydrates is cleaved to acetyl-CoA and formate, which is converted to H_2 and CO_2 by PFL under low pH conditions. Under these conditions, it is also active lactate dehydrogenase (LDH), which catalyzes the reduction of pyruvate to lactate, thus diverting NADH and one molecule of pyruvate from the formation of hydrogen. The acetyl-Coa produced together with formate can be converted to acetate or to ethanol (Hallenbeck, 2009).

In dark fermentations it was demonstrated that, owing to metabolic and thermodynamic limitations, only a maximum of 4 mol of H_2 per mol of glucose can be obtained. This limit, called *Thauer limit*, is due to the need for the cells to obtain at least 1 mol of ATP per mol of glucose consumed via substrate level phosphorilation (Thauer 1977).

3.3 Phofermentative hydrogen production

In photofermentation, anoxygenic photosynthetic bacteria, in particular purple non-sulfur (PNS) bacteria, reduce H^+ ions to gaseous H_2 using the reducing power deriving from the oxidation of organic compounds (e.g. low-molecular weight fatty acids) and the energy deriving from sunlight. This process is generally considered very promising, due to the high substrate conversion yields that can be achieved, the possibility to use a wide spectrum of wavelengths of sunlight, the absence of O_2 evolving reactions (that would inhibit the H_2-producing enzymes) and the possibility to couple this kind of H_2 production processes with waste disposal (Basak and Das 2007).

In the photofermentation process (Fig. 2), PNS bacteria oxidize organic compounds and the electrons obtained in this way are channeled, through reverse electron flow, to Ferredoxin and to N_2ase, the enzyme that catalyze hydrogen formation in PNS bacteria. This process requires large amounts of ATP for both the reverse electron flow and the activity of N_2-ase.

The maximum amount of hydrogen obtainable from the most frequently utilized organic acids can be calculated from the stoichiometric reactions of their complete oxidation:

Lactate: $C_3H_6O_3 + 3H_2O \rightarrow 6H_2 + 3CO_2$

Acetate: $C_2H_4O_2 + 2H_2O \rightarrow 4H_2 + 2CO_2$

Malate: $C_4H_6O_5 + 3H_2O \rightarrow 6H_2 + 4CO_2$

However, it has to be stressed that part of the organic substrate is utilized as carbon source for cell growth and also that some organic acids, such as acetate and butyrate, can be used by the cells for the synthesis of poly-beta hydroxybutyrate (PHB) instead of for H_2 production (Vincenzini *et al.*, 1997). Thus, most frequently the conversion of the substrate to hydrogen remains below the theoretical value. With the only exception of a 100% conversion efficiency reported by Sasikala *et al.* (1990) for a 2 ml culture, the conversion efficiencies mainly range for acetate between 69 and 75%, for lactate between 50 and 85% and for malate from 25 to 88% (Adessi and De Philippis, 2011).

The conversion efficiency is strongly affected by the C/N ratio in the culture: a high C/N ratio in the culture media usually leads to a high hydrogen production while a low C/N ratio leads to a high cell growth (Redwood *et al.*, 2009).

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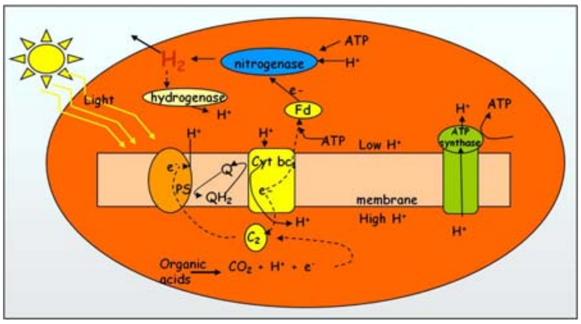

Figure 2: Photofermentation in a purple non sulfur bacterium. PS = photosystem; Fd = ferredoxin; Cyt bc_1 = Cytochrome bc_1; c_2 = Cytochrome c_2.

3.4 Integrated hydrogen producing systems

Dark fermentation, as above reported, can lead to the maximal production of 4 mol H_2 per mol of glucose consumed, with the contemporaneous release in the culture medium of a number of partially oxidized organic compounds, mostly low-MW fatty acids, that still could be further utilized as electron donors. However, their oxidation for obtaining reducing power is thermodynamically unfavorable, owing to the very positive values of ΔG0' of these reactions. For instance, in the case of the oxidation of acetate to CO_2 and H_2 the value of $\Delta G^{0'}$ is +104.6 kJ mol^{-1}.

As a consequence, this process is unfeasible for chemoheterotrophic bacteria but becomes possible through the photofermentation of acetate or of the other compounds carried out by PNS bacteria, as the energy required for this endoergonic reaction can be obtained from sunlight.

Moving from these considerations, in recent years many research groups investigated the possibility to use a two stage process where, in the first stage, the dark fermentation of organic substrates is carried out by chemoheterotrophic bacteria, which produce H_2 and of low-MW organic acids, while in the second stage the acids previously produced are utilized by PNS bacteria as electron donors for the nitrogenase-mediated hydrogen photoevolution (Fig. 3).

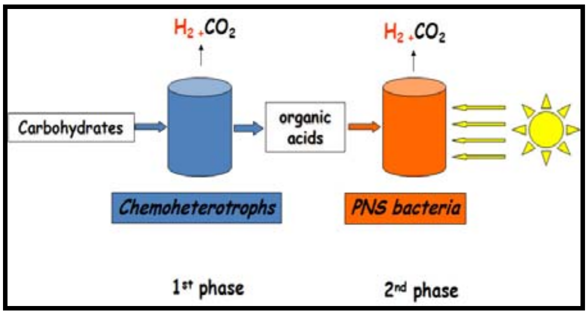

Figure 3: Scheme of the two-phase integrated processes for biohydrogen production.

The studies carried out with the integrated systems have been recently exhaustively summarized by Redwood *et al.* (2009) and by Adessi and De Philippis (2011), pointing out a number of interesting results obtained in the last years. For instance, very promising results have been obtained by Kim *et al.* (2006), with an overall productivity of 8.3 mol H_2 per mol of glucose using algal biomass as starting material. Also the use of sweet potato starch residues (Yokoi *et al.* 2001, 2002) has lead to a very interesting productivity (7.2 mol H_2 mol glucose). Beet molasses have been used as well (Özgür *et al.* 2010) obtaining a productivity of 6.85 mol H_2/mol glucose.

4 LIMITATIONS AND CHALLENGES

The main advantages and drawbacks of the above described biological hydrogen producing processes are summarized in Table 1.

In particular, due to the low hydrogen production rates and to the problems created by the oxygen produced by photosynthesis to the enzymes that catalyze H_2 formation, it seems not much realistic to propose biophotolysis of water for the industrial production of hydrogen.

Process	Microorganisms	Advantages	Drawbacks
Biophotolysis of water	Green microalgae; cyanobacteria	-Production from H_2O and sunlight	- H_2 producing enzymes sensible to O_2 -Low production rates
Dark Fermentation	Fermentative bacteria (various genera)	- Anaerobic process - It can utilize poor, renewable carbohydrate containing substrates - Not affected by dark/light cycles - High production rates	- Incomplete utilization of substrates - Negatively affected by the increase in H_2 partial pressure -Produces CO_2, which must be separated from H_2
Photofermentation	Photosynthetic bacteria (PNS bacteria)	- Use compounds contained in industrial wastewaters or deriving from dark fermentation - Uses a broad range of wavelengths of sunlight	- Nitrogenase consumes large amount of ATP - Nitrogenase is sensitive to NH_4^+ - Low light conversion efficiency

Table1: Main advantages and drawbacks of the biological hydrogen-producing processes.

On the other hand, dark fermentation shows high H_2 production rates and the capability to utilize complex organic substrates (cellulose, food wastes, municipal wastes etc), but the yield in hydrogen is low, around 17%, and the incomplete oxidation of the carbohydrates leaves in the fermentation broth a number of partially oxidized compounds.

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Photofermentation seems to be rather promising, due to its capability to utilize a large number of organic compounds released by dark fermentation or present in industrial wastes and to its capability of taking advantage of the energy of sunlight with a photosynthetic process that does not release O_2. On the other hand, photofermentation shows low light conversion efficiency and nitrogenase, the enzyme catalyzing H_2 formation in PNS bacteria, is sensible to ammonia, a compound frequently present at high concentrations in the broths deriving from dark fermentation.

In this respect, the use of integrated systems in two stages is realistically the most suitable process for future biological energy production.

As above reported, the most frequently utilized microorganisms in the dark fermentative hydrogen production processes are mesophilic bacteria, belonging to *Clostridia* or to *Enterobacteria*, and some thermophilic bacteria such as *Caldicellu-losiruptor* and *Thermotoga*. The advantages in using thermophilic bacteria are related with their faster kinetics of H_2 production and the ease in controlling the contamination with other microorganisms that could balance the energetic costs. However, it has to be stressed that the use of hot residual waters deriving from industrial processes would lead to overcome the problem of the costs of a process using thermophilic bacteria.

For what concerns the photofermentative stage, PNS bacteria are the most suitable organisms due to the high substrate conversion yields that can be achieved, the possibility to use a wide spectrum of the sunlight, and the wide variety o organic compounds that they can metabolize for hydrogen production.

Though the integration of these two processes seems to be the elected solution for biological production processes, several problems have still to be overcome.

In addition to the limitations of the two processes taken singularly, the integration brings to light some additional problems: first of all the cultivation modes have an influence on the integration strategy, as the first stage biomass needs to be separated from the effluent before starting the second stage. Further- more, the effluent has to be sterilized (if axenic single-microorganism processes are used) and properly treated (i.e. integrated with some limiting nutrients, or refined in nitrogen composition), leading to an increase in the costs. In the case of the photofermentative stage, the most challenging problem is related with the possibility to have a good distribution of the light in the photobioreactor, a parameter that drastically affects the light conversion efficiency (Dasgupta *et al*., 2010; Gadhamshetty *et al*., 2011).

5 CONCLUSIONS

In spite of the excellent research work done in the last decades on biological hydrogen production, this process never reached the level of large scale plants owing to a number of constraints that still limit its industrial exploitation. However, many progresses have been achieved owing to the increased know- ledge on the physiology and molecular biology of the microorganisms involved in biological hydrogen production. In the next future new efforts have to be done in developing the integrated systems, both improving the efficiencies of each of the two stages and reducing the negative impact of some of the compounds contained in the fermentation broth deriving from the first stage. Moreover, one critical point is related with the need of increasing the light conversion efficiency of the second stage, an improvement that could be achieved genetically manipulating the PNS bacteria used or optimizing the light distribution in photobioreactors. Moreover, low cost, highly transparent photobioreactors has to be developed for the second stage. Finally, in order to give the possibility to use a wide range of renewable biomass as substrate, it should also be developed the use of specific microorganisms or hydrolytic enzymes for releasing carbohydrates from complex matrices such as ligno-cellulosic residues in order to increase the economical and environmental sustain- ability and profitability of biological hydrogen production.

For concluding this short review, it is worth mentioning a recent paper where Hallenbeck and Ghosh (2009) stated that dark fermentative hydrogen production rates obtained utilizing real wastes reached practical levels, comparing the results obtained for H_2 production with the production of bioethanol from lignocellulosic substrates.

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DISINTEGRATION OF LIGNOCELLULOSIC BIOMASS FOR 2ND GENERATION BIOFUELS & BIOREFINERIES

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ABSTRACT

Excessive usage of fossil resources causes global warming and depletes readily accessible crude oil. The conversion of lignocellulosic biomass into bioethanol and biochemicals is becoming vital to the solution of these problems. Lignocellulosic feedstock, however, have a complex composite structure and their efficient utilization requires separation of their main polymeric components, cellulose, hemicelluloses, and lignin. Therefore, it is necessary to liberate cellulose and hemicelluloses from the lignin complex for the effective production of fuels and chemicals from lignocellulosic biomass. In lignocellulosic biorefinery, conversion of lignin to valuable aromatic chemicals is also important issue. In this seminar our recent approaches to convert lignocellulosics to bioethanol, biomethane and value-added chemical products are introduced, in highlight of biological and chemical lignin-degrading technologies and characterization of pretreated biomass and its components.

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1 INTRODUCTION

Biorefinery is a production system for fuels, chemicals, materials and energy from biomass in integrated chemical and energy industries, and it provides a new concept to change the petrochemical industry developed in 20th century. There is a growing demand to establish biorefinery to solve the problems of global warming and deficiency of fossil fuels. Because biomass is our only "carbon-based" renewable resources, biorefinery plays a key role to replace oil-based chemical industry. Among the potential industrial biorefineries, lignocellulosic biorefinery has immense potentials to replace oil refinery, due to large quantities of lignocellulosics, non-competitiveness in food supply and availability as plant wastes. Fractionation of cell wall components, cellulose, hemicelluloses, and lignin is a key technology to establish the lignocellulosic biorefinery.

In enzymatic conversion of lignocellulosics it is necessary to decompose the network of lignin prior to the enzymatic hydrolysis because lignin makes the access of cellulolytic enzymes to cellulose difficult. Production of value added products from lignin is also important issue due to its abundance and demand for the supply of aromatic chemicals from renewable resources. Thus, effective lignin-degrading pretreatments are needed for the lignocellulosic biorefinery.

Our group has been focusing on ligninolytic systems of selective white rot fungi and microwave-assisted degradation of lignin, aiming at producing biofuels and chemicals from a wide range of biomass including recalcitrant softwood. In enzymatic saccharification of lignocellulosics, the access of the enzymes to exposed cellulose surfaces is a key initial step in triggering hydrolysis. However, knowledge of the structure-hydrolyzability relationship of the pretreated biomass is still limited. We used fluorescent-labeled recombinant carbohydrate-binding modules from *Clostridium josui* as specific markers for exposed crystalline cellulose (*Cj*CBM3) and non-crystalline cellulose (*Cj*CBM28) to analyze the complex surfaces of pretreated lignocellulosics. For the effective use of residual lignin, chemical structures of the isolated lignin is studied using advanced NMR with a cryoprobe and ultra-high resolution mass spectrometry.

2 APPLICATIONS OF LIGNIN-DEGRADING FUNGI FOR BIOREFINERY

Development of conversion systems from lingocellulosics into biofuels and chemicals has received much attention due to immense potentials for the utilization of renewable bioresources. In particular, keen attention has been paid to the fermentation of lignocellulosic carbohydrates obtained by saccharification with cellulolytic enzymes. Since lignin makes the access of cellulolytic enzymes to cellulose difficult, it is necessary to decompose the network of lignin prior to the enzymatic hydrolysis. Biological pretreatment with white rot fungi in combination with thermochemical or physicochemical treatment is one possible approach for this purpose.

We applied pretreatments with selective white rot fungi for enzymatic saccharification and fermentation of lignocellulosics. Selective white rot fungi degrade lignin with minimum loss of cellulose. A white rot fungus SKM2102 isolated in Japan was identified as a new strain belonging to the genus, *Phellinus* sp. by nucleic acid sequence of ITS region. Cultivation conditions for the pretreatment of Japanese cedar wood with *Phellinus* sp. SKM2102 were optimized using a solid-state fermentation reactor and various media. A solid state fermentation was conducted by suppressing growth of competing microorganisms under the partially sterilized conditions. Pretreatments with *Phellinus* sp. SKM2102 and microwave irradiation increased enzymatic saccharification of Japanese cedar wood.

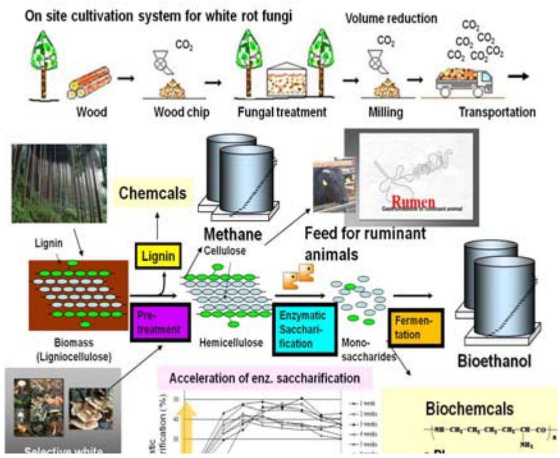

Fig 1: Applications of white rot fungi for biomass conversion

The pretreatments with white rot fungi were applied to methane fermentation of wood biomass. The fungal pretreatments with *Ceriporiopsis subvermispora* and *Phellinus* sp. SKM2102 increased yields of biomethane. The use of white rot fungi and biomimetic radical reactions is attractive not only for increasing saccharification yields but also for decreasing input energy for milling and transportation of biomass.

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3 BIOETHANOL PRODUCTION BY MIROWAVE SOLVOLYSIS AND GNENE ENGINEERED BACTERIA

There is growing demand to develop new, highly efficient technology to produce bioethanol from woody biomass. For enzymatic conversion of lignocellulosics, pretreatments to expose cell wall polysaccharides is necessary. Efficient conversion of the enzymatic hydrolyzates by ethanologenic microbes are also the key factor for bioethanol production. In our NEDO project (R&D member: RISH, Kyoto Univ., Prof. Hideshi Yanase, Fac. Eng. Tottori Univ., Japan Chemical Engineering & Machinery Co. Ltd., and Toyota Motor Corporation), we have applied ligninolytic systems of selective white rot fungi and microwave-assisted solvolysis to the pretreatment. We have developed new continuous microwave reactors by applying 3D electromagnetic simulation technique. Through cell-surface engineering based on genome DNA analysis information, novel, high-performance ethanologenic bacteria, *Zymobacter palmae* and *Zymomonas mobilis*, which efficiently convert hexoses and pentoses to bioethanol, secrete β-glucosidase, and

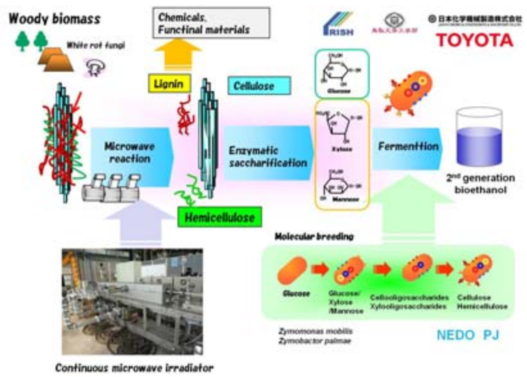

Fig 2: Production of bioethanol from woody biomass by microwave reaction and ethanologenic bacteria (NEDO project).

display cellulase on the surface of the cells, are being bred. A high-performance fermentation process using the ethanologenic bacteria is being developed to produce bioethanol from fast growing wood. A bench scale plant for bioethanol production using the pretreatment system and genetically engineered bacteria was build in 2010.

4 CHEICALS AND VALUE-ADDED PRODUCTS FROM LIGNIN FOR LIGNOBIOREFINERY

Lignin is important as a raw feedstock for the production of aromatic chemicals due to difficulties to obtain aromatics from carbohydrates and its abundance next to cellulose. Production of value-added chemicals from lignin increases overall cost performance of biorefinery including bioethanol production. Therefore, we have been studying conversion of lignin to functional polymers and low

Fig 3: Bench scale plant for bioethanol production from woody biomass (NEDO project).

molecular mass aromatics by microwave reactions. We also investigate detailed structure of lignin by ultra-high sensitivity NMR and ultra-high resolution mass spectroscopy. Potentials of lignin-derived chemicals are discussed.

5 CONCLUSIONS

In enzymatic conversion of lignocellulosics it is necessary to decompose the network of lignin in lignified plant cell walls. Production of value added products from lignin is also important issue due to its abundance and demand for the supply of aromatic chemicals from renewable resources. Thus, effective lignin-degrading pretreatments are needed for the lignocellulosic biorefinery. Our group has been focusing on ligninolytic systems of selective white rot fungi and microwave-assisted degradation of lignin, aiming at producing biofuels and biochemicals from a wide range of biomass including recalcitrant softwood. We have applied the fungal pretreatments to the production of bioethanol, biomethane, and feed for ruminant animals. New microwave reactors have been developed using a simulation technique for an irradiation cavity, and used for the pretreatment of various woody biomass. Pretreatments with white rot fungi and microwave irradiation increased efficiency of enzymatic saccharification and fermentation. A bench scale plant for bioethanol production using the pretreatment system and gene-engineered bacteria has been built in 2010. A joint research covering the lignin utilization and production of biofuels acelerates the establishment of lignocellulosic biorefinery.

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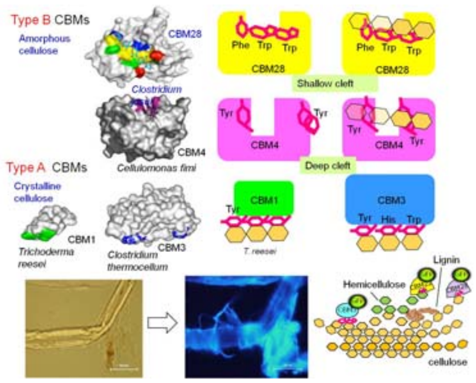

Fig 4: Characterization of exposed carbohydrates with fluorescence-labelled carbohydrate binding module (CBM) in pretreated biomass

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POTENTIAL OF USING ORGANIC FERTILIZER TO CULTURE *CHLORELLA VULGARIS* FOR BIODIESEL PRODUCTION

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ABSTRACT

In the present study, organic fertilizer was used as nutrient source to culture *chlorella vulgaris* for biodiesel production. High concentration of nitrate content in the culture medium was found to be one of the determining factors to promote the growth of *chlorella vulgaris*, in which the specific growth rate was increased from 0.132 day^{-1} to 0.229 day^{-1} with initial nitrate content of 1.33 mg/L and 26.67 mg/L, respectively. Apart from that, *chlorella vulgaris* was found to grow favourable under 24 h of continuously illumination. The growth rate was retarded by nearly 17 % if illuminated under a photoperiod of 12 h light: 12 h dark. In addition, *chlorella vulgaris* could sustain their growth within a wide boundary of pH value, ranging from 4 and 8. Nevertheless, slow specific growth rate was observed when *chlorella vulgaris* was cultured at open environment; a reduction of 27 % was recorded in comparison with controlled environment. On the other hand, it was found possible to reutilize the water to re-culture *chlorella vulgaris*. This observation reflects the high adaptability of *chlorella vulgaris* towards the environment surrounding and suitable to be cultured at outdoor. Total lipid of 18.1 % from *chlorella vulgaris* biomass was successfully extracted and the fatty acid methyl ester profile was proven to be suitable for making biodiesel.

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1 INTRODUCTION

Sustainable energy development has become one of the key challenges in this century. Depletion of fossil fuel in the near future and the effect of green house gases towards human and environment have escalated the search of renewable energies, such as solar, wind, hydro, wave and geothermal power. However, these renewable energies are heavily dependent on regional or local condition that can be very unpredictable and inconsistence. On the other hand, biodiesel being recognized as a green and alternative renewable fuel has attracted great interest from researchers, governments, local and interna- tional traders. Some of the advantages of using biodiesel instead of fossil diesel are it is a non-toxic fuel, biodegradable and lower emission of green house gases when burned in diesel engine [1]. Commonly, biodiesel is produced through transes- terification reaction, in which vegetable oil reacts with short chain alcohol (e.g. methanol) in the presence of catalyst (e.g. sodium hydroxide, NaOH) [2]. Edible oils such as soybean, sunflower, rapeseed and palm oil are the common feedstock for biodiesel production. However, continuous growth of the biodiesel industries in the last ten years has raised a tragic social issue, the food versus fuel dispute. It is strongly believed that further expansion of biodiesel industries in the global market will result to more undernourished people suffering from hunger and malnutrition [3].

Currently, microalgae have been identified as a superior feedstock for biodiesel production, mainly due to their fast growth rate (100 times faster than terrestrial plant) and ability to double their biomass in less than one day under favourable culture conditions [4]. Apart from that, certain microalgae strains are able to accumulate sufficient amount of lipid that can be expected to beat the terrestrial crops. For example, microalgae with 30 % of lipid content have the potential to produce 54 tonnes of oil/ha/year, whereas palm oil and jatropha are only able to produce 3.62 and 4.13 tonne of oil/ha/year, respectively [5, 6]. Thus, culturing microalgae for biodiesel production requires only a minimum land area and up hold an important key for more sustainable land utilization. Apart from that, an added advantage of culturing microalgae is the ability of the microorganism to act as a carbon sink and able to assimilate CO_2 from atmosphere and flue gases through photosynthesis; a golden opportunity for carbon credit program [7, 8].

However, one of the limitations to culture microalgae in industrial scale is the availability of nutrients sources. Chemical or inorganic fertilizers are commonly used to achieve promising growth rate of microalgae. Nevertheless, a life cycle assessment on microalgae cultivation has underlined that 50% of energy use and green house gases emissions are associated with chemical fertilizer production [9]. Meanwhile, utilizing secondary or tertiary wastewater as nutrients source to culture microalgae appears as a promising choice to reduce the overall energy input [10-12]. This is because wastewater contains high concentration of nitrate and phosphate that promote the growth of microalgae and at the same time, microalgae can act as a purifier to further remove the nutrients from wastewater before discharging to water sources. Nevertheless, the key challenges of using wastewater are the susceptible contamination and inconsistent of nutrients compos- ition, in which these factors will directly retard the growth of microalgae significantly. On the other hand, using organic fertilizer as a nutrient source offers an alternative and convenient way to minimize the contamination level in water. Organic fertilizer can be obtained through a series of decomposition and refining process from waste materials such as manure, sewage sludge, biomass and food [13]. Normally, organic fertilizers available in the market are made in dry pellet form for easy transportation and storage. Hence, recycle and reuse the waste materials to culture microalgae for biodiesel produc- tion appears as a greener solution to drive the industry towards sustainable growth.

Thus, the objective of the present study is to evaluate the potential of using organic fertilizer to culture *chlorella vulgaris*. Various culture parameters such as amount of nitrates, illumination duration, pH, outdoor cultures and the effect of reuse the recycling water were investigated systematically. All experi- ments were performed using 5 L photobioreactor (except seed culture) instead of using lab scale conical flask and tap water was used rather than using distilled or sterilized water. This approach was used with the intention for easier up-scaling purposes and to minimize the overall energy consumption in cultivating microalgae in commercial scale.

2 METHODS

2.1 Pure microalgae strain and culture condition

A wild-type *chlorella vulgaris* was isolated from local freshwater located at Penang, Malaysia. The microalgae was preserved and cultured in Bold's Basal Medium (BBM), consisting of $NaNO_3$ (25 g/L), $CaCl_2{\cdot}2H_2O$ (2.5 g/L), $MgSO_4{\cdot}7H_2O$ (7.5 g/L), K_2HPO_4 (7.5 g/L), KH_2PO_4 (17.5 g/L), NaCl (2.5 g/L), EDTA anhydrous (5 g/L), KOH (3.1 g/L), $FeSO_4{\cdot}7H_2O$ (0.05 g/L), H_2SO_4 (1 mL), H_3BO_3 (0.11 g/L), $ZnSO_4{\cdot}7H_2O$ (0.088 g/L), $MnCl_2{\cdot}4H_2O$ (0.014), MoO_3 (0.007 g/L), $CuSO_4{\cdot}5H_2O$ (0.016 g/L), $Co(NO_3)_2{\cdot}6H_2O$ (0.005 g/L). The initial pH of the medium was adjusted to 6.8. The culture was grown in a 100 mL Erlenmeyer flask containing 50 mL of medium, aerated with compressed air, surrounding temperature of 25-28^oC and illuminated with cool-white fluorescent light (Philip TL-D 36W/865,

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light intensity of 60-70 μmol m^{-2} s^{-1}) continuously for 24 hours.

2.2 Microalgae cultured in organic fertilizer medium

Organic fertilizer (Baja Berbajadi Humus 27) was purchased from a local market. 10 g of the fertilizer was immersed in 600 mL tap water and stirred for 24 hours using magnetic stirrer. Non-soluble particulate solids were observed after the stirring process and were filtered using filter paper (Double Rings 101). The resulting organic fertilizer medium was dark-brown in colour with initial nitrate content of 1.33 g/L. Subsequently, a pre-determined volume of the organic fertilizer medium was introduced into a photobioreactor with 5 L of tap water (without steril- ization) and the pH of the medium was adjusted according to pre-determined values. Then, *Chlorella vulgaris* with initial cell concentration of 0.3×10^6 cell (around 10 mL from the seed culture) was introduced into the photobioreactor. The photobioreactor was aerated with compressed air continuously and illuminated with cool-white fluorescent light (Philip TL-D 36W/865, light intensity of 60-70 μmol m^{-2} s^{-1}.

2.3 Measurement of microalgae growth

A correlation between the optical density of *chlorella vulgaris* and their biomass was pre-determined. Optical density was measured daily at 540 nm using spectrophotometer (Shimadzu UV mini-1240). Then, 10 mL of sample were centrifuged at 10 × 1000 g for 5 min. The supernatant was slowly decanted back into the culture medium whereas the microalgae biomass was dried in an oven at 100°C for 24 h. All the samplings were performed in triplicate to ensure the accuracy of the data. The correlation is shown in Eqs (1):

$$Dry\ weight\ (g/L) = 0.4541 \times OD_{540}\ ,R^2=0.9890 \quad (1)$$

The specific growth rate (μ) was measured by using Eqs (2):

$$\mu\ (day^{-1}) = \frac{\ln(N_2/N_1)}{t_2 - t_1}$$

where N_1 and N_2 are defined as biomass (g/L) at time t_1 and t_2, respectively.

2.4 Measurement of nitrate content in medium

Nitrate concentration was determined according to the Ultraviolet Spectrophotometric Screening Method [14]. A sample (1 mL) collected from photobioreactor was centrifuged at 10×1000 g for 5 min. The supernatant was collected and optical density was measured at 275 nm and 220 nm by using spectrophotometer (Shimadzu UV mini-1240). Then, the absorbance reading at 275 nm was subtracted two times from the reading at 220 nm to obtain the actual absorbance caused by the presence of nitrate. Dry potassium nitrate (KOH) at different concentrations was used for calibration purposes.

2.5 Microalgae harvesting and biomass collection

When *chlorella vulgaris* had grown to stationary phase, air aeration to the culture medium was stopped. The microalgae were let to settle naturally to the bottom of photobioreactor for two days. Two distinguish layers were observed, in which the upper layer consist of water with suspended microalgae cells and the bottom layer was microalgae biomass. The upper layer water was slowly decanted, leaving behind the microalgae biomass which was further dried in an oven at 100°C for 24 hours. The dried microalgae biomass was collected and sealed in an empty container for lipid extraction.

2.6 Microalgae lipid extraction

10 g of dried *chlorella vulgaris* biomass was placed in a cellulose thimble and extraction process was performed using Soxhlet extractor. Four types of solvent were used to compare the extraction efficiency, such as n-hexane, methanol, ethanol and mixed methanol-chloroform with volume ratio of 2:1 [15]. A total of 250 mL for each solvent was placed in Soxhlet extractor and heated at 60-65°C for 24 hours. After that, the solvent was evaporated in a rotary evaporator and the leftover lipid was collected. The residues (solid material) after evaporation were subjected to repeated extraction twice using the same solvent. Thereafter, crude microalgae lipids were measured gravimetrically. All samplings were performed in triplicate.

2.7 Transesterification reaction and fatty acid methyl ester (FAME) analysis

Transesterification was performed by using 0.1 mg of crude *chlorella vulgaris*, methanol to lipid molar ratio of 15:1 and 3 wt % of concentrated sulfu- ric acid (H_2SO_4) as catalyst. The reaction was carried out in a water bath shaker at 60°C for 3 h. Upon completion of the reaction, 1 μL of the reaction product was subjected to gas chromatography-mass spectrometry (GC-MS; PerkinElmer Clarus 600) ana- lysis. The GC was equipped with flame ionization detector (FID) and Elite 5-MS column (30 m × 0.25 mm ×0.25 mm). The initial oven temperature was 65 °C, held for 2 min and raised to 280 °C at ramping rate of 8 °C/min and held at 280 °C for 10 min, while the injector temperature was set to 250 °C. The compounds detected were identified and quantified by using NIST Mass Spectral Search Program.

3 RESULT AND DISCUSSION

3.1 Effect of nutrients concentration

Supplement of sufficient nutrients for microalgae to grow is the first key step to produce

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bulk quantity of microalgae biomass. Among all the nutrients required, adequate supply of nitrogen sources such as nitrate (NO_3^-), ammonia (NH_4^+) and urea appear to be the most significant contributor to sustain microalgae growth. In the present study, synthesized nutrient from organic fertilizer were diluted to different concentration with the purpose to test the ability of *chlorella vulgaris* in up taking the nutrients. Fig. 1 depicts the growth of *chlorella vulgaris* with different nitrate concentration. From the figure, *chlorella vulgaris* were found to be grown even under limited nutrients condition (5 and 10 ml of nutrients), however, the quantity of biomass produced were unsatisfactory which were 0.1-0.14 g/L even after 12 days of cultivation. In comparison, *chlorella vulgaris* grew much better with sufficient supplement of nutrients (80 and 100 ml). The biomass produced were 0.29-0.31 g/L, which was nearly 150 % of increment.

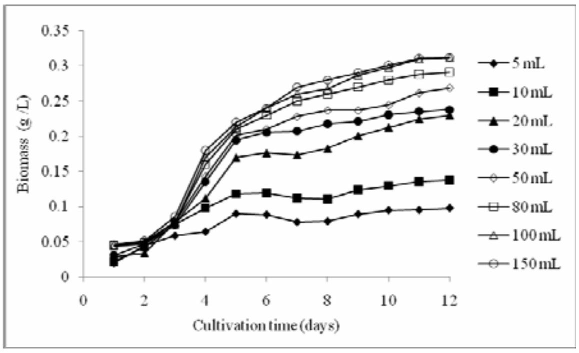

Fig.1: Effect of initial nitrate concentration towards the growth of *chlorella vulgaris*. Other culture condition: pH = 7 and illuminated for 24 h continuously. Initial nitrate content (mg/L): 5 mL= 1.33, 10 mL= 2.69, 20 mL= 5.38, 30 mL=7.98, 50 mL= 13.09, 80 mL= 20.94, 100 mL=26.67, 150 mL= 40.71.

Apart from that, specific growth rate of *chlorella vulgaris* was also improved with sufficient nutrients supplement. For example, the specific growth rate was 0.132 day^{-1} and 0.229 day^{-1} for 5 and 100 ml nutrients added respectively; an increment of 73 %. This observation indicates that huge quantities of nutrient sources are the mandatory step to promote microalgae growth and consequently, to attain higher biomass yield. Hence, utilization cheap nutrient sources such as organic fertilizer will be more bene- ficial in large scale microalgae culture industry, typically in term of cost saving and environmental protection. On the other hand, the growth curve for *chlorella vulgaris* with excess amount of nutrients (150 ml) exhibited the same trend as 100 ml, in which no further improvement of biomass yield was observed. This is because other parameters such as photoperiod and pH control will become the growth determining factors if adequate nutrients are supplied.

3.2 Effect of photoperiod

The effect of photoperiod on microalgae cultivation exhibited an exceptional significant influence towards microalgae photosynthetic activity and growth rates in a typical photobioreactor [16, 17]. However, it should be noted that microalgae with excessive light exposure would not only resulted to unwanted electricity consumption (energy waste) but also inhibit the growth of microalgae [18]. Fig. 2 shows the effect of photoperiod towards the growth of *chlorella vulgaris*.

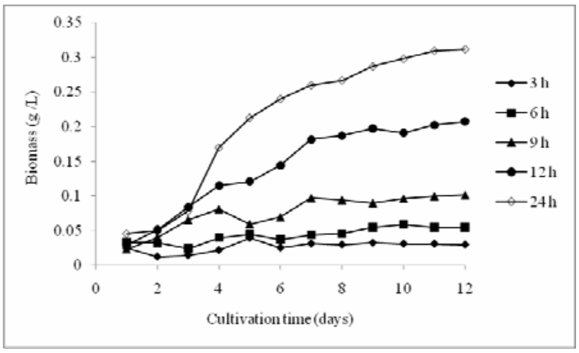

Fig.2: Effect of photoperiod towards the growth of *chlorella vulgaris*. Other culture condition: Initial nutrients volume = 100 mL (nitrate content of 26.67 mg/L) and pH = 7.

From the figure, the growth of *chlorella vulgaris* were strongly affected by photoperiod, with the highest biomass attained at 0.31 g/L through continuous 24 h light illumination for 12 days (specific growth rate of 0.228 day^{-1}). In addition, for photoperiod of 3 h, 6 h and 9 h, the biomass attained were only 0.03-0.1 g/L which was exceptional unsat- isfactory. Therefore, longer photoperiod will result to continuous uptake of nutrients by *chlorella vulgaris* through photosynthesis and subsequently, increase their biomass at the same cultivation time. In other words, this result also indicates that *chlorella vulgaris* is weak in storing substantial amount of light energy for later use in the dark period (exergonic reactions) [17]. Hence, surface area of illumination should be taken into consideration when designing a photobioreactor to culture *chlorella vulgaris* (typically outdoor cultivation), so that optimum light energy can be utilized by the microalgae efficiently. From Fig. 2, it can also be clearly seen that the quantity of biomass at 12 h of photoperiod time (equivalent to outdoor sunlight cycle) is almost double the quantity of biomass obtained at photoperiod of 3 h, 6 h and 9 h. This result proved that it is feasible to culture *chlorella vulgaris* at the outdoor with open system; however, it is still dependent on the local weather condition and temperature.

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3.3 Effect of initial pH

One of the determining factors to grow microalgae is the pH of the culture medium. Generally, pH of 7-8 is plausible for microalgae to grow. Nevertheless, it is worth to mention that microalgae strains for biodiesel production should not only contain high lipid yield, but able to withstand pH changes during cultivation cycle. For example, high pH value (9.0-9.5) was observed in photobioreactor due to shortage of inorganic carbon after rapid consumption by microalgae [19]. Sometimes, pH can also drop drastically if high concentration of CO_2 is continuously supplied to the culture medium and caused adverse effect towards the growth of microalgae [11]. More importantly, in the near future, when flue gases are used to culture microalgae in industrial scale, the selected microalgae strains should be able to tolerate incon- sistence concentration of CO_2 in the flue gases that indirectly resulted to pH variation in the culture medium. Fig. 3 shows the effect of initial pH in the culture medium for *chlorella vulgaris*.

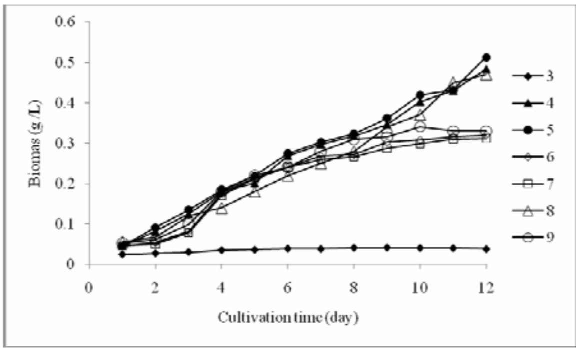

Fig.3: Effect of initial pH towards the growth of *chlorella vulgaris*. Other culture condition: Initial nutrients volume = 100 mL (nitrate content of 26.67 mg/L), illuminated for 24 h continuously.

From the figure, *chlorella vulgaris* exhibited almost a linear growth at pH of 4, 5 and 8 with maximum biomass of 0.47-0.51 g/L obtained after 12 days of cultivation. Furthermore, higher specific growth rates were obtained at pH 4 (0.265 day^{-1}), 5 (0.270 day^{-1}) and 8 (0.263 day^{-1}) compare to culture growth at neutral condition, pH 7 (0.229 day^{-1}). This result indicates that *chlorella vulgaris* can adapt very well even at low or high pH. One of the advantages of growing microalgae at low or high pH is the simultaneous elimination of contaminants such as fungus and able to sustain the growth of microalgae naturally. Nevertheless, at pH 3, the growth of *chlorella vulgaris* was reverted and no significant increment of biomass was detected after 12 days of cultivation. On the other hand, although the growth of *chlorella vulgaris* is satisfactory at pH 6, 7and 9, a lag phase was observed from day 1 to day 4 and the growth was almost stagnant from day 9 to day 12 with a maximum 0.33 g/L of biomass obtained.

3.4 Effect of culturing in open environment

Up to date, research on microalgae culture is focussed on indoor culture where a control environment can be easily achieved. Nevertheless, for the purpose of culturing microalgae in a comer- cial scale, outdoor culture will be the most feasible option due to easy access to sunlight. This will reduce overall energy input (mainly for illumination and temperature control) and consequently, enhance the cost effectiveness to produce biodiesel from microalgae. However, one of the limitations to culture microalgae at outdoor is the inconsistent changes in local weather, temperature and light intensity. In this scenario, microalgae strains with high lipid content and ability to tolerate with these external changes will be an added advantage. Fig. 4 shows a comparison growth of *chlorella vulgaris* at indoor and outdoor condition. Error bars were indicated in the figure for microalgae cultured at outdoor due to the inconsistent changes in weather and temperature.

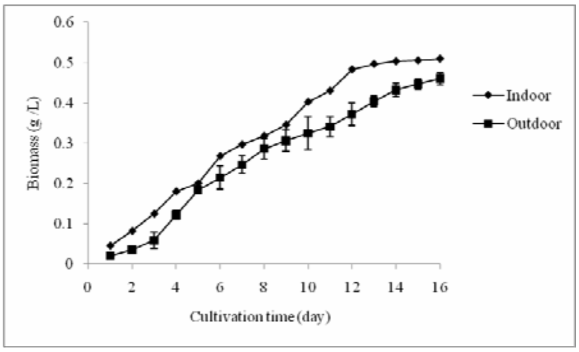

Fig.4: Effect of outdoor towards the growth of *chlorella vulgaris*. Culture condition: Initial nutrients volume = 100 mL (nitrate content of 26.67 mg/L), illuminated for 24 h continuously and pH = 5.

From the figure, *chlorella vulgaris* grew slower at outdoor compared to the indoor culture. Furthermore, a lag phase was detected from day 1 to day 5, indicating that the microalgae required some time to adapt themselves to new environment. Therefore, longer cultivation time is definitely required to grow *chlorella vulgaris* at outdoor, typically 16 cultivation days was needed to reach the optimum biomass yield as shown in Fig. 4. As a consequence, the specific growth rate of microalgae cultured at outdoor dropped nearly 37.8 %, to 0.196 day^{-1} in comparison with indoor culture (0.270 day^{-1}). Nonetheless, the growth of *chlorella vulgaris* was not seriously affected by the outdoor temperature and weather, since the temperature varied from minimum 23°C to maximum 35°C and lack of sunlight during raining days. Thus, it is expected that *chlorella vulgaris* is relatively easier to culture in open system due to their high tolerance characteristic towards the

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surrounding environment. Apart from that, it was found that the *chlorella vulgaris* cultured at outdoor attained higher biomass yield (0.37 g/L) on day 12 compared to indoor culture with 12 h illumination (0.21 g/L). This may be due to the intensity of sunlight that is much more suitable to grow *chlorella vulgaris* compare to illumination by fluorescent lamp.

3.5 Effect of using recycle water for culture

Effective uses of water resources play a significant role in microalgae cultivation since micro- algae are aquatic microorganisms that need water to survive. In large scale microalgae cultivation system, huge amount of water is required to reach optimum quantity of microalgae biomass. Therefore, recycle and reuse of water during microalgae cultivation not only can save the overall operation cost, but also helps to preserve the water for other purposes. However, not all microalgae strains are able to grow effectively in recycle water because it is susceptible to contamination by fungus and bacteria, typically in an open pond system. Hence, the selected microalgae strains for biodiesel production should be able to grow rapidly and withstand with other contaminants. Fig. 5 represents the effect of using recycle water from the previous batch culture (indoor and outdoor, respectively) without any pre-treatment or puri- fication process. The recycled water still contained minimum amount of nutrients and microalgae cells which were not completely harvested.

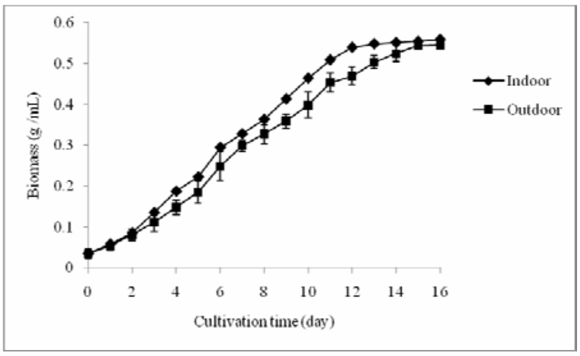

Fig.5: Effect of reuse the recycling water towards the growth of *chlorella vulgaris*. Culture condition: Initial nutrients volume = 100 mL (nitrate content of 26.67 mg/L), illuminated for 24 h continuously and pH = 5.

From the figure, the growth of *chlorella vulgaris* in indoor was not affected by the use of recycle water and produced satisfactory quantity of biomass within 16 days of cultivation time. Apart from that, for *chlorella vulgaris* cultured at outdoor using recycle water, the lag phase from day 1 to day 5 was greatly minimized if compared with the use of fresh water (Fig. 4). This is because *chlorella vulgaris* that were not harvested and remain in the recycle water have already adapted to the outdoor environment. Therefore, once new organic nutrients were supplied to the culture medium, the microalgae can utilize the nutrients immediately. Consequently, higher quantity of biomass (0.55 g/L) was obtained compared to outdoor culture with the use of fresh water (0.37 g/L).

3.6 Lipid extraction and biodiesel production from chlorella vulgaris

Up to now, lipid extraction using chemical solvent is the most reliable method to determine the overall lipid content presence in microalgae. Extraction efficiency by chemical solvent is higher than physical method (mechanical pressing machine), typically due to the high polarity of fatty acids toward the chemical solvent. Nevertheless, the effectiveness of different type of chemical solvent used is strongly dependent on microalgae strains, especially when the existence of cell wall in microalgae will indirectly impede chemical solvent extraction efficiency [20]. In the present study, four different types of solvent were used for lipid extraction study: n-hexane, methanol, ethanol and methanol mixed with chloroform with volume ratio of 2:1 (Bligh and Dye method). Bligh and Dye method gave the highest extraction efficiency, in which 18.1 % of lipid was extracted out. This value is comparable to the total lipid of *chlorella vulgaris* reported in the literature, which lies between 5-40 % under phototrophic culture condition [21]. The solvent extraction efficiency in descending order is methanol (15.5 %), ethanol (10.7 %) and n-hexane (3.2 %). Although n-hexane has been widely used in oil extraction study, nevertheless, it is a not a suitable choice to extract lipid from *chlorella vulgaris*. This is because n-hexane is a non-polar solvent which has poor permeability in cell walls and hence, resulted to only extracellular lipids are extracted [22].

The fatty acids methyl ester composition of *chlorella vulgaris* are mainly consisted of C16:0, C16:1, C16:2, C18:1, C18:2 and C18:3 as shown in Fig. 6. From the figure, C16:0 (palmitic acid methyl ester), C18:1 (oleic acid methyl ester) and C18:2 (linoleic acid methyl ester) contributed the major portion of fatty acid methyl ester composition, accounted for total of 83.2 %. These fatty acids are naturally found in oil bearing crops, such as soybean, sunflower, cottonseed and palm oil, in which the fatty acids are suitable for biodiesel production. Apart from that, unsaturated fatty acids methyl ester (C16:1, C16:2, C18:1, C18:2, C18:3) are predo- minant in the fatty acids profile, accounted for 85.1 %. It is important to note that higher composition of unsaturated fatty acids methyl ester can reduce the pour point of biodiesel and making it feasible to be used in cold climate countries. Other saturated fatty acids methyl ester except C16:0 (palmitic acid methyl ester) were also identified, such as C14:0 (myristic acid methyl ester), C15:0 (pentadecanoic acid

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methyl ester) and C18:0 (stearic acid methyl ester). However, the composition is minimum, accounted for only 1.56 % of the overall lipid composition.

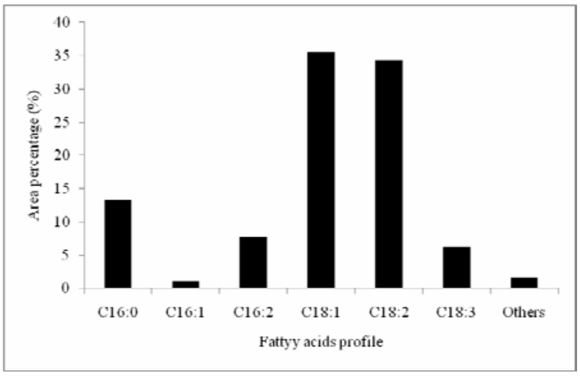

Fig. 6: Fatty acid profile of *Chlorella vulgaris*.

4 CONCLUSIONS

The result in this study showed that it is feasible to use organic fertilizer to culture *chlorella vulgaris* for biodiesel production. This approach offer several advantages in term of environmental perspective and cost effectiveness. In addition, this method can be easily adopted since organic fertilizers are available worldwide and *chlorella vulgaris* is easy to cultivate due to their high adaptability towards the environment surrounding. Furthermore, reutilize the water to re-culture *chlorella vulgaris* is possible and thus, further driven the microalgae biodiesel industry towards more sustainable development.

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BIOFUELS IN PERU
CURRENT SITUATION AND PERSPECTIVES

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1 INSIGHT ON BIOFUEL IN PERU

Any country seeking growth through development must necessarily commit to infrastructure and human capacity building. The connection between these two concepts is competitiveness. And Peru is hindered by a series of structural obstacles to competitiveness. As it is, only the total deficit of infrastructure is estimated in almost 35 billion Dollars, approximately a third part of Peruvian Internal Gross Product[2]. Energetic infrastructure deficit in Peru is estimated in more than 12 billion Dollars, representing almost a tenth of the IGP[3]. Out of this deficit, 8.3 million Dollars correspond to deficit in electric infrastructure, and 3.7 million Dollars to deficit in natural gas infrastructure[4]. In accordance with certain estimates by the Peruvian Economy Institute, at present, approximately 615 million Dollars investment is required in electric generation, transmission and coverage and 345 million Dollars in infrastructure to exploit natural gas[5].

A large part of this deficit is found in the poorest zones of the country, both urban but mainly rural, where it is difficult and costly to put the energetic offer to work. This caused the deficit to directly influence the poorest population segments which, added to other road infrastructure deficits, communications and health, deepen the conditions of inequality and margination, in which a large number of Peruvians subsist. As it is, a very clear relation is observed in Peru between limited energetic coverage and poverty. If we just analyse the electric coverage, such Departments with high index of poverty as Huanuco, Loreto, San Martin, Apurimac, and Huancavelica show a low coverage, while Departments that register a lower index of poverty, such as Lima, Tacna, Arequipa and Lambayeque, at the same time enjoy higher coverage of the electric service. Diverse structural conditioning both geographical as demographical and social, makes the implementation of rural energetic coverage very expensive, or at least, not always guarantees positive social results (current net social values higher or equal to zero[6]) and in all cases generates negative private results (current net social values lower than zero[7]). All of which cause it to be seen as a “marginal” issue among infrastructure implementation priorities in the country.

However, improper energetic supply, at integral level, that is, not only concentrated in large urban centres but also in rural zones, give rise to impacts that transversally affect the country's productive forces. Under this perspective, public policies oriented to diversifying the energetic matrix, taking advantage of our so valuable potential of renewable and natural gas, added to the task to reduce this deficit in energetic infrastructure, if duly applied, may generate an important impact not only on the direct beneficiaries but on the country´s competitiveness.

The economic impact of the integral energetic shortage, that is, such which covers the whole of the Peruvian population, is unfolded in two sub-impacts. Each of them influences different temporary horizons. Direct impact is immediate: lack of energy makes it difficult, if not impossible, for the population to gain access to an essential utility, and wastes the possibility that would be offered by modernity to raise their life condition. Starting from the proper preparation of meals to the necessary light to study, going through access to information technologies such as Internet and the appropriate public safety; absence of appropriate energetic supply in rural zones becomes the most important element for population to gain access to these services.

Approximately a fifth of the Peruvian population, that is, almost 6 million people live in condition of poor energetic supply. Who are automatically "left out" of modernity, including quality utilities, market economy and its potential benefits. This generates a material and sustained impairment of life quality of the ones directly affected, who are irremediably dragged to deepen their condition of poverty. In addition, it creates an incentive for migration in search of access to better utilities, which results in an ever-growing crowd up of urban centres that little by little find their own structured collapsed, in the face of the explosive increase of demand.

Indirect impact is distributed along the various transversal planes, which stops focusing the problem only on affected populations. The first thing impacted is the economic growth, that is, if the commitment to infrastructure is, as expressed above, an essential requirement to economic growth. A first corollary is also true: investment in infrastructure further leverages economic growth. According to certain studies[8],

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[2] Estimated as of year 2008.

[3] Again, 2008.

[4] See *Instituto Peruano de Economía (Peruvian Economy Institute) Infrastructure challenge to 2018.* Lima, 2009.

[5] Ibíd.

[6] VAN social >0.

[7] VAN private <0.

[8] See Calderon Cesar and Luis Serven. The Effects of Infrastructure Development on Growth and Income Distribution. Central Bank of Chile.

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an improvement in the level of Peruvian infrastructure, in general, only at the level of the leading country in Latin America in the matter, that is, Costa Rica, would increase its economic growth rate by 3.5%, while should it reached the level of Chile, the impact on the growth rate would be by 2.2%. Therefore, a second corollary is also true: that infrastructure impairment lessens the potential for growth, and, to aggressively stop the effort to close all the sectorial infrastructure gaps hampers and reduces the potential of whole country, and not only the specific community which suffers the gap.

In addition, energetic shortage seriously affects the horizon of people that have been impacted by it since infancy and their possibility to properly compete in labour and product markets (goods and services). In other words, energetic shortage for these Peruvian citizens translates in loss of human capital because it prevents them from receiving suitable educational and health services, including the nutritional element, which are indispensable for human development. Such decapitalization impairs competitiveness of each individual, though, in the long term, impairs territorial and the country's competitiveness.

Likewise, impairment of human development level as a result of the permanent fall of life quality and the utility supply-demand gap derived from the migration countryside-city, irremediably impacts, in the long term, on policy pressure for raising tax to pay for, as social welfare, releasing the poverty resulting from these processes. As any fight for tax resources, integral energetic shortage is the perfect scenario for social conflict that affects the investment environment and the increase of the country's capitalization, both necessary instruments to raise employment, personal incomes, and the consequent life quality of the residents of a country. In turn, tax increase produces further distortions to economy and loss of irrecoverable social efficiency, which conspire against a more competitive performance of the national productive forces, in a transversal manner. Tax increase generated by not timely responding to energetic requirements for rural and peri-urban zones, end up thus hardly alleviating, in a very insignificant proportion, the permanent economic loss created in shortage or inexistence of provision of utilities. It thus looses its re-distributive potential, only to become some kind of payment to postpone the time to address the problem.

The other problem is that deficient energetic rural structure deepens inequality countryside-city, and inequality in the country, in general. It is estimated that if Peru reached the infrastructure levels of Costa Rica and Chile, reduction in its Gini-coefficient of national income distribution [9] would be 0.070 and 0.044 respectively[10], which implies that just providing with a better infrastructure at the levels already arrived at by other Latin American countries could lavishly improve the existing situation of huge social inequality in Peru. As a matter of fact, the impact of a proper and diversified energetic infrastructure would be very powerful on the lower income social strata, since it is in these poorest zones where the implementation of a suitable energetic infrastructure generates strong and dramatic changes in families' incomes and the acquisition power of the poor. Some estimates show that the sole implementation of electric and water services would generate an increase by almost 30% in household incomes which lack the same[11].

In consideration of all the foregoing, the economic impact of not addressing this energetic infrastructure gap on the most depressed locations in the country, which belong to the rural and peri-urban sphere, is not limited to the loss of life quality of those directly affected, as it is usually considered. The truth is that an idea is conveyed that addressing these demands is a "human" or "solidarity" issue. However, it turns out to be an issue of national competitiveness, due to the derived effects, which ends up

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Series Working Papers No. 270. Santiago de Chile, 2004.

[9] Gini-coefficient is a measurement typically used to measure inequality of income in a human group, usually a country. It is a number between 0 and 1, where 0 corresponds to perfect equality (all with the same income) and 1 corresponds to perfect inequality (one person earns all the income and the others none).

[10] See Calderon and Serven, *op. cit*. Hypothetically, if such infrastructure levels may be reached as those of Korea, the impact would be much greater: a reduction of 0.10 in the Gini-coefficient.

[11] See Escobal, Javier and Maximo Torero 2004. *Analysis of rural infrastructure services and life quality in rural zones of Peru. Final Report. Analysis Group for Development (GRADE). Lima, 2004. According to this study* it is estimated that if, in addition, these impoverished households were provided with sewage, income increase would reach 40%, and, if they were provided with telephones, their income would increase by 60%.

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impacting on the whole of the economy, since tax increase is promoted, production costs for the business sector become more expensive, and goods and services used by consumers also become more expensive.

Why is competitiveness so important? Because, nowadays, a country's development is intimately related to its capacity to compete against one another within a global context. The most competitive countries are also the most developed. Notwithstanding, Peru is still not among the most competitive in the world. In line with the criteria of the World Economic Forum, the basic pillars of competitiveness are institutions, macro-economic stability, basic health and education, and infrastructure. With respect to the latter, as far as quality of infrastructure is concern, Peru is in position 103 out of 133 countries. But, in making finer comparisons, it may be appreciated that the infrastructure quality index in Chile is almost twice as much as Peru's[12]. In addition, if we only considered the energetic sector, in regard to electricity, Chile's national coverage is 99.4%, while Peru's only reaches 83.5%, according to official reports[13]. If we add that to Peru's very modest levels of basic health and education, items intimately related to infrastructure, no wonder it is a country where high levels of poverty are registered historically.

As a consequence, public policies oriented to promoting diversification of the energetic matrix and closing the infrastructure gap will directly influence the improvement of competitiveness and therefore, economic development.

[12] See World Economic Forum. *Global Competitiveness Report 2009-2010*. In order to have a clearer idea of the meaning of this position in terms of "quality distances" between countries, Peru's score is 3.0, while Chile's is 5.6, within a scale of 1 to 7, where 1 is absolutely inefficient and 7 fully efficient. Top countries in infrastructure such as Switzerland, Singapore and Hong Kong are around over 6.7.; United States registers a 5.9.

[13] In the case of Chile, we have used information form the Ministry of Planning (MIDEPLAN) through the Survey on Socio Economic Characterization CASEN of 2007, and for Peru, information from the Ministry of Energy and Mines (MINEM) according to estimations.

2 ANALYSIS OF LEGISLATION ON BIOFUELS[14]

At present, interest in the production and use of biofuel has reached high levels, and the regulatory framework is being developed promoted by the interest put in these product as energetic alternative; concern for the environment, and the high prices of oil. On the other hand, the importance of the involvement of the private sector in the production of raw material is evident and in obtaining biofuel due to the level of investments required to supply the local market and for possible exports. The State's duty to set forth clear rules for the whole biofuel production chain that would guarantee the investor's investment is unavoidable, in addition to a competitive product in the fuel market. Likewise, a clear regulatory system is essential to govern the chain: producer (farmer and industrial) – investor – trader – purchaser – refiner – distributor – regulator – auditor – exporter – final user. As long as the whole production and use chain has clear regulations to guarantee an acceptable profitability and a market-competitive fuel, the conditions to have access to local environmentally friendly fuel are thus created, made with local raw material, and, which in addition, generates jobs and productive chains in the agricultural, trading and financial sectors.

3 EVOLUTION OF REGULATORY SYSTEM FOR BIOFUELS

The legal framework which governs the activities of the private sector for the production of biofuels is being developed in connection with the specific interest of certain countries in these products as energetic alternative. To that effect, Brazil is the first country which oriented its activities towards that perspective; and therefore, it was forced to create the conditions to promote the production and use of ethanol and subsequently biodiesel. At present, different countries are in a different phase of development (Chile, Cuba, El Salvador, Mexico, Nicaragua, Panama, Peru, Dominican Republic, Uruguay, Venezuela), and in some cases, laws updating in such countries as: Costa Rica, Guatemala and Honduras.

[14] OLADE, 2008, Analysis of Legislation on BioFuel in Latin America.

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4 METHODOLOGY OF ANALYSIS

The central idea of this analysis is to acknowledge and assess the measures, actions, and policies adopted by a State to promote the production and use of biofuels, making this activity attractive to investors. In relation hereto, the following analysis parameters have been determined: Definition of biofuel, Authority to apply the regulation - Institutional framework, Biofuel Producer Requirements, Biofuel distributor requirements, Subjects benefited by the promotional regime, Tax Regime, Terms or periods for the application of the regulation, Percentage of biofuel mix, Environmental aspect and Regulations. Likewise, the issues to analyse comprise interinstitutional participation, clarity of control and inspection functions, legal security, guarantee for investments, proper pricing policy, guaranteed market for biofuel, product final consumer rights, etc. The scheme proposed intends to analyse indispensable elements such as efficiency, because it is strictly related to the situation, which will govern by means of the harmonic combination of technical and legal aspects; and, efficacy of public policies, which is given through the compliance with the objectives proposed.

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BIOFUELS IN PERU CURRENT SITUATION AND PERSPECTIVES

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1 ETHANOL

Peruvian law mandates the mix of 7.8% of anhydrous ethanol with gasoline for the production of GASOHOL and BIODIESEL

1.1 Gasohol

- The salient provisions of the Biofuels Act include the following
- In Peru Ethanol is produced based in the cultivation of sugar cane mainly in the coastal deserts areas of the north of the country. Peru enjoys very favorable conditions for the production of sugar of cane being able to produce 50% more per Hectare than Brazil. Water in northern Peru has become the key issue since rivers are basically dry most of the year with the exception of the Chira River and rain is very scarce.
- Peru currently has one plant built with Brazilian technology specifically for the processing of anhydrous ethanol. This plant is capable of producing about 350,000 liters of anhydrous ethanol per day. There are two additional facilities being installed also in Northern Peru. One belongs to Maple Ethanol which is a subsidiary of Maple Energy which is basically built with the same Brazilian technology as the one currently operating and will produce approximately the same amount of liters of anhydrous ethanol as the first one. This second facility will start production during the second semester of 2011.
- The third facility is being built with Praj India's technology and bases its production on molasses it currently obtains from the production of sugar. This third ethanol plant belongs to a Peruvian group who own the largest sugar cane plantation in Peru. The Rodriguez group own about 60,000 Hectares of land planted with sugar cane and of course have the largest sugar market share in the country. Right now the produce hydrogenated alcohol which will be transformed into carburant ethanol. The installed capacity is said to be of about 300,000 liters per day.
- These three facilities can handle all current and future Peruvian needs of anhydrous ethanol for the production of GASOHOL.
- To date Peru does not have an official Technical Standard for the domestic production of anhydrous ethanol It bases its standards on the US ASTM Norm for this product but it requires a maximum of 0.3% of water content in volume which is a bit more strict than the US Standard.
- Peru is now self-sufficient in sugar as it was until the late sixties and is on the verge of recuperating its former condition of sugar exporter. There is no conflict between the sugar production in Peru and the use of sugar cane for the processing of anhydrous ethanol. Production of sugar cane has increased significantly due to better agronomic practices, more discipline in the workplace and – of course – large investment in equipment and research. With the current amount of cultivated Hectares of sugar cane Peru is capable to satisfy its needs of sugar, industrial and domestic alcohol and anhydrous ethanol for the mix with gasoline. Current sugar production is nearing one million metric tons in close to 80,000 Hectares.
- The law that promotes the biofuels sector in Peru goes back to August of 2003. It has been a matter of continuous political and environmental debates in which lobbyist from all sides of this issue have pushed their own agendas to a point that today after almost eight years it still not clear where the country is going and how the energy matrix will look like vis a vis ethanol and biodiesel. The current government has approved a Decree called State Energy Policy 2010-2040 in which biofuels are almost non existent in the energy matrix although the concept of biomass is mentioned in the context of waste used as a source of energy.
- The current mix mandated by law in Peru is of 7.8% of carburant ethanol. At this time this is being applied in Northern Peru only. The distribution of GASOHOL in the largest market which of course is in Lima the Capital City of Peru has been postponed a number of times due to "insufficient domestic production". GASOHOL is supposed to be distributed at the retail level starting June 1st, 2011 but we are already hearing voices that claim that there is no guarantees that delivery trucks and gas stations tanks are hermetic in order to prevent the new carburant containing ethanol will absorb water before it gets into car tanks. This issue can cause yet another delay which may leave this issue to be

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resolved by the new government to be inaugurated on 7/28/2011.

1.2 Biodiesel

- Is being produced in various areas in Peru. There are Projects being developed in coastal areas as well as in the northeastern rain forests.
- African palm trees are is the plant of choice but lately Jatropha Curcas L has become a strong choice too for the newer Projects being currently developed.
- Currently there is one refinery in place located in the San Martin Region capable of producing 50,000 Metric Tons of biodiesel per year. This quantity is still not sufficient to cover the demand which has grown dramatically since January 1st, 2011 when the government mandated an increased mix to 5%.
- Oil refineries in Peru claim that biodiesel produced in the country does not satisfy their quality standards although no proof of this problem has been presented so far. The claim is basically centered on the inability of Peruvian produced biodiesel of performing in high altitudes and at low temperatures. At this time the biodiesel needs of the Peruvian market is satisfied by means of the importation of foreign material brought in primarily from the US, Argentina and at a lesser scale from Ecuador.
- Notwithstanding the above the Peruvian government increased the mix from 2% (B2) to 5% (B5) as of 1/1/2011.
- Osinergmin, the Peruvian governmental official regulator in charge of controlling the quality of the biofuels being delivered to consumers still lacks of the necessary means to cover nationwide this important task. This situation applies to biodiesel and gasohol as well.
- Imported biodiesel enters Peru duty free unless there are proven dumping practices or subsidies in which case the governmental Trade Commission will establish special tariffs as it has done with US biodiesel being imported. Dumping and subsidies make investments more difficult and at times Projects become not viable.

2 PLANTATION TRUST

2.1 For the farming of Jatropha Curcas

<u>Definition</u>

A plantation trust is established when an individual or an entity puts its property or part of it into a trust which will be managed by a financial institution which obliges itself to comply with the purpose for which the trust has been created under the laws of the country and to use the property only in the best interests of the Trust and of its Trustees. Trusts in Peru can only be established for thirty years but can be extended for similar periods of time.

<u>Fundamentals</u>

SOCIAL: The farming of Jatropha Curcas or Pinon Blanco as we usually call it in Peru promotes the participation of men and women adult and young alike which allows them an opportunity of personal fulfillment giving them a sense of being productive using middle tier technologies which facilitate their social inclusion.

ENVIRONMENT: The farming of Jatropha Curcas demands less stress of soils that have been deforested and marginalized for many years. Jatropha can be farmed in idled areas which have been deforested and degraded with little risk of plagues thus reducing the use of agrochemicals. Jatropha itself can be used as life fences avoiding the use of wood and produces two to three times per year enough foliage which is plenty of organic material that can be used as natural fertilizer reducing the need of expensive and some times inaccessible chemical fertilizers.

ETHICS: Oil produced from Jatropha can only be used to produce biodiesel thus avoiding competition with feedstock used for food. It also allows the farmer to intercrop it with other plants which he can farm for his own consumption or sell. Farmers can also plant exotic tropical flowers like orchids as we have been able to see in places of the rainforests of Brazil and of Peru enabling them to get more income while waiting to harvest the Jatropha.

ECONOMY: Jatropha is a plant that can produce fruit for 30 to 40 years and perhaps even further as we have heard it does in other parts of the world. Therefore it allows the Farmer to enter into consortiums and/or joint ventures which will help him be an active participant in a modern economy.

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2.1 Efects of this type of Trust

- Trustors commission their property to the Trustee with the purpose of receiving the best possible return.
- All sort of goods or property can be commissioned: land, rights, future cash flow, etc.
- The commissioned property forms and independent and autonomous estate.
- The Trustee operates watching the interests of the Trustors. Fiduciary operates as the owner based on the exclusive interests of the Trustees.
- Trustee is forced to comply with the purposes for which the Trust was created
- Trustee has to render checks and balances. If he does not it can be replaced.
- The property of the Trust is not at risk at any time because it is isolated from any Trustee and/or Trustor own risks.
- Participants have limited liability.

3 BUSINESS MODEL

3.1 Sustainable

- Economic and ecologic respect for the territorial zoning Territorial.
- Environmental responsibility. Ecological and conservationist management of the Amazon rain forest, its natural resources and biodiversity.
- Development of certified plantations for the purpose of having them comply with the highest international standards and better agronomic and agro-industrial

3.2 Inclusive

- Promotes transparent relationships among all agents in the
- Compatible with international standards of Corporate Social Responsibility with respect to workers rights and the needs and wished of the community and its own social laws.
- Fair benefits for all the participants of the chain or cluster.

3.3 Competitive

- Regional investment in infrastructure generate favorable conditions for businesses to flourish.
- Long term financial and economic developments enable this business to ensure its viability.
- Calls for the strategic development of a competitive biofuels cluster.
- Calls for continuous research and development commitments for the purpose of sustaining better agronomic and agro-industrial practices.

3.4 Advantages Of This Business Model

- Investor has to deal with only one agent (Trustor) and not with hundreds of Farmers.
- Trustor is regulated by the government through the Banks and Insurance Superintendence.
- Contracts are drafted “tailor made” to suit the specific needs of the Trust.
- Risks of each party do not affect the business or the Trust.
- Participants (Trustees) share of the business are predetermined.
- Open possibility to incorporate more future Trustees.
- Quick and simple decision making process is defined in the contracts. The investor can become the operator of the Trust.

4 TRUSTEES ROLES

4.1 Investor Farmer

- Manages and operates the business. Contributes with his land for 30 years expecting fixed or variable return.
- Fund the business to make the project Contributes with his land for 30 viable years expecting fixed or variable return
- Contributes its Know How, technology Offers his manpower should he and market. Qualify.
- Develops the business with international standards.
- Shares in the profits should he choose the variable income option

4.2 Trustor Regional Government

- Structures the Trust and represents Invest in road energy, communications,
- Farmers with the Investors infrastructure

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- Administers the Trust according to Contributes with its leadership its regulations and instructions.
- Contracts and supervises Operator Contributes in getting the Farmers properties legal.
- Invest in basic services in cluster territories.

5 THE CONTRACT

Structures an autonomous estate with the property included in the productive process.

Gives the Trustor the legal capacity to act directly as the manager of the productive process or contracting a third party specialized on it. The third party is usually the Investor Company.

Trustees transfer the following estate in the form of a fiduciary domain in favor of the Trustor which is usually a well established financial institution:

- The land destined to the cultivation of Jatropha.
- The Jatropha seeds, seedlings and plants.
- The infrastructure which will be installed as a consequence of the investment and further financing.
- The machinery and the equipment financed.
- The cash flow generated during the productive process over which the Farmers have rights.
- These contracts are signed and executed by the Investors, the Farmers and the Financial Institution.
- By law in Peru these contracts may have an extension of up to 30 years but can be renewed.

6 FIDUCARY MANDATES AND RIGHTS

6.1 Land Owners (Farmers)

- Transfer administrative rights to the Financial Institution for 30 years.
- Mantains the property of the land.
- Have first rights to become a productive operator.
- In case of a sale of the land acknowledges the Investor's first right of refusal.
- Has preferencial rights at the time of liquidating each season.
- Acknowledges the property of the investment and of the future production in favour of the Investor.
- Benefits of the income agreed upon as per the plan selected when signing into the Trust: fixed or variable income.

6.2 Investors

- Defines who would be the Operator of the Project. Usually it is the Investor.
- Has first right of refusal if a land owner (Trustee) wants to put his property for sale. The price of the land is established according to a formula agreed upon at the time of entering into the Program.
- In case of a sale to a third party the Investor has the right to receive the current and future value.
- Has the right to the entire production given to the refinery and/or to its commercial entity.

7 GROUND WORK

Maps of different aspects of the territory needed to be developed.

8 REQUIRED CONTRACTS

- <u>Master contract</u> (Land owners, Investors, Financial Institution, Regional Government).
- <u>Trust contract</u> (Land owners, Investors, Financial Institution).
- <u>Contract of future production sale</u> (Financial Institution or Operator and the Market).
- <u>Service contract with Operator</u> (FI + Land owners).
- <u>Contract with Operator entity</u> (FI + Operator).

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TOWARDS LIGNOCELLULOSIC ETHANOL PRODUCTION : CURRENT DEVELOPMENT AND COLLABORATION IN KHON KAEN UNIVERSITY

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ABSTRACT

Khon Kaen University has strong focus on biofuel research since 2009 despite continuing research on the topic long before that year. The reason mainly concerns with the pathway to become one of the national research universities that stimulated the university to form research clusters of which biofuel cluster is one of them. Bioethanol is one big group of the research in biofuel cluster. Various research aspects on bioethanol have been carried out. The interest in lignocellulosic ethanol production has been seen in 2 parts, which include lignocellulosic biomass conversion and glucose/xylose conversion to ethanol. Physical and chemical treatments are the main hydrolysis methods employed. Researches on discovery of cellulolytic enzymes have also been carried out. The research on lignocellulosic ethanol production focuses on the use of the wild type yeast already in the national collection, *Candida shehatae* TISTR 5843. Assessing the ability of the yeast to utilize glucose and xylose, which are the main sugars from lignocellulosic hydrolyzate, for growth and ethanol production is current concern. Attempt in genetic modification of a *Saccharomyces* strain for xylose uptake has started under collaboration with researcher in Microbiology Department.

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1 INTRODUCTION

Thai government promotion on commercial ethanol production as alternative fuel has been around since the fuel crisis. In 2003, the first fuel ethanol production plant has operated commercially. Currently, 19 ethanol plants are in operation with maximum daily capacity of 2.925 million liters. In 2011, 4 ethanol plants are due to operate and another plant in 2012. These will contribute an extra maximum daily capacity of 1.82 million liters to the current number (Department of Alternative Energy Development and Efficiency, Ministry of Energy).

The main raw materials for ethanol plants in Thailand are molasses and processed/fresh cassava. Most ethanol plants operated from 2009 have focused on using cassava as their raw materials. Many of the ethanol plants that use molasses are linked or associated with sugar industries.

The raw materials used for ethanol production in Thailand, especially cassava, are also food sources and not only used for ethanol production but also other industries such as ethanol for consumption and other biochemical productions as well. The foreseen problem of competition for raw materials is concerned in addition to food versus fuel problem considering the relatively constant production of these 2 main crops during the past 10 years (Fig 1).

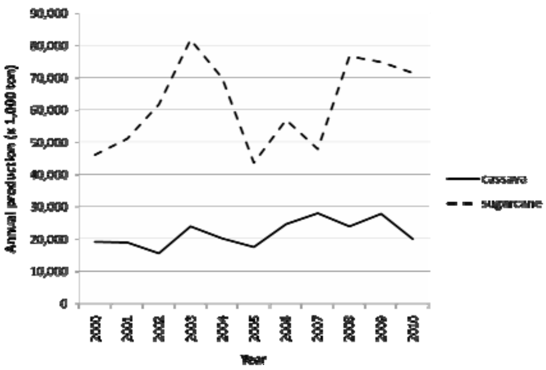

Fig 1: Cassava and sugarcane production in Thailand from year 2000–2010 (data from ASEAN Food Security Information System)

The above concerns have initiated an interest in finding alternative raw materials for ethanol production. Generally, 2 major approaches in focus are finding alternative energy plants and utilizing lignocellulosic materials. Thailand, as an agricultural–based country, has obvious opportunity in using the agricultural waste and the refuse from agro–industries such as oil palm and sugarcane industries as alternative raw materials not only for ethanol production but also in other biochemicals that employ the use of microorganisms as well.

This report includes 2 sections regarding the research on ethanol production in Khon Kaen University (KKU). The first part focuses on the university–wide perspective on ethanol production research. The second part is about the current researches leading to the lignocellulosic ethanol production process.

2 RESEARCH ACTIVITY REGARDING ETHANOL PRODUCTION IN KKU

Being a multidisciplinary university, KKU consists of faculties that could well involve in ethanol production. The major players in ethanol researches are in the Faculty of Agriculture where the researches in alternative energy plant are active, while ethanol researches in Faculty of Technology and Faculty of Science involved in the areas related to ethanol production.

The alternative plants for ethanol production undergone the studies in KKU are sweet sorghum and Jerusalem artichoke. The researches have been carried out in collaboration with researchers from Faculty of Technology since 2003 to evaluate the potential ethanol production from these plants. Exploration on various production methods using the plants is still ongoing with the aim to obtain options for production processes and to find the suitable ethanol production process.

Fig 2: Sweet sorghum (left) and roots of Jerusalem artichoke (right), the 2 alternative energy plants in focus at KKU

Another aspect of ethanol research that has been carried out in the university concerns the hydrolysis of lignocellulosic materials. Although not as the major theme on KKU ethanol research as compared to the use of alternative plants, researchers in Microbiology Department, Faculty of Science has been active in searching for cellulolytic enzymes. Physical and chemical treatments of agricultural wastes such as oil palm trunk, bagasses, corn cobs and rice straw are commonly practiced not only in ethanol research but also in biohydrogen research which is also carried out in the university.

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From 2009, biofuel have become one of the main research focuses of the university. Various research clusters were established as a part of university's pathway to national research university. Biofuel cluster was one of 6 clusters formed. Grants were awarded in 2010–2011 to 56 projects under Biofuel cluster. Almost 50% of the grants involved the works that relate to fuel ethanol business including raw materials, hemicellulosic and cellulosic hydrolysis enzymes, production process and logistics.

3 RESEARCH TOWARDS ETHANOL PRODUCTION FROM HEMICELLULOSIC FRACTION

Interest in lignocellulosic ethanol production in KKU mainly concerns 2 aspects; finding cellulolytic enzymes and using the hydrolysate for ethanol production. Conventional hydrolysis of the lingocellulosic materials would result in hydrolysate from hemicellulose and cellulose parts. In our laboratory, interest in utilizing the hemicellulosic part has been concentrated as we believe that conventional ethanol production could be employed with hydrolysate from cellulosic hydrolysate which would mainly contain glucose. The long term goal for the research is the total conversion of biomass fractions to ethanol using natural (non–genetically modified) organism/s.

The research has been using *Candida shehatae* TISTR 5843 from TISTR Culture Collection Bangkok MIRCEN as a model organism. The strain's ability in ethanol production from xylose was reported. The preliminary works has been carried out using pure glucose and xylose to assess the growth and ethanol production abilities. Initial study showed that biomass of *C. shehatae* was able to convert xylose to ethanol. Attempt to test the conversion of mixed sugars (glucose–xylose) to ethanol using the sequential conversion by *Zymomonas mobilis* and biomass of *C. shehatae* was successful (Boonmee *et.al.* 2007). The biomass of *C. shehatae* showed potential to be used repeatedly at least twice to produce ethanol from xylose but with reduced yield and deteriorated xylose uptake (Uan–yuan and Boonmee 2007).

The strain was further evaluated separately in pure glucose and xylose. *C. shehatae* TISTR 5843 has a limitation on using glucose and xylose under the static cultivation of the biomass to produce ethanol. Approximately 55–60 g/l of glucose could be uptake by the yeast. Reduced glucose uptake was evident at higher glucose concentrations than 60 g/l as compared to lower concentrations. Xylose uptake was capped at 70–80 g/l. Maximum ethanol concentrations produced from both sugars were in the range of 24-27 g/l. Superior ethanol yield per sugar utilized was observed when glucose was used. The ethanol yield values laid between 0.33–0.43 g/g when using xylose and averaging around 0.44 g/g when using glucose. Much lower ethanol productivity compared to *Saccharomyces cerevisiae* was to be considered for improvement in further studies (Pornin *et.al.* 2008).

As *C. shehatae* produces ethanol only from its biomass in static condition, some sugars from hydrolysate was presumably used in producing biomass. Further study on using alternative sources in biomass production, prior to ethanol production, was also of interest as a way to maximize the use of sugars only for ethanol production. A study on using glycerol to produce the cells of *C. shehatae* TISTR 5843 had shown that the cells produced using the alternative substrate still maintain the ability in producing ethanol. However, ethanol concentration from cells produced from glycerol was slightly lower than the cells produced from xylose, when using xylose as the substrate for ethanol. The ethanol yield from cells produced from glycerol was also lower than those produced from xylose (Promwieng *et.al.* 2009).

Apart from the work in Biotechnology Department, Faculty of Technology, collaboration with researcher from Microbiology Department, Faculty of Science in constructing the genetically modified *Saccharomyces cerevisiae* has been ongoing. Attempt to make *S. cerevisiae* to uptake xylose was done with the aim to make the yeast to produce ethanol more efficiently as *S. cerevisiae* showed more tolerant to ethanol than xylose–utilizing yeast.

More studies on the uses of mixed sugars (glucose/xylose) and lignocellulosic hydrolysate as substrates are ongoing. In the raw material hydrolysis part, repeated acid treatments and the use of ionic liquid are currently under investigation. However, the focus would be on ethanol production. Current projects involve the investigation into various cultivation techniques in order to develop the cultivation methods that maximize the use of sugars in hemicellulosic hydrolysate to product ethanol. More screening work to obtain potential isolates that has improved ability in sugar, especially xylose, uptake and ethanol production would also be carried out in parallel.

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STUDY ON WOODY WASTE PRETREATMENT FOR CELLULOSIC ETHANOL PRODUCTION

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ABSTRACT

The study on pre-treatment processes of woody wastes from pulp and paper integrated mills for enzymatic hydrolysis of cellulose was presented. Combined mechanical (grinding, milling) and chemical methods (liquid hot water (LHW), acid, alkaline) pretreatments were developed in order to achieve the complete saccharification of cellulosic component of woody biomass.

Acacia and Eucalyptus wood rejects from the chipping process of pulp production were grinded to reduce the size of biomass. Hemicellulosic component was then extracted from saw dusts by liquid hot water pretreatment. The solid products with or without delignification were pulverized by ball-milling in order to improve enzymatic digestibility. Delignification was performed by using sodium hydroxide solution. The enzymatic hydrolysis experiments showed that the conversion of cellulosic components into glucose attained 90-99 mol% under optimal conditions. The enzymatic hydrolysate and prehydrolysate were ethanol fermented by *Saccharomyces sereviciae* and *Pichia stipitis*, respectively.

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1 INTRODUCTION

Currently, Viet Nam Pulp and paper Industry and Wood Chips Exporters use annually around 4 million tons of timber. The amount of forest residues and woody wastes may go up to 800 thousand tons per year. These sources of biomass are now being used ineffectively. Due to their abundance and renewability, there was a great deal of interest in using such the biomass for the production of biofuels.

The hard wood species such as *Acacia* spp. and *Eucalyptus* spp. are main sources for pulp and paper industry in South-East Asia Region, in general, and in Viet Nam, in particular. Woody wastes from chipping process may consist of 4-5% of the timber wood and can be served for bio-ethanol production.

Cellulosic hydrolysis has been accomplished by using either acids or enzymes. In order to convert lignocellulosic biomass to fermentable sugars for ethanol production, enzymatic hydrolysis is an attractive option as it produces higher ethanol yields than those produced by acid-catalyzed hydrolysis. The development of an effective pre-treatment method for lignocellulosic biomass is the most important aspect of the enzymatic saccharification process. Pretreatment generally refers to the disruption of the naturally resistant carbohydrate-lignin shield that limits the access- ibility of enzymes to cellulose and hemicelluloses (Wyman, 2005).

Various pretretment methods have been classified in to mechanical, physical, chemical, biological and combination of these (Mousdale, 2008 and Mtui, 2009).

In the present study, the different pretreatment methods were applied to woody wastes such as grinding, planetary ball-milling, LHW, acid and alkaline treatments… It founded that combining mechanical (griding, pulverization process) and chemical (LHW, dilute sulfuric acid, sodium hydroxide) methods would achieve the complete saccharification of the total cellulose portion of the biomass (Yang and Wyman, 2007).

2 MATERIALS AND METHODS

2.1 Samples

Woody wastes of *Acacia mangium* and *Eucalyptus urophylla* used in this study were received from Viet Nam Paper Corporation. The woody wastes were air dried and stored in plastic bags before use.

It was shown that particle size greatly affected the rate of enzymatic hydrolysis. The woody wastes were cutter-milled and to pass through a 0.2 mm-size sieve flours used for the research. Obtained woody flour samples were store in plastic bags for moisture homogeneity and use in all experiments.

2.2 Chemicals

Commercial enzyme including cellulase and hemicellulase were purchased from Novozymes, Denmark. And other chemicals used in the experiments were PA grade.

2.3 Preparation of samples

Preparation of samples and composition analysis were performed according to standards of LAP (Laboratory Analytical Procedure, NREL, USA), and TAPPI (Technical Association of the Pulp and Paper Industry, USA).

2.4 Chemical pretreatment

Chemcal pretreatments using different agents were carried out in reactors at appropriate temperature for certain time with suitable reagent concentrations.

Liquid hot water pretreaments of wood flour samples were performed at 160-200°C for 10-30 minutes with liquor-to-wood weight ratio (L/S) 10-20.

Sulfuric acid solutions with concentration of 0.1-0.5% were used to pretreat the samples at 125-165°C for 5-25 min.

Alkaline pretreatment was performed using sodium solution of 1-5% concentration at 30-100°C for 1-4 h.

2.5 Ball-milling process

The ball-milled process was performed in a Planetary Ball Mill (Pulverisette 5; Fritsch, Germany). The pretreated samples were then milled at a rotational speed of 200 rpm with a cyclic mode of 10-min milling followed by a 10-min pause for a prescribed time period of 30 min up to 90 min (Teramoto Y. *et al*., 2008).

2.6 Reducing sugars content

Reducing sugar concentration was determined by the Dinitrosalicylic Acid (DNS) method, based on the colour formation of DNS reagent when heated with reducing sugars (Adney and Baker, 2008).

The intensity of colour was measured using Spectrophotometer SP–300 (Japan) at the wave length of 540 nm. The concentrations of reducing sugars in hydrolysates were calculated from the calibration curve of synthetic glucose solutions.

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2.7 Enzymatic hydrolysis

The pretreated solid products were hydrolyzed by the mixture of cellulase and hemicellulase (NS 50010, **N**S 50013, **N**S 50014, Cellic CTec and Cellic HTec; Novozymes, Denmark) in citrate buffer (pH 4.8). The enzymatic hydrolysis prepared samples were then incubated at 50±1°C with shaking.

2.8 Ethanol fermentation

Prehydrolysate and enzymatic hydrolysate have been fermented using the bacteria (*Pichia stipitis* 09, *P. stipitis* 36…) and yeasts (*Saccharomyces cerevisiae* 17, *S. cerevisiae* HN05…) from the microbial collection of Institute of Biotechnology, Viet Nam Academy of Science and Technology (VAST).

3 RESULTS AND DISCUSSION

3.1 Composition analysis

The composition of lignocellulosic materials depends on their sources. Table 1 summarizes the dry basis contents of collected woody (*Acacia mangium* and *Eucalyptus urophylla)* wastes.

Table 1. Composition of woody wastes, % odw

Component	*Acacia mangium*	*Eucalyptus urophylla*
Cellulose	36-42	40-43
Pentosans	17-23	18-22
Lignin (Klason)	24-27	24-25
Ash	1.3-1.5	0.6-0.7
Extractives in Acetone	2.6-3.5	2.3-3.0

Odw: oven dried wood

The cellulose content of woody wastes (36-43%) was high enough to serve as feedstocks for bioethanol production.

3.2 Dilute acid pretreatment

Dilute sulfuric acid-based chemical pretreatment is the most popular pretreatment method (Mosier *et al.*, 2005). This process, however, might have some undesirable effects such as the formation of furfural and hydrometylfurfural due to an excessive degradation of the produced monosaccharides. These result in a lowering the conversion yield of polysaccharides and an inhibition for the ethanol fermentation process.

Pretreatment with sulfuric acid has been the subject of research for over two decades. It was overseen that 80-90% of hemicelluloses sugars are recoverable by dilute acid technology (Wyman C. E., 2007).

At the optimum, woody flours were pretreated at 150-165°C, for 15 min with dilute sulphuric acid concentration of 0.1-0.25 w%. This results in dissolved reducing sugars of 21.5-23 % odw.

3.3 LHW pretreatment

Liquid hot water pretreatment is usually used in dissolved pulp cooking. In this process, hemicelluloses are mainly dissolved. This mild hydrolysis condition could avoid the formation of aldehydes.

LHW pretreatment was used as the first pretreatment step to extract hemicelluloses from woody flours. At the optimal conditions (temperature: 190-200°C, L/S ratio: 15 and retention time: 10-15 min) the amount of 27.5-31% of original substances were dissolved, among them 11-12.5% odw was reducing sugars.

3.4 Alkaline pretreatment

Alkaline, in particular sodium hydroxide is a good agent for lignin removal. This process can cause the fibrillation of wood chips; therefore, it makes enzymes easy of access to the fibre surfaces and improves the cellulose saccharification.

The good lignin extraction from LHW treated wood flour samples was performed by using sodium hydroxide of 5% concentration at 100°C for 1 hour. The same result could be obtained by pretreating the samples with 1% NaOH solution at 100°C for 4 hour.

3.5 Enzymatic hydrolysis

For saccharification of cellulose of pretreated woody samples, different enzymes products of Novozymes were used.

The treated solid products were, subsequently, pulverized by ball-milling in order to destroy the crystalline structure of cellulose component. This process notably improves the enzymatic digestibility.

The LHW or sulfuric acid pretreated solid products were then ball-milled by pulverized ball-milling for the period of 30 to 90 minutes (cyclic mode: 20-min milling and 10-min pausing). The results of optimal LHW-treated followed by ball-miling could significantly improve the yield of glucose. The reducing sugars in enzymatic hydrolysate attained 94-99% of cellulose content with milling time 30-90 min (Fig. 1).

The enzymatic hydrolysis of LHW followed by sodium hydroxide pretreated samples could achieve cellulose-to-glucose conversion of 81.5-82%.

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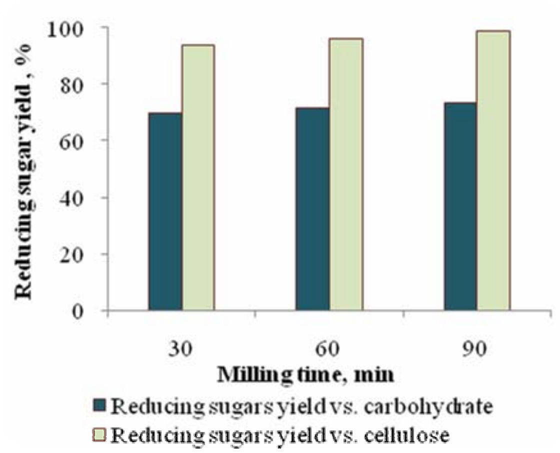

Fig 1: Cellulose-to-Glucose conversion of pretreated and ball-milled woody samples

3.6 Ethanol fermentation

The acid and LHW hydrolysate were treated for removing fermentation inhibitors (furfural, HMF, organic acids, phenolics…). The pentoses were fermented to ethanol using yeasts (*Pichia stipitis* 09, *P. stipitis* 36…).

Glucose in the enzymatic hydrolysate was fermented by the yeasts (*Saccharomyces cerevisiae* 17, *S. cerevisiae* HN05…).

The ethanol yield was around 130 mL/kg woody waste.

4 CONCLUSION

The woody wastes from Pulp and Paper Industry could be use as a feedstock for ethanol production.

Pretreatments must be advanced and carefully integrated with the rest of the process to realize the full potential of cellulosic ethanol. For example, the combination of chemical (either acid; or LHW; or LHW-alkaline) pretreatments followed by ball-milling could achieve 99% of cellulose-to-glucose conversion.

5 ACKNOWLEDGEMENTS

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CURRENT STATUS OF BIO-FUELS DEVELOPMENT IN VIET NAM

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ABSTRACT

Viet Nam is richly endowed with a relatively large amount of renewable energy (RE) resources distributed throughout the country. There are lot of biomass feedstock available for energy production. Biomass is available with agricultural products and residues at equivalent more than 10 million tons of oil per year. Biogas energy potential is approximately 9 billion m^3 per year, which can be collected from landfills, animal excrements and agricultural residues. The potential of small hydropower (<30MW) is larger than 7,000MW. Solar energy is abundant with average solar radiation at 5kWh/m^2 per day. Thus the Government of Viet Nam recognizes the importance of making sure that Viet Nam's RE resources endowment as well as bio-fuels are fully developed, and that the institutional, regulatory and financial barriers are overcome so that RE can make its proper contribution to economic development, energy security and sustainable development.

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1 INTRODUCTION

Renewable energy (RE) has long played a large role in Viet Nam and in its energy mix. About 60% of the rural population still relies on biomass fuel for cooking. Furthermore, biomass fuel is the traditional fuel for many local handicraft facilities and production of brick, porcelain, and ceramic. Agricultural residues, especially bagasse, are extensively used in cogeneration. In remote rural areas, small hydro power (household scale Pico-hydropower generators as well as community sized projects) have long provided at least some limited access to electricity.

That the share of RE having recently declined is largely a reflection of Viet Nam's dramatic transition of the past twenty years into a modern market-based economy – a transition greatly assisted by cheap global energy prices, the coal resources of the north, and the discoveries of gas in the southern region of Viet Nam.

But the era of cheap fossil fuels that has played a central role in the economic development of many countries is seen by many as ending in the very near future. Unprecedented increases in oil prices reflect the demands of the new emerging economic superpowers. Yet very shortly Viet Nam will become more exposed to international energy prices because its economic growth is expected to outpace its own domestic energy resources – the potential for large hydro will be largely exhausted in this decade, while limited supplies of gas and coal means that Viet Nam will soon need to import energy for power generation. Thus the Government of Viet Nam recognizes the importance of making sure that Viet Nam's RE resources endowment as well as bio-fuels are fully developed, and that the institutional, regulatory and financial barriers are overcome so that RE can make its proper contribution to economic development, energy security and sustainable development.

2 POLICIES AND PROGRAMME TO PROMOTE RE/BIO-FUELS

In 2001 the Government of Viet Nam launched the Renewable Energy Action Plan (REAP)[2] which set the general directions of Government intervention to encourage RE. Most of the effort (and the success) has been in the development of an institutional framework to facilitate grid-connected RE, bio-fuel projects and a comprehensive package of reforms that will be in place by the end of 2008.

The 2004 Electricity Law permits support to electricity generation from RE sources in the forms of investment incentives, preferential electricity prices and preferential taxes.

The National Energy Development Strategy issued in December 2007[3] sets specific targets for RE to reach 5% of primary commercial energy in 2020, and 11% by 2050.

The most recent policy initiative was issued in 2008,[4] which established a minimal tax on income derived from the sale of carbon offsets under the Kyoto Protocol, and, more importantly, establishes a framework for providing subsidy to certain RE technologies.

Viet Nam's RE policies are driven by the needs to supply sufficient energy for economic development and ensure environmental protection. Energy demand is expected to increase 4 times in the period 2005-2030 and electricity demand 9 times in the period 2005-2025.

The essence of RE development are going to be finalized in the documents titled "Strategy for Renewable Energy Development of Viet Nam for the period up to the year 2030, with outlook to 2050" and "Master Plan for Renewable Energy Development of Viet Nam for the period 2011 to the year 2020, with outlook to 2030" that MOIT has commissioned the Institute of Energy to prepare, and approval.

For bio-fuels, the government has targeted annual output of 100,000 tons of E5 and 50,000 tons of B5 by 2010, equivalent to 0.4% of the country's projected oil and gasoline demand; 1.8 million tons of ethanol and vegetable oil, or 5% of oil and gasoline demand by 2025 (Decision No. 177/ 2007/QD-TTg).

In order to achieve these targets, the Government has provided various incentives to investors and other actors. Incentives include subsidies, tax incentives, state investment credits, subsidized interest rates, loans guarantees, priority in land leasing and priority for loans. It is expected that these policies will create the appropriate conditions for planning and implementing RE projects as well as the sale of RE products in Viet Nam. While some of these policies have been put in place, various incentives and other support measures are still missing from the policy framework.

3 THE MAIN CHALLENGES

The main challenge for the Government of Viet Nam is how to balance the long term goal of environmental sustainability with the immediate goals of economic development. General decalrations of policy such as the granting of preferential

[2] Viet Nam-Renewable Energy Action Plan, World Bank ESMAP Technical Paper 21, March 2002.

[3] Prime Minister Decision 1855/QD-TTg, 27 December 2007

[4] Inter-Ministerial Circular 58/2008/TTLT-BTC-BTN&MT

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pricing for RE in the Electricity Law run into the practical realities of funding incremental costs.

Even if one could show that the benefits of RE the avoidance of the environmental damage costs of fossil usage, which impose real health damage costs to Viet Nam - outweigh the increased costs, passing these costs to consumers is exceptionally difficult at a time when fighting inflation is a primary objective of Government policy. If fossil fuel price cannot be raised even to meet the general requirements of a financially healthy energy/power sector, the expectation that the incremental costs of RE which can be funded by a consumer levy or from the general State budget may be unrealistic.

As a signatory to the Kyoto Protocol, Viet Nam recognises its obligations to the international community. At the same time, no reasonable definition of equity could hold that the cost of reducing Viet Nam's GHG emissions be carried by Viet Nam's consumers (or the Government of VN).

These realities shape the main criteria for designing a realistic policy framework for RE development. Priority should be given to:

- Developing those RE resources that are economic, but whose development is constrained primarily by institutional and regulatory problems.
- Maximising the revenue from sales of carbon credits to the international community.
- Developing local capabilities in those RE technologies of greatest significance for Viet Nam in the short- to medium term.

4 BIO-FUEL DEVELOPMENT

4.1 *Biomass energy*

In recent years, the development of biomass energy in Viet Nam has been driven by investments in two main areas:

- Heat and electricity production
- Improvement of efficiency and environmenttal effects of biomass use.

In biomass-based electricity production**,** some of the typical projects in Viet Nam are bagasse-based co-generation (bagasse-fired cogeneration) of around 38 sugar companies; rice husks - based co-generation (1 rice husk fired power plant) and rice husks - based electricity generation (8 rice husk fired power plant under F/S report)

4.2 Bio-fuels

There are 3 types of feedstock for bio-fuel production:

- Edible energy crops such as corn, soy bean, cassava, sugar cane, sweet sorghum, etc. (first generation)
- Non-food energy crops such as jatropha, buffalo grass, algae, etc. (second generation)
- Industrial, and agricultural wastes such as animal fats, etc. (second generation).

The main feedstock for bio-fuel in Viet Nam in the period up to 2015 with outlook to 2025 were identified as: (1) used fats and oils including used oils collected from food industries, (2) algae, considering its low demand of land and ecological conditions and short development cycle, (3) agricultural crops such as bagasse, cereal, sesame, peanut, coconut, and basa fish fat, (4) jatropha which can be grown on around 9 million hectares of bare land or land strips along the national highways.

Since the deployment of the Governmental Decision No. 177 on a "Scheme for Development of Bio-fuel up to 2015 with a vision to 2025", investments in bio-fuel research and production have increased. Research is focused on bio-fuel technologies, application in electricity generation and transportation use. Unlike the situation during 1990s when researches were still spread on a broad scope and mainly focused on lab or field research, which did not link to the market, research and development of bio-fuel in recent years have been oriented towards applying international and regional technological advancement into Viet Nam's conditions.

Pilot and commercial production has been increasing in the last 5 years in which 2009 can be considered as the year of the bio-fuel industry kick-off in Viet Nam. During 2009 many ethanol plants were built in Quang Nam, Phu Tho, Quang Ngai, Binh Phuoc and Dong Nai. It is planned that by 2011, there will be 6 ethanol producers with a total installed capacity of 365,000 tons per year-enough for mixing 7.3 million tons of E5.

In the next two decades, development of bio-fuel as a partial replacement to fossil fuels is an attractive option, particularly for oil-importing countries. Bio-fuel technologies are not relatively complex and can utilize local feedstock. When mixed with fossil fuels, there is no need to change the mechanics of vehicles. Bio-fuel also is competitive on sales price compared to gasoline and diesel. Consequently, the international market for bio-fuel will create the opportunities for the sector to grow in Viet Nam by taking advantage of the tropical climatic conditions of the country and the agricultural base of its economy.

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4.3 Biogas

In Viet Nam, the climatic conditions are favourrable for biogas production. Biogas is produced mainly from agricultural residues, livestock and urban wastes. Bio-digesters made from bricks, concrete and plastic have been tested and applied throughout Viet Nam. Small scaled biogas plants are considered as the most complete technologies.

Attempts have been made to research and pilot testing use of biogas to replace gasoline and diesel in vehicles, motors and electricity generators. These efforts have been scattered and at small scale in size. In rural areas, farmers with more than 15 pigs can build biogas plants with capacity of 15-20 m^3 and use the gas for electricity generation.

In addition to the benefits of a more stable energy supply and environmental protection, biogas technologies provide several other benefits. The residue "bio-slurry" used as fertilizers helps increase agricultural yields and reduces the use of chemical fertilizers and pesticides.

Biogas projects in Viet Nam have been financed by the state budget, international development institutions, private investments from industries and households. At the household level, the biggest biogas project in Viet Nam is The "Biogas Program for the animal husbandry sector of some provinces in Viet Nam", 2003-2011: This program received assistance from the Netherlands Government to the Livestock Department. In the first phase (2003-2006). The program reached 12 provinces and helped to build 18,000 plants with a total cost of US$ 17.7 million. In the second phase (2006-2011) the program has targeted developing 150,000 biogas plants in 35 provinces and cities, providing 800,000 people with improved energy service. This program promotes the fixed dome biogas plants built underground from bricks and cement that last at least 15 years.

5 CONCLUSIONS

In addition to the purely economic arguments based on the increasing cost of fossil fuels, renewable energy projects have other attributes that make them desirable: i). Small renewable energy projects are generally located in remote rural areas, and are more likely to catalyse rural development and generate rural employment opportunities than thermal projects. Construction activities in such remote areas often require development of local roads that improve access of rural communities; ii). Renewable energy displaces thermal generation, and thereby reduces environmental damage costs from coal, gas and oil-based projects. While the magnitude of these impacts is uncertain (particularly when compared to other emission sources, notably urban transportation), the human health consequences of fossil fuel emissions represent a real economic cost to the economy; iii). Many renewable energy projects will qualify for the sale of carbon credits to the international community, representing an additional transfer of resources to Viet Nam.

However, the main challenge for Viet Nam is how to balance the long term goal of environmental sustainability with the immediate goals of economic development with a question need to be answered - who will pay for incremental costs of RE?

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CHALLENGES AND OPPORTUNITIES OF THE PRODUCTION OF BIOFUELS IN MEXICO

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ABSTRACT

In this document one appears a general view the activities developed in the last years in relation to the production of biofuels in Mexico. Starting off from the present situation of the power ones, where the exhaustion of the hydrocarbon reserves, the increasing demand of fuels for the transport, the stagnation in the growth of the refinement infrastructure, the preoccupation by the mitigation of greenhouse effect gases, and mainly the results of the projects developed in the world for the production of biofuels, gave rise to the creation of the legal frame in the matter in our country, that has as well given to rise to programs and projects derived from the Law of Promotion of the Bioenergetics. This law focuses to the production of bioetanol and biodiesel from crops that do not compete with the basic food production like the maize and the sugar cane. Supporting the development of projects sustained in non food crops for the generation of second and third generation biofuels. For it official documents of the environment secretariats were used, energy and agriculture among others. The studies and projects that have been realized starting off of the guidelines by the Law of Bioenergetics, in which it is orchestrated with the participation of the Mexican Government through the secretariats of energy, environment, agriculture, economy and property, as well as of universities, research centers, private companies and crop producers, are in the right direction, through the programs of introduction of the bioenergetics, climatic change, of renewable energies and energy efficiency. Making lack greater clarity and coordination between this great set of actors of these programs and projects, that without a doubt will give the results waited by Mexican authorities, without being free of corrections and adjustments in the goals and reaches in the time. Combined to the previous thing, Mexico owns earth and the will to producer biofuels of second and third generation that allows mitigate the environmental impact, the use and operation of their fossil fuels in benefit his habitants and the APEC region.

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1 INTRODUCTION

Mexico is a country located to the north of the American continent with a surface near the 2 million square kilometers and one population of 112,3 million inhabitants in agreement with the census of population realized in 2010. Traditionally it is considered as a energy generator country and a net importer of raw materials like maize and oily.

The Mexican power sector is constituted by the Secretariat of Energy (SENER), the public companies of electricity (CFE) and petroleum (PEMEX), and the research centers of petroleum (IMP), nuclear power (ININ) and electricity (IIE).

The total consumption of energy in our country in the last years (2008) by type of power is dominated by gasoline with 33,4%, is followed by the petroleum diesel engine with the 16,9%, the electricity with the 13,8%, solid fuels like coal, wood, bagasse of sugar cane and petroleum coke with the 12,2%, the dry gas with the 9,8%, gas LP with the 9,4% kerosene with the 2,7% and the fuel oil with the 1,9%. The petroleum production has decreased of a maximum value of 3,6 million barrels per day reached in 2004, to a production of 2,4 million barrels per day in 2008. On the other hand it has existed a growth of the 15,7 % and the 53,0% in the production and gasoline consumption respectively, as a result of the growth of the vehicle number and the stagnation of the refinement capacity. On the other hand the petroleum reserves go to the loss with the exhaustion of the well-known oil zone like "Sonda de Campeche", that during two decades has provided the greater production from petroleum to our country. Of year 2001 to year you reserve them to 2009 proven reserves of 23.500 million barrels of petroleum to 14.300 million barrels, which implies an average life smaller to 10 years.

The greenhouse gas effect in Mexico have been increased 40,2% of 1990 to year 2006 reaching a value of 715 MtCO2e, contributing 33% of the emissions the energy use, 27% the energy generation, 18% agriculture and the use of ground change, 14% the residues, and 8% the industrial processes, reason why the sector energy significantly contributes with 60% of the emissions, considering the sum of its generation and consumption. In the same way in sequence of contribution, the change of ground use and agriculture that it includes the cattle activity would represent along with the sector energy 78% of GEI emissions. Of not being applied measured of mitigation, the tendency it indicates that 882 MtCO2e in 2020 and of 1.089 will be reached values of MtCO2e in 2050.

In the agricultural sector, Mexico has a surface of for sowing of weather of 20 million hectares, and of irrigation of 8,5 million hectares. The maize, original of Mexico, takes place in all the agricultural regions of the country, represents the main food of Mexican people with an average consumption of 500g by person per day.

In 2005, the internal demand of maize was of 18,6 M ton for human consumption, 4,6 M ton for cattle consumption and 3,7 Mton for industrial consumption. However, Mexico is the third import country with 6,8 M ton, after Japan and the Republic of Korea that concern 16,6 and 8,5 maize M ton, respectively.

In Mexico, approximately 7 M hectares are seeded and 20 M ton of maize are produced. If the yield would be increased to 5 or 6 ton/ha, would duplicate the production without increasing the seeded surface, the challenge is to carry out it in the reality, since this us would make self-sufficient from the nourishing point of view and would exist surpluses to produce bioethanol, saving gasoline imports, reducing the dependency of petroleum and mitigating the emissions that contribute to the climatic change.

The sugar cane is another important culture in our country, that sowing in a surface of 750 thousand hectares for an annual production of 50 million tons of sugar cane and one production of 5 million tons of sugar. In present conditions this crop, is focused to the sugar production, thus its use is restricted for the production of bioethanol, mainly by the price of guarantee that it has in our country equivalent to the double or more of the international price, that increases the production cost of bioethanol making its production nonviable.

The other source of first generation biofuels, is the cultures of the oily crops like soya, sunflower, palm oil, among others. In this subject, Mexico also is deficit since of the 5,5 M of tons of vegetable oil that consumes annually, it only produces of the order of a half of million tons, consequently it is seen in the necessity to import the main part of this consumption.

In the last years it has been impelled on the part of the government, the production of oily like soya, canola and sunflower, as well as the oil production from the palm, in the south-east region of the country, with the National Oily Production Project 2007-2012, of the SAGARPA (Agriculture Ministry). Nevertheless in a market of net import, it is little less than impossible, to propose the production of biofuels from these crops.

2 PROMOSION LAW AND DEVELOPMENT OF THE BIOFUELS

In February 1ST of year 2008, was published Law of Promotion and Development of Biofuels, that it establishes like objective to promote and to develop the bioenergetics with the purpose to help the diversification of fuels and the sustainable

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development conditions that allow to guarantee the support to the Mexican field, and establishes the bases which they promote the production of consumptions for Biofuels, with base in viability criteria and considering driving this program in zones of high and very high marginality, from farming and forest activities, with biotechnological and enzymatic production of seaweed in the Mexican field, without putting in risk the nourishing security of the country.

The Law establishes that Energy Ministry (SENER), Environment Ministry (SEMARNAT), Agriculture Ministry (SAGARPA), Property Ministry (SHCP) and Economy Ministry (SE) of the Mexican Government, will form a commission that joint with universities, research centers, local governments and companies, will formulate and carry out the National Strategy of Energy through projects derived from the programs orchestrated by the different sectors, like the Program of Sustainable Production of Biofuels Resources and Scientific and Technological Development 2008-2012. The Special Program of Climate Change (PECC) orchestrated by the SEMARNAT, as well as the Special Program of Renewable Energies (PEER) through the SENER. In some aspects these programs have common projects that own the attributes of energy, environment and agriculture.

3 PROGRAM OF SUSTAINABLE PRODUCTION BIOFUELS RESOURCES AND SCIENTIFIC AND TECHNOLOGICAL DEVELOPMENT

The Program of Sustainable Production of Biofuel Resources and Scientific and Technological Development 2008-2012, articulates within the framework with the Program of Introduction of Biofuels of the Inter-ministry Strategy of the Biofuels of the SAGARPA and with the consensus of the members of the chain; which will have by objective to foment the viable production of biofuels resources and their commercialization, being given certainty, increasing the competitiveness and major yield to the Mexican field by means of the scientific and technological development.

The Program of Introduction of Biofuels, leaves from the diagnosis of which in Mexico the conditions for the production of biofuels exist and that the contribution of the biomass to the national energy balance in 2006 of the 3,4% was contributed by the firewood and the sugar cane bagasse, also it indicates that it is possible in period 2008 to 2012, to produce bioethanol to replace 2% in gasoline for the three main cities of Mexico, replacing MTBE (Methyl Therbuthyl Ether) at the moment used, in agreement with studies realized by the SENER. Reason why the central axis of this program is based on initially producing 200 million annual liters for the city of Guadalajara, and continuing with 150 million liters for the city of Monterrey and 530 million liters for the city of Mexico, reaching in 2012, the 880 million liters for the three cities, from additional surpluses crops as sugar cane, beet, yucca and sorghum. The participation and disposition of PEMEX the oil public company in charge to produce and to commercialize fuels in Mexico is fundamental, since in agreement with the Law the introduction of bioethanol to replace the MTBE is its attribution.

4 SPECIAL PROGRAM OF CLIMATE CHANGE (PECC)

The PECC arises from international and voluntary commitments of Mexico to reduce 50,65 $MtCO_2$ in year 2012, emissions in the energy generation sector (18,03 $MtCO_2$), use of energy (11,87 $MtCO_2$), agriculture and change of land use (15,29 $MtCO_2$), and wastes (5,46 $MtCO_2$).

The reductions in the energy sector will be obtained in projects like self supplying (3,68 $MtCO_2$), co-generation (0,9 $MtCO_2$) and improves in operative efficiency in oil industry (1,24 $MtCO_2$), saving of energy in electric home appliances (2,68 $MtCO_2$), wind generation (1,2 MtCO2), projects of hydrogenation (1,91 $MtCO_2$) and reinjection of bitter gas in oil wells (6,9 $MtCO_2$). In which it refers to mitigation in the sector transports are expected reductions of 0,9 $MtCO_2$, by clean fuels and 1.1$MtCO_2$ by retirement of circulation of old cars, 1,6 $MtCO_2$ by modernization of railway system, 1,2 $MtCO_2$ by reduction of distances by means of road construction. It is important to stand out that by means of the program of efficient stoves in rural means they will be reduced 1,62 $MtCO_2$ by the introduction of 600 thousand stoves to year 2012.

5 SPECIAL PROGRAM OF RENEWABLE ENERGIES (PEER)

The PEER additionally looks for the beginning of the energy transition in our country, in the context of a greater energy demand and fuels, and a diminution of the petroleum reserves and fuel production. The goals of this program 2008-2012 are based on impelling the development of the industry of renewable energies in Mexico, starting off of a energy contribution of the 3,3 % in 2008, to a contribution of the 7,3% in the national energy balance in the 2012; distributed in 4.3% of wind energy, 0,77% of minihydraulics, 1,65% of solar energy and 0,85% in biomass and biogas. It excels of this program the generation with energy of the wind to interconnect 2.473 MW from 2009 to 2012.

The SAGARPA in last three years through Trust of Shared Risk (FIRCO), it has destined

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funds for the installation of 305 biodigesters in equal number of farms. With this action, they have been stopped emitting to the atmosphere more than 1,3 million tons of carbon dioxide and 4,460 GWh are generated, and so the demand of the own productive units is covered.

6 CROP PRODUCTION OF BIOFUEL RESOURCES

Within the frame of the Law of Promotion and Development of the Biofuels, it have been destined public and deprived resources for the production of second and third generation biofuels that do not put in risk the food production, such as the culture of the Jatropha curcas, residues of harvest with high ligno-cellulose content and the use of microorganisms modified like the microalgaes.

6.1 *Jatropha curcas*

The Jatropha curcas also known as pinion is original of Mexico and Central America, Is a non-evergreen shrub that belongs to the Euphorbiaceae family. The fruits are elliptical capsules, yellow color with 2 to 3 seeds by fruit. It is developed well in the tropical and semitropical in grounds poor and sandy climate regions in altitudes that go of the level of the sea until the 1.600 meters over sea level. One adapts to poor grounds of low fertility and owns the capacity to recover grounds eroded by the great amount of organic matter that produces (Henning, 1998). It has of two to three periods of flowering and fruition is in the months of July December. It has a yield average of oil of 1.590 Kg/ha-year, exist toxic and nontoxic species in our country; the content of dry matter of the seed is greater of 95%, the content average of protein is of 25% and fat of 55%. In the first year it produces 1.100 kg of seed and 440 kg of oil by hectare. It exist a potential surface for his culture in our country among 2,5M and 7M of hectares. The project more hard is in the State of Chiapas with a worked surface of 10 thousand hectares that hopes to reach the present year with a surface of 16 thousand hectares. The biodiesel produced in this project was used in commercial flight of an Airbus A320-214, equipped with motors CFM56-5B4/3. The airplane took off of the International Airport of Mexico City to the International Airport of Tuxtla Gutiérrez, Chiapas. For it, the flight was realized with a mixture of 27% of Biodiesel and 73% of fossil fuel. The biodiesel only was in one of the motors, whereas the other filled of power the conventional one.

6.2 *Castor Oil (Higuerilla)*

Its area of adaptation is very extensive: From 0 to 1.800 meters over sea level it tolerates temperatures of 41ºC, but an excessive heat during the flowering affects the relation of flowers male/female. The densities of sowing vary from 11,2 to 15,7 kg/ha with populations between 20.000 and 40.000 plants/ha. It is not developed in grounds karsts. A neutral pH is adapted. The cycles of production are between the 120 and 150 days. In Mexico, the INIFAP of SAGARPA has estimated “productive potential half of the castor-oil plant in areas with no irrigation” in 6.3 million hectares. The average yields in different countries vary between 500 and 1.600 kg/ha. The seed can be stored maintaining a humidity of 10%. When being finished the agronomic process, the following step is the extraction of the oil and its transformation. It is important to mention that the seeds have a toxin that is neutralized during the process of extraction of the oil, without contaminating. Yield average of seed: 1.1 t/ha (0.5-1.6 t/ha) Contained of oil in the seed: 40-55% Yield of extraction oil: 90% Yield: 460 kg of oil/ha, and cycles of 4-5 months.

6.3 Palm Oil

In Mexico the plantations of greater productivity are in the region of “Soconusco” in the south-east of the country, since plantations exist, the majority without irrigation, with yields average in the region of 19,5 ton by there are and 4,3 ton of oil. In the last years the palm oil consumption has been increased exponentially, since in the year 2000, Mexico concerned 50 thousand tons and the past year a volume of import was reached a half of million tons. When defining the Governing Plan of the production of Oil Palm in our country in 2004, 16.830 hectares of palm in the state of Chiapas were had, today are more than 40 thousands, they are hoped to arrive at 100 thousands in the 2012 and a potential of more calculates than 900 thousand hectares in all Chiapas. Considering the main producing states in the country of palm of oil like Veracruz, Campeche, Tabasco and Chiapas, a potential of the culture is in 6 million hectares is estimated.

6.4 *Lignocelulosic Materials*

One hopes that of a production of bioethanol from lignin of 3 billions of gallons (11,3 billions of liters) in 2006, it is increased to more than 25 billions of gallons (94,6 billions of liters) in 2020. In Mexico investigation is in progress, since in the specific case of the sugar cane the possibility exists of producing additionally to bioethanol that it is possible to be produced by fermentation of the juice, of the order of 125 liters of bioethanol by ton of sugar cane, without putting in risk the sugar production. One has focused this work in the construction of microorganisms for the fermentation of lignin, celluloses and hemi-celluloses to bioethanol, combining the characteristics of microorganisms like the Escherichia coli and the Saccharomyces among others for such intention. The lignin sources are varied like the sugar cane, harvest left-over, wood and forest residues, among others many.

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6.5 *Microalgaes*

The Mexican group Biofields owns an exclusive patent of the company Algenol Biofuels, allows work with blue green seaweed to produce ethanol from the solar radiation, salt water, nutrients and carbon dioxide. The process that follows the company to create this biofuel of third generation absorbs a great amount of CO_2. The first project of BioFields in Mexico is called Sonora Fields and it is located in Puerto Libertad Sonora at the northwest of Mexico. For this project 22 thousand hectares of nonpro- ductive earth were bought. BioFields glides to produce 250 million gallons (946 million liters) of the fuel for year 2013. It is a project that has many benefits and viability to generate bioethanol in significant quantities, that they diminish the depen- dency of petroleum, nevertheless, the possibility of escape of organisms modified to the environment, worries to the public opinion, by the possible impacts on natural seaweed and the ecological balance.

7 CONCLUSIONS

Mexico is a country that is in stage of development of its biofuels, in a context of environmental an energy needs as the greenhouse effect, where one of their more important components is the emissions derived from fossil fuels for the transport. From the energy point of view without consider the economic one, the import of these fuels is increasing due to the growth of the vehicles number, the stagnation of the petroleum refinement and today diminution of petroleum reserve. Simultaneously, the resources like the maize and the sugar cane to produce biofuels of first generation (bioethanol and biodiesel), compete with food, reason why they have been contemplated in the Law of the Biofuels and they will only be used when there are surpluses of these cultures, situation that can happen more by through the increase of the crop yields than by the extension of culture surfaces. Giving great margin for the production of biofuels of second and third generation, as the conversion of residues of harvests and the use of microalgaes, and cultures little known in our country like jatropha curcas and the castor-oil plant. The studies and projects that have been realised starting off of the guidelines marked by the Law of Biofuels, in which it is orchestrated with the participation of the Mexican Government through the ministries of energy, environment, agriculture, economy and property, as well as of universities, research centers, private companies and specially field producers, are in the right direction, through the programs of introduction of the biofuels, climate change, of renewable energies and energy efficiency. Making lack greater clarity and coordination between this great set of actors of these programs and projects, that without a doubt will give the results waited for by the Mexican authorities, without being free of corrections and adjustments in the goals and reaches in the time. Combined to the previous thing, Mexico owns earth and the energy to produce biofuels of second and third generation, that allow to mitigate the environmental impact, the use and the operation of their fossil fuels in benefit of their inhabitants and APEC region.

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CURRENT STATUS AND PERSPECTIVES OF BIOFUELS IN MEXICO

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1 REGULATORY AND LEGAL FRAMEWORK

The legal framework for biofuels and bioenergy started to take shape in year 2008. Two laws were approved by the Parliament, and their corresponding regulatory Decrees followed some months later. The Parliament also approved budget allotments to several programs, steadily increasing to reach about 170 MUSD by 2011.

1.1 Law of Promotion and Development of Biofuels, regulatory Decree.

This Law encourages the establishment and development of national Biofuels companies, including the social sector (farmers associations and cooperatives).

It also aims to develop a modern and competitive rural sector, able to produce and market efficiently raw materials and biofuels. Furthermore, it promotes the education, training and technological and scientific research on Biofuels, and make provisions to attract investment in the production of raw materials and Biofuels

The regulation Decree, established an Inter-secretarial Commission (formed by four Ministries) to act as implementing body. It also required that Official Mexican Norms have to be issued for all aspects regarding production, distribution, marketing and environmental safety of biofuels. It states that maize can only be used to produce ethanol in case of surplus production, which is a very much unlikely event.

Since 2009, the Parliament approved in each fiscal year specific budget lines for biofuels promotion, reaching 100 MUSD by 2011. This funding was mainly applied to: a feedstocks R&D program, issuing grants to improved irrigation technologies, giving financial support to biogas production from manure in big farms.

1.2 Law for the Better Use of Renewable Energy and the Financing of Energetic Transition

This law addresses specifically the electricity production and end uses. It promotes and encourages the use of renewable sources of electricity, energy efficiency and energy saving measures and technologies and the diversification of primary energy sources. Moreover, it created a national fund for these purposes within the federal government budget. About 70 MUSD were allotted in fiscal year 2011, and applied to several programs to foster energy savings (subsidies to efficient refrigerators, replacement of light bulbs, improving energy efficiency in public buildings). Finally it grants a program for the same purposes, focused on industrial energy consumers.

1.3 Bioenergy policy

Two programs have been issued: a liquids biofuels program and a R&D program.

The Secretary of Energy launched in 2008 a program for the substitution of MTBE by ethanol in gasoline, aiming to replace 6% in volume by 2014 in most of the country. However, due to the low prices set in the public biddings, no supply contracts were signed, and the Mexican state-owned petroleum company PEMEX has not yet started to mix ethanol in gasoline.

The research and development program operates through annual calls for projects under the national research agency CONACyT . Over 400 projects were approved and funded between 2008-2010. Most of them deal with feedstocks production technology (first generation) and basic science. Less than 9% aims to develop / transfer technologies.

2 CURRENTS STATUS

2.1 Sources of Energy

In Mexican primary energy matrix, Oil Products account for 64%, Natural Gas 10% and Electricity 14%. Biomass share is 7% (5% from firewood plus 2% bagasse).In 2008, overall primary energy used was 4,815 PJ.

Mexico exports about one third of its oil production, but this is steadily declining and according to most forecasts, the country will become a net importer by 2015. Most of the coal and one fifth of the natural gas are currently imported.

2.2 Research and Development on bioenergy

From 2008 to 2010, governmental funding for R&D in biofuels added up to 28 MUSD, supporting above 400 projects. Most of them were small-scale (20–50 thousand USD budget) and short-term (1-2 years). The majority focus on basic science and production technologies of feedstocks for 1st generation liquid biofuels. Only a few were involved in technology transfer, economic assessment and environmental impacts of biofuels. Just two cases focused on Life Cycle Assessment (LCA) of GHG emissions and energy balances. Less than ten research projects dealt with solid biomass sources, its technologies and end uses.

2.3 Biofuels Projects

Several projects have been proposed for biofuel production, and a few have started the implementation phase, either at pilot or demonstration scale. Small farmers established Jatropha plantations with little success in five thousand hectares, while big companies in about

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one thousand hectares achieved slightly better results. Three small companies produce Biodiesel from used vegetable oils and fats. One corn-ethanol plant was built but is not currently in operation.

Above 300 biodigesters have been installed, mostly in pig farms, however a large number of them burns the biogas without energy recovery at all. Solid bioufels have received very little attention.

One big project was announced for the production of biodiesel from algae, and started operations as a pilot plant in 2010 in Sonora State.

2.4 Sate of the art

Solid Biofuels such as firewood and charcoal are a traditional source of energy for cooking and for small industries (bricks, tiles, pottery, mezcal, brown sugar) in Mexico. Some 20 Mt (DM) /year of firewood are used in rural households and small rural enterprises. Charcoal is mostly used in cities for cooking; some 0.6 Mt/yr are produced with traditional technologies (earth mounds) from native forests, from which a small fraction are under proper forest management.

Industrialized forest fuels as wood chips and wood pellets are seldom used in Mexico, in spite of the huge potential and actual availability of forest residues.

The production of liquid biofuels is currently at an incipient stage: Ethanol is produced but not used for fuel, while Biodiesel is produced from residual fats and oils in small scale and currently used in experimental/demonstrative projects. Bioturbosine (jet fuel mixed with 37% of modified biodiesel from recycled fats and oils) was successfully used in one demonstration flight in May 2011.

Biogas from landfills is beginning to be captured. In one big landfill (Monterrey) it is successfully used for power generation in a 11 MW dual fuel station (diesel / biogas). Biogas from manure digesters is mostly burnt without energy recovery, because the tariff paid for electricity by rural users is heavily subsidized and thus the net return on investment for power generation is low.

3 PERSPECTIVES OF BIOFUELS AND BIOENERGY

The future development of biofuels and bioenergy in Mexico depends on a combination of drivers, which may change over time:

- Supply and domestic prices of oil-derived fuels, which are presently abundant and cheap but are more scarce and costly in a near future
- Stronger policies to foster the production and use of biofuels and biomass for energy
- Availability of mature and cost-effective technologies for biofuels production plus the investment capital needed
- Compromise to attend both the needs of food and fuels

3.1 Potential supply of biomass for energy

Mexican potentials for biomass production are big. At least 2,800 PJ/year can be sustainably obtained from native forests, crop and agro-industrial residues, and urban waste. New dedicated crops could add 720 PJ/year in the mid-term. Altogether, biomass energy could supply up to 70% of the primary energy used in 2008, 15% coming from new dedicated crops and plantations.

Source	Biofuel	Mt/year	PJ/año	Mha
Forest and jungle management	firewood, charcoal	101	1,515	
Agro-industrial waste	bagasse	29	431	
Forest plantations	firewood, charcoal	23	345	*2.924*
Sugarcane	etanol	206	**338**	*2.953*
RACs	celulósicos	15	227	
Grain Sorghum	etanol	10	**202**	*2.656*
Oil Palm	biodiesel	13	**121**	*1.860*
Agro-industrial waste	cellulosic	8	114	
Dedicated Crops	various	6	86	
Forest residues	lignocellulosic	3	63	
Jatropha curcas	biodiesel	3	**57**	*3.173*
Livestock manure	Biogas	35	35	
Municipal waste	Biogas	n.a.	35	
TOTAL			3,569	

3.2 Dedicated energy crops

The area with good aptitude for new, dedicated crops and forest plantations is 13.7 M ha, or about 50% over the presently cropped area (24 M ha). Most of these lands are presently used for pasture, and could be replaced by annual or perennial crops without sensible reductions in food production, also avoiding deforestation. However, replacing pastures by crops will certainly decrease carbon stocks in soils, generating strong CO_2 emissions, unless proper technologies ensuring conservation of organic matter in the soil are applied. In any case, the overall potential contribution of dedicated crops for 1st generation biofuels is much lesser than the other accessible sources.

3.3 Crops and agro-industrial residues

Residues from crops and agro-industrial operations (including forestry and forest industries)

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are by far, the biggest potential source of biomass in Mexico, with a sustainable output around 2,350 PJ/yr. Moreover, neither capital investment nor waiting time is needed to tap these sources that are presently available.

Although cost effective commercial technologies for conversion of lingo-cellulosic materials into bioethanol and/or biodiesel are not yet available, there is a variety of alternatives for its use to replace fossil fuels in industrial applications and electricity generation with current technologies.

3.4 Technology gaps and challenges

When considering 1st generation liquid biofuels, there are a few technology gaps, mainly related to the development of cropping systems that can at the same time produce raw materials for food and fuel, or food co-products along with fuel products. These systems should be energetic efficient and generate low GHG emissions, implying the use organic fertilizers and avoiding the use of fossil fuels in the industrial phase. Otherwise, the carbon mitigation targets already set for biofuels will not be reached. The challenge is to produce biofuels without impairing food supply and at the same time to avoid the use of fossil energy. These constraints do not apply to solid fuels obtained from residues, thus opening wide opportunities for their development in the short term.

For 2nd generation biofuels, the main challenge is to develop commercially viable technologies for the conversion of lingo-cellulosic material into liquid fuels. This depends to a certain extent on the market prices for fossil fuels, but mainly on the technologies by themselves, which must be energy efficient and environmentally acceptable.

3.5 Fuel versus food

A great debate exists on this issue all around the world. In Mexico, the law forbids the use of corn (the national staple human food) for fuels. But Mexico imports about one half of the food it needs; food security is by no means granted. A great national challenge is how to feed the people and fuel the economy at the same time.

First generation biofuels offer little hope to achieve both goals. In the best case, the expansion of dedicated crops for biofuels may not make things worst, but certainly will not make them better in Mexico. Only new multipurpose crops producing fuel, food and fodder will help. Non-toxic Jatropha varieties may be a partial solution, as well as grain sorghum that can produce ethanol and DDGs. Sugarcane for ethanol has not these capabilities. New and promising crops are being studied, but research and development of a new culture may take many years.

4 REMBIO ACTIVITIES

The activities of REMBIO spread out in several directions: research, communication and dissemination, capacity building. REMBIO works as a network that includes institutions and people involved in all these fields.

4.1 Assessment of biomass potentials

Starting from overall studies on biomass potentials at country level, REMBIO is refining these assessments and is currently conducting case studies at local and regional scale. By using GIS technology, the assessment of potentials can be geographically referred and applied to more detailed feasibility studies. Several publications ca be found at www.rembio.org.

4.2 Case studies and projects development

Case studies are a powerful tool for better understanding the complexities of biomass utilization for energy, which only are evident at detailed levels of analysis. REMBIO has performed several of these studies (ethanol from grain sorghum and sugarcane, cogeneration in sawmills, impacts of fuel crops on soil and water use and conservation).

4.3 Life Cycle Analysis

To fully understand the environmental, energy and economic results of biofuels production systems, a full Life Cycle Analysis has to be made. It is also a mandatory requirement to export biofuels or feedstocks to the European Union, in order to prove that substantial mitigation of GHG emissions is achieved and no harmful effects to the environment are due to the biofuels production chain. REMBIO has performed the LCA of several 1st generation biofuels in Mexico.

4.4 Building capacities

Strengthening national capacities, both at institutional and personnel levels, is a requisite for the advancement of biofuels and bioenergy. REMBIO has a Training Program providing short courses on several issues: efficient stoves for rural households, LCA, biofuel technologies, biofuels projects assessment.

4.5 International cooperation

In the last five years, REMBIO has cooperated in different ways with international agencies like the World Bank, the International Energy Agency, and FAO. It is also a partner in national biofuels initiatives involving GIZ and the Secretaries of Environment, Energy, and Agriculture of the Mexican Government.

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5 CONCLUSIONS

The Mexican context and the potentials for bioenergy and biofuels are promising. Mexico needs to advance in the energy transition, pointing to a new energy model based on renewable energy, where bioenergy is a key element for the sustainable supply of energy in the future.

To fully and safely develop Mexican bioenergy potentials, it is needed that:
Liquid biofuels strategies, technologies and policies move from 1st to 2nd generation

- Biofuel technologies achieve better energy balance and significant mitigation of GHG emissions
- New feedstock options be developed, moving from sugars / starches / fats to lingo-cellulosic materials / waste / wood
- New targets be set for technologies development; multipurpose production systems with high energy efficiency and effective GHG mitigation that may produce simultaneously food, fodder, fibers and fuels.

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ASSESSING THE BIOFUEL POTENTIAL OF PHILIPPINE MICROALGA ISOLATES

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ABSTRACT

Rising prices and the negative environmental impacts of fossil fuels serve as the driving force for tapping alternative fuel sources. First-generation source of biofuels compete for arable land used for food production. There is a need for biofuel sources that have high photosynthetic efficiency, ability to grow in non-arable lands, and the potential to produce carbon-neutral fuels currently being met by second-generation biofuel sources like the microalgae.

Four species of microalgae namely *Chlorella vulgaris*, *Chlorella sorokiniana, Spirulina platensis* and *Chlorococcum humicola* were studied for potential for lipid production. Growth curves including growth rates and generation time of each species was studied. The effects of various treatments of nitrate (0.0015 g/l, 0.015 g/l, 0.15 g/l and 1.5 g/l as control) for the first three species were assessed with respect to lipid production. For *Spirulina platensis* nitrate treatments were the following .25g/l, 0.025g/l, 0.025 g/l, and 2.5 g/l as control.

There had been variations on the growth rates and generations time of each species. *C. vulgaris*, *C. sorokiniana, S. platensis* and *Chlorococcum humicola* showed growth rates (per day) respectively 0.06, .180, 0.20 and 0.140 with doubling times (days) of 5.270, 1.672, 1.504 and 2.144 respectively.

Growth rate and generations time during the logarithmic and stationary phase of growth were determined. *C. vulgaris* and *C. sorokiniana* produced significant amounts of lipids in 0.0015 g/l treatment during the logarithmic phase while *Chlorococcum humicola* produced significant amounts of lipids at 0 concentrations during both growth phases. *C. sorokiniana* though had higher lipids at 0.015 g/l during the stationary phase. *S. platensis* on the other hand produced 25% lipids in a nitrate concentration of 0.0025.

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There is a potential for microalgae to produce biofuels. Cost effective technologies such as mass production, harvesting and lipid extraction must be developed to support the findings of this research.

1 INTRODUCTION

1.1 Background of the Study

Rising prices and the negative environmental impacts of fossil fuels serve as the driving force for tapping alternative fuel sources that are renewable and capable of capturing carbon dioxide from the atmos- phere. However, first-generation source of biofuels such as corn, soy, sugar cane, and palm are impractical because of their potential to compete for arable land used for food production. In addition, cultivating land for these biofuel sources increases the carbon dioxide emissions through the clearing of natural forests that are effective carbon sinks (Dismukes, *et al*., 2008). Hence, there is a need for biofuel sources that have high photosynthetic efficiency, ability to grow in non-arable lands, and the potential to produce carbon-neutral fuels. These are currently being met by second-generation biofuel sources, one of which is microalgae (Schenk, *et al*., 2008).

The Philippines has a diverse collection of microalgae. However, there are only few studies con- ducted to identify which species are best used for biofuel production. In addition, the culture conditions that maximizes the fuel produced from algae has yet to be determined.

1.2 Statement of the Problem

The viability of microalgae as an alternative fuel source depends on two factors: (1) their biomass productivity which affects the volume of fuel produced and; (2) their potential to produce significant amounts of lipid which affects the concentration of the fuel produced (Liu, Wang, and Zhou, 2008, Courchesne, 2009, Pruvost and Van Vooren. These two factors must be maximized in order to fully tap the potential of microalgae as an alternative fuel source.

These factors vary depending on the species of the microalgae used, as well as environmental and culture conditions (Gerashchenko *et al*., 2002). Thus it is important to determine the following: (1) the specific algal species that meets the viability requirements and; (2) the specific culture conditions that optimize growth and lipid production of the algal species.

1.3 Objectives of the Study

- To screen potential microalgal isolates for lipid production
- Determine the effects of varying nitrogen concentrations on the growth and lipid production of *Chlorella vulgaris*, *Chlorella sorokiniana Spirulina platensis* and *Chlorococcum humicola*
- Determine the effects of varying nitrogen concentrations on the fatty acid composition of *C. vulgaris* (ADMU and UPLB strains), *C. humicola* and *S. platensis*

1.4 Significance of the Study

The Philippines being considered a megadiverse country in the aspect of biodiversity should be taken advantage of in the potential of micro algal species for biofuel production. Our country is highly dependent on the Middle East and other oil producing countries for fossil fuels. With the soaring price of oil and concerns on its limited supply, including their significant contribution to greenhouses gases, alternative sources of fuel that are renewable at the same time capable of capturing carbon dioxide in the atmosphere have to be explored. Considering the biology and physiology of the microalgae, it has been shown that these organisms have the potential as alternative source of biofuel.

2 METHODOLOGY

2.1 Growth Curve Monitoring

A defined volume of the microalgae in the logarithmic phase (9-day old) was diluted to 1L with BG-11 such that the initial culture density will be 3.0 x 10^6 cells/mL. The growth curve monitoring started immediately after inoculation until the stationary phase. Optical density and cell counts were used to monitor algal growth.

Wavelength determination for optical density measurements were done by scanning the microalgae using a Shimadzu UV-1601 UV-Vis spectrophotometer from 300 to 700 nm. The optical density was then measured by reading the absorbance using the determined wavelength. Cell count, expressed as the number of cells/mL, was also performed using the Neubauer haemocytometer. Cell count and optical density were correlated to obtain a regression equation (Quin, 2005). This was used to estimate the number of cells from the absorbance readings in the succeeding experiments. The optical density and cell counts were plotted against the number of days to determine the growth curve of the algae.

2.2 Manipulation of the Growth Medium

The standard BG-11 solution has several sources of nitrogen: ferric ammonium citrate,

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cobalt nitrate ($Co(NO_3)_2$), and sodium nitrate ($NaNO_3$). Because this experiment focused on the effects of N as NO_3, ferric ammonium citrate and cobalt nitrate were replaced with equimolar salts of iron citrate and cobalt chloride ($CoCl_2$) respect-tively, to remove other sources of nitrogen in the medium other than $NaNO_3$. Aside from this modification, the formulation for other components of the BG-11 medium was followed. 1.5g/L of $NaNO_3$ served as the control.

The method used by Piorreck, Baasch, and Pohl (1984) were adopted wherein only the initial concentration of nitrogen was manipulated. Only one form of nitrogen source was varied: nitrate (NO_3^-) from the salt sodium nitrate ($NaNO_3$). Slight modifications from the concentration levels used by Piorreck and Pohl (1984), the concentrations of sodium nitrate were used: 0.15, 0.015, and 0.0015 grams per liter (g/L). The growth rate was monitored daily while the lipid content was monitored twice for the entire duration of the set-up: one for the logarithmic phase and one for the stationary phase.

For the analysis of fatty acid composition, the following concentration levels of sodium nitrate in BG-ll were used: 1.5, 0.75, 0.075, 0.0075 and 0 grams per liter (g/L) while other sources of nitrogen were replaced, ferric citrate for ferric ammonium, and cobalt chloride for cobalt nitrate. In Zarrouk the concentration levels: 2.5, 1.25, 0.125, 0.0125 and 0 grams per liter (g/L) were used. In both media all other components followed the original formulation.

2.3 Growth Rate Determination

The growth for this study is expressed as the optical density which was obtained by scanning the algae at λ_{max} using a Shimadzu UV-1601 UV-Vis spectro- photometer. The measurement was done daily until the termination of the set-ups. The abiotic parameter (N) was then be correlated to the optical density.

Growth is also expressed in terms of the growth rate (K) and average generation time (G). These were calculated using the optical density measurements. Growth Rate (K) is equal to:

$$K = \frac{[(\log OD_t - \log OD_0) \times 3.322]}{T}$$

Where:

- OD_t is the terminal optical density
- OD_0 is the initial optical density
- T is the time in days.

Average generation time (G) was calculated using the equation:

$$G = \frac{0.301}{K}$$

Where:

- G is the average generation time
- K is the proliferation rate (Quin, 2005).

2.4 Quantification and Characterization of Lipids

Two lipid harvestings were done for the entire duration of the set-up: (1) in the logarithmic phase, and (2) in the stationary phase.

Lipids were quantified gravimetrically. The gravi- metric determination of lipid content slightly modified the procedure described by Lee, Yoon, and Oh (1989). Cells were first be harvested by pelletizing the algal cells in the centrifuge at 5000 revolutions per minute (rpm). The pelletized algal cells were then dried at 60°C for 24 hours. This was weighed and recorded as dry weight. The algal cells were crushed with a mortar and pestle and sonicated with 20mL of chloroform /methanol (2:1 v/v) for 1 hour. It was then filtered and transferred to a separatory funnel.

The lipids were recovered in the lower chloroform phase washed 20mL of 5% (w/v) NaCl. The solution were evaporated to dryness, and weighed gravime- trically, and recorded as total lipid. The crude lipid extract were then sent to the Philippine Institute for Pure and Applied Chemistry (PIPAC) for fatty acid characterization.

The total lipid content of the algae relative to its dry weight was obtained using the equation below:

$$\%\ lipid = \frac{total\ lipid}{dry\ weight} \times 100$$

2.5 Quantification and Characterization of Fatty Acids

Dried pelletized algal cells were sent to the National Chemistry Instrumentation Center (NCIC) at Ateneo de Manila University. Total lipid was determined after drying of chloroform/methanol extract under nitrogen gas and vacuum drying. Fatty acids were quantified using gas chromatography-flame ionization detection (GC-FID) and gas chromatography-mass spectroscopy (GC-MS) after derivation using the Boron Triflouride Method.

3 RESULTS AND DISCUSSION

The growth curves of three microalgae namely, *Chlorella vulgaris, Chlorella sorokiniana, Chloro cocum humicola* and *Spirulina platensis* were studied and compared. Growth was monitored daily through cell counts and absorbance readings. Regression analysis showed that cell counts and optical density have a strong positive correlation.

Chlorococum humicola when inoculated into the BG 11 medium showed fast adjustment that lag phase was not significant and commenced into the

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logarithmic phase which lasted for approximately 19 days. Plateau stage was observed for five days after which death of the culture commenced as shown in Fig. 1. Growth rate per day (Table 1) is 0.140 while doubling time is 2.144. *Chorella vulgaris* on the other hand had two days of lag phase before it entered the logarithmic phase which was maintained up to the 30th day. After which, the culture of *C. vulgaris* went into the plateau stage and then into the death phase (Fig. 2). Growth rate per day for *C. vulgaris* (Table 1) is 0. 06 while doubling time is 5.270. *Chlorella sorokiniana*'s growth curve is shown in Fig. 3 with 8 days of lag phase. The plateau stage commenced by the 20th day, after 11 days of fast growth. Growth rate per day for *C. sorokiniana* (Table 1) is 0. 18 while doubling time is 1.672. *Spirulina platensis* (Fig. 4) showed a lag phase of three days, then logarithmic phase up to the 12th day and plateau stage commenced. Growth rate of *S. platensis* is 0.200 as shown in Table 1 and doubling time is 1.504.

3.1 Growth Curves

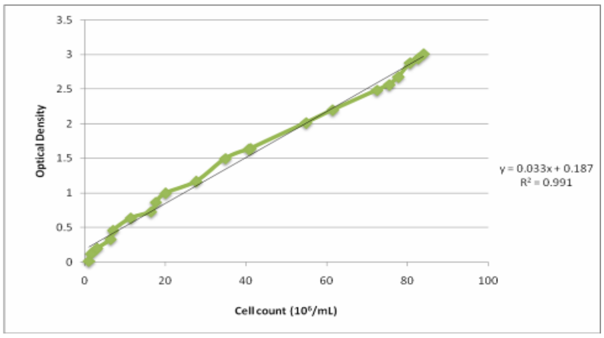

Fig 1: Growth curve of *C. humicola*

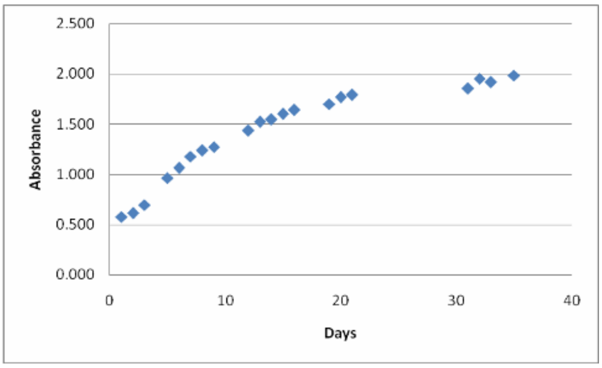

Fig 2: Growth curve of *C. vulgaris*

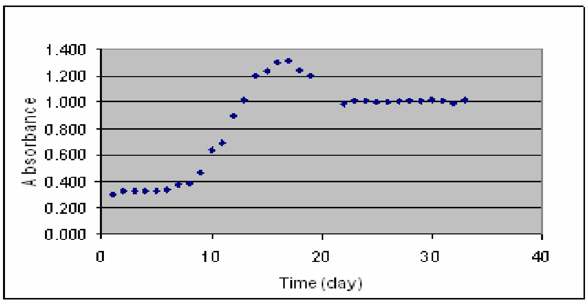

Fig 3: Growth curve of *C. sorokiniana*

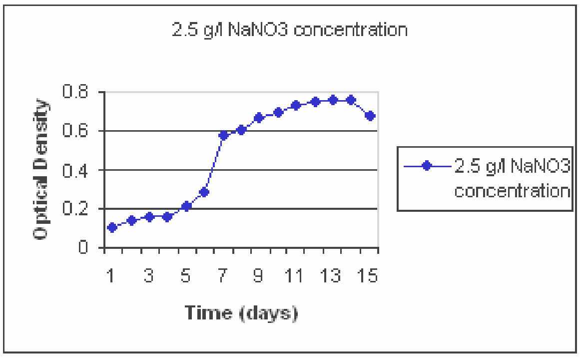

Fig 4: Growth curve of *S. platensis*

Table 1: The growth rate and doubling time of four microalgal species

Microalgae	Growth Rate (day^{-1})	Doubling Time (days)
Chlorella vulgaris	0.060	5.270
Chlorococcum humicola	0.140	2.144
Chlorella sorokiniana	0.180	1.672
Spirulina platensis	0.200	1.504

3.2 Effects of Nitrogen on Growth and Lipid Production: Chlorella vulgaris

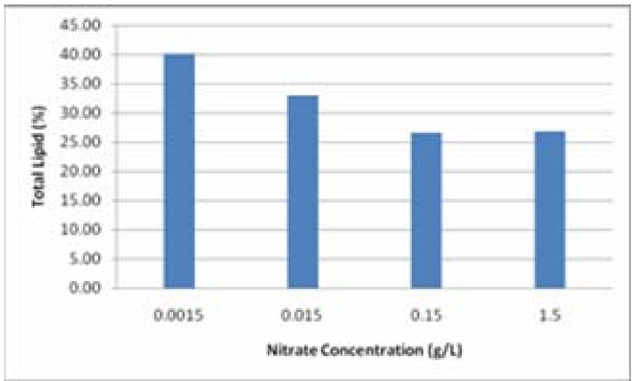

Fig 5: Lipid content (% lipid relative to dry weight) of *C. vulgaris* in the logarithmic phase.

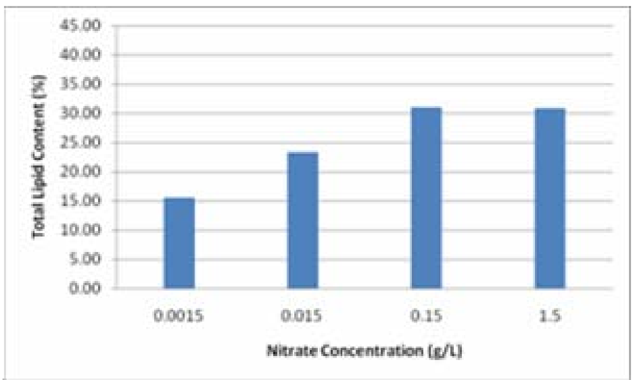

Fig 6: Lipid content (% lipid relative to dry weight) of *C. vulgaris* in the stationary phase

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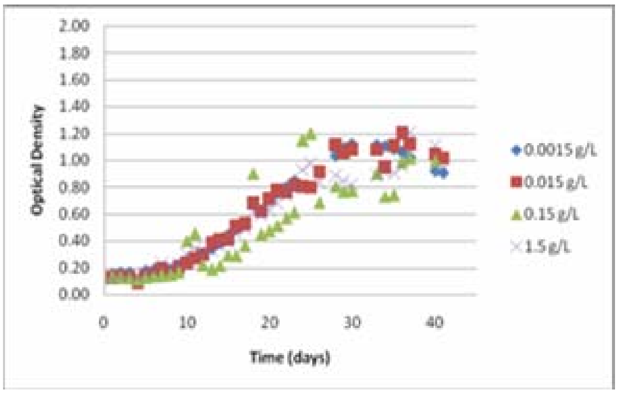

Fig 7: The growth curve of *C. vulgaris* in different levels of Nitrate in the growth media.

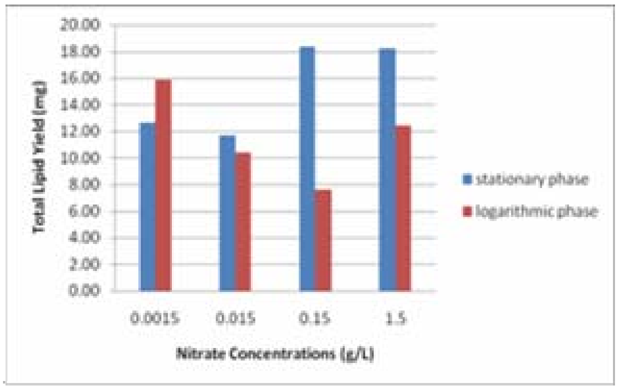

Fig 8: The comparison of oil yields (mg) in two growth phases of *C. vulgaris.*

Table 2: The growth rate and generation time of *C. vulgaris* in different nitrate levels.

Nitrate Concentration	Growth Rate (K)	Generation Time (G)
0.0015 g/L	0.05	6.02
0.015 g/L	0.06	4.64
0.15 g/L	0.06	5.13
1.5 g/L	0.06	5.22

The effect of exposure to different levels of nitrate concentration (0.0015 g/l, 0.015 g/l, 0.15 g/l and 1.5 g/l as control) on the growth rate and generation time of *C. vulgaris* was studied including the percentage lipid content in both the logarithmic and stationary phase as shown in Fig. 5 and Fig. 6. The growth rate of *C. vulgaris* exposed to 0.015 g/l, 0.15 g/l, and 1.5 g/l was 0.06 in contrast to a concentration of 0.0015 g/l which is 0.05 (Fig. 7). Generation time of this microalgae exposed to 0.0015 g/l was 6.02 as shown in Table 2. Lipid content of *C. vulgaris* exposed to 0.0015 g/l had the highest lipid content compared to the control during the logarithmic phase (Fig. 8). The stationary phase yielded lower lipid production compared to the logarithmic phase although the highest yield was found in the control and the 0.015 g/l of the nitrate concentration. Statistical tests show that growth and lipid production in different levels of nitrogen are not significantly different ($p < 0.05$).

3.3 Effects of Nitrogen on Growth and Lipid Production: Chlorella sorokiniana

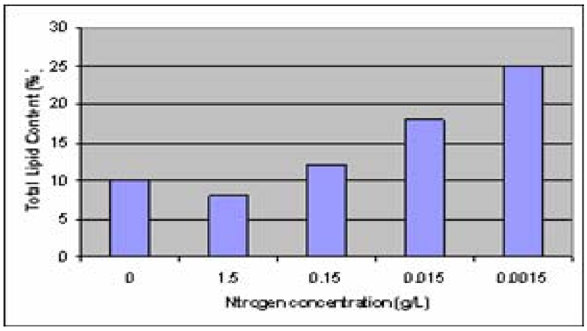

Fig 9: Lipid content (% lipid relative to dry weight) of *C. sorokiniana* in the logarithmic phase

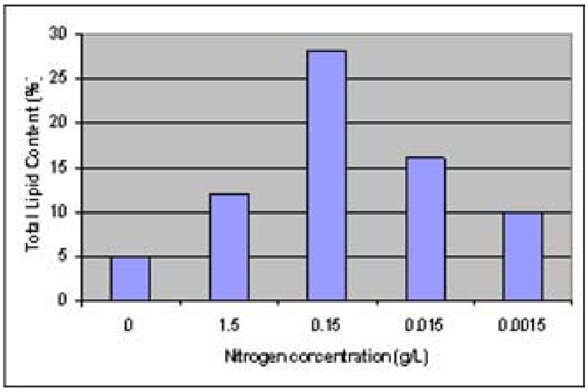

Fig 10: Lipid content (% lipid relative to dry weight) of *C. sorokiniana* in the stationary phase

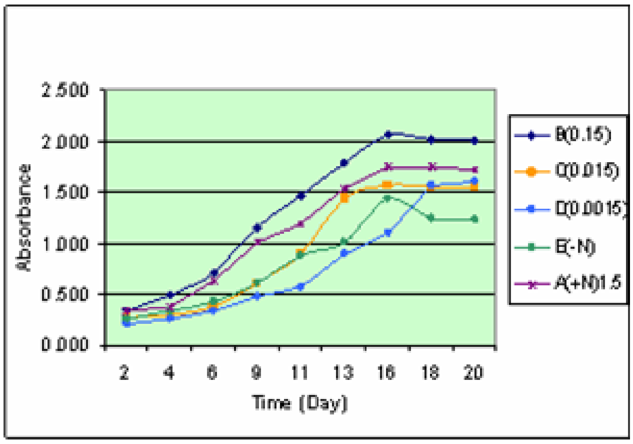

Fig 11: The growth curve of *C. sorokiniana* in different levels of Nitrate in the growth media. Statistical tests show that growth and lipid production in different levels of nitrogen are not significantly different ($p<0.05$).

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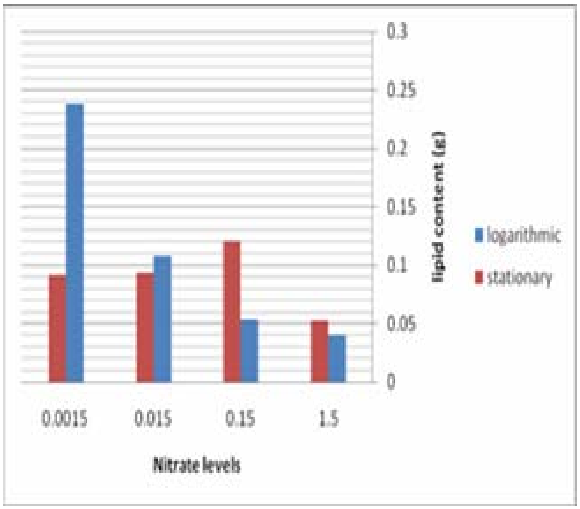

Fig 12: The comparison of oil yields (mg) in two growth phases of *C. sorokiniana*

Table 3: The growth rate and generation time of *C. sorokiniana* in different nitrate levels

Nitrate Concentration	Growth Rate (K)	Generation Time (G)
0.0015 g/L	0.122	2.47
0.015 g/L	0.041	7.37
0.15 g/L	0.182	1.65
1.5 g/L	0.180	1.67

The effect of exposure to different levels of nitrate concentration (0.0015 g/l, 0.015 g/l, 0.15 g/l and 1.5 g/l as control) on the growth rate and generation time of *C. sorokiniana* was studied including the percentage lipid content in both the logarithmic and stationary phase as shown in Figs. 9-11. The growth rate of *C. sorokiniana* exposed to 0.15 g/l is 0.182 and the lowest was observed in 0.015 g/l which is 0.041 as shown in Table 3. Generation time of this microalgae exposed to 0.015 g/l was 7.37 (Table 3). Lipid content of *C. sorokiniana* exposed to 0.0015 g/l had the highest lipid content compared to the control during the logarithmic phase (Fig. 12). The stationary phase yielded lower lipid production compared to the logarithmic phase although the highest yield was found in the control and the 0.15 g/l of the nitrate concentration. Statistical tests show that growth and lipid production in different levels of nitrogen are not significantly different ($p < 0.05$).

Table 4: The growth rate and generation time of *C. humicola* in different nitrate levels

Nitrate Concentration	Growth Rate (K)	Generation Time (G)
0.00 g/L	0.137	2.19
0.0015 g/L	0.135	2.19
0.15 g/L	0.135	2.23
1.5 g/L	0.140	2.14

3.4 *Effects of Nitrogen on Growth and Lipid Production: Chlorococcum humicola*

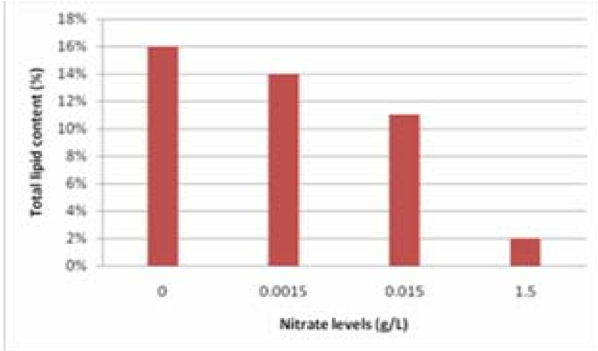

Fig 13: Lipid content (% lipid relative to dry weight) of *C. humicola* in the logarithmic phase.

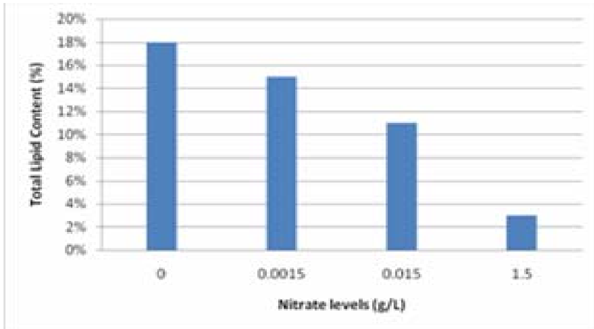

Fig 14: Lipid content (% lipid relative to dry weight) of *C. humicola* in the stationary phase

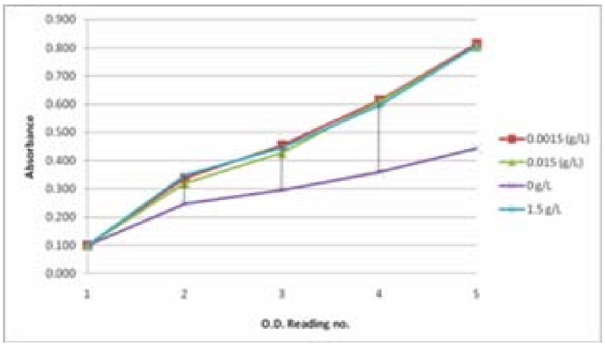

Fig 15: The growth curve of *C. humicola* in different levels of Nitrate in the growth media

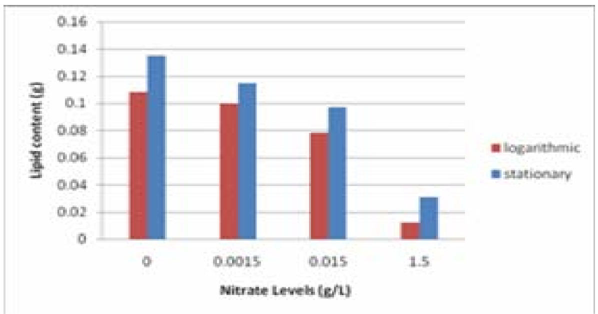

Fig 16: The comparison of oil yields (mg) in two growth phases of *C. humicola*

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The effect of exposure to different levels of nitrate concentration (0.0015 g/l, 0.015 g/l, 0.15 g/l and 1.5 g/l as control) on the growth rate and generation time of *C. humicola* was studied including the percentage lipid content in both the logarithmic and stationary phases as shown in Figs. 13-15. The growth rate of *C. humicola* exposed to the different concentrations of nitrate ranged from 0.135 to 0.140 shown in Table 4. Genera- tion time of this microalgae exposed to the different concentrations of nitrate ranged from 2.14 to 2.23 (Table 4). Lipid content of *C. humicola* was higher in the stationary phase compared to the logarithmic phase (Fig. 16). The absence of nitrate in the medium in both the logarithmic and stationary phases enhanced the production of lipids. The control (1.5g/l) yielded the lowest lipid in both two growth phases while the 0.0015 nitrate concentration produced the higher lipids. Statistical tests show that growth and lipid production in different levels of nitrogen are not significantly different ($p < 0.05$).

3.4 Effects of Nitrogen on Growth and Lipid Production: *Spirulina. platensis*

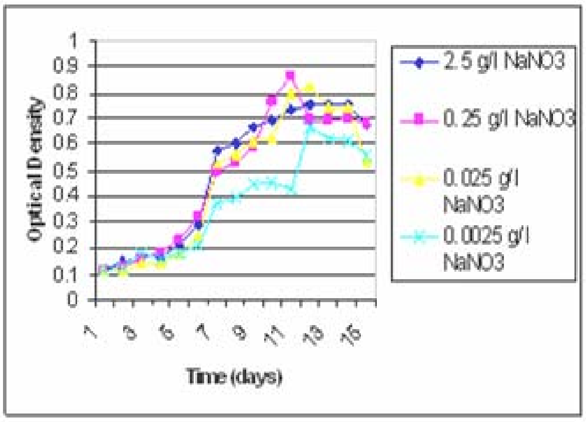

Fig. 17: The growth curve of *S. platensis* in different levels of Nitrate in the growth media.

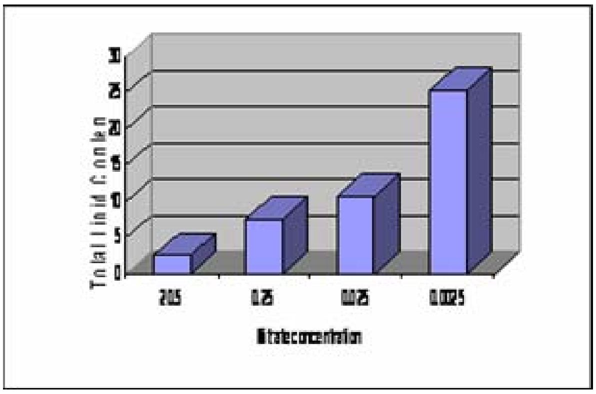

Fig. 18: The oil yield (mg) in the stationary phase of *S. platensis*

Table 4: The growth rate and generation time of *Spirulina platensis* in different nitrate levels

Nitrate Concentration	Growth Rate (K)	Generation Time (G)
0.0025 g/L	0.217	1.39
0.025 g/L	0.221	1.36
0.25 g/L	0.242	1.24
2.5 g/L	0.231	1.30

Spirulina platensis was grown in Zarrouk's medium while the rest of the microalgae were inoculated in BG 11. The amount of nitrate in the control is 2.5 g/l and the treatments for the nitrate levels were 0.25g/l, 0.025g/l, 0.025 g/l (Fig. 17). The growth rate for the various treatments ranged from 0.217 to 0.242 while generation time ranged from 1.24 to 1.39 (Table 4). As shown in Fig. 18, there was increasing trend in the lipid production of *S. platensis* when the nitrate concentration is lowered from 0.25g/l to 0.025 g/l.

3.5 Effect of Nitrogen on the Fatty Acid Profile of C. vulgaris (ADMU and UPLB), C. humicola and S. platensis in both logarithmic and stationary phases

The effect of different concentrations of nitrate (0ml, 0.05ml, 0.5ml, 5ml and 10ml) on the fatty acid composition of *Chlorella vulgaris* ADMU strain was studied in the logarithmic stage. There were 21 identifiable fatty acids. The smallest amount of fatty acid produced was 0.033167 %weight of tridecanoic acid in the 5ml NaNO-$_3$concentration while the largest amount of fatty acid produced was 22.58938% weight of 9,12-octadecadienoic acid in the 0ml $NaNO_3$ concen- tration. Of the unidentified fatty acids, the 0.05ml $NaNO_3$ concentration produced the most 16.109 29% weight unknown fatty acids.

The effect of different concentrations of nitrate (0ml, 0.05ml, 0.5ml, 5ml and 10ml) on the fatty acid composition of *Chlorella vulgaris* ADMU strain was studied in the stationary stage. It has 15 identifiable fatty acids. The least amount of fatty acid produced was 0.088299% weight of tetradecanoic acid in the 10ml $NaNO_3$ concen- tration while the largest amount was 29.00478% weight of 9-octadecenoic acid produced in the 0.05ml $NaNO_3$ concentration. The 0ml $NaNO_3$ concentration produced the most 5.6317% weight unknown fatty acids.

The effect of different concentrations of nitrate (0ml, 0.05ml, 0.5ml, 5ml and 10ml) on the fatty acid composition of *Chlorella vulgaris* UPLB strain was studied in the logarithmic stage. It listed 15 identifiable fatty acids. The smallest amount being 0.013875 % weight of dodecanoic acid produced in the 0.5 ml $NaNO_3$ concentration while the largest amount was 19.99099258% weight of 9-

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octadecenoic acid produced in the 5ml $NaNO_3$ concentration. The greatest amount of unknown fatty acids 22.2812954% weight was found in the 5ml $NaNO_3$ concentration as well.

The effect of different concentrations of nitrate (0ml, 0.05ml, 0.5ml, 5ml and 10ml) on the fatty acid composition of *Chlorella vulgaris* UPLB strain was studied in the stationary stage. There were 13 fatty acids identified. The least amount produced was 0.0855391% weight of dodecanoic acid in the 0ml $NaNO_3$ concentration while the biggest was 72.2104 5688% weight of 2,3-dimethyl-undec-1-en-3-ol in the 0.5ml $NaNO_3$ concentration. The largest amount of unknown fatty acids was 165.658408% weight in the 10ml $NaNO_3$ concentration.

The effect of different concentrations of nitrate (0ml, 0.05ml, 0.5ml, 5ml and 10ml) on the fatty acid composition of *Chlorococcum humicola* was studied in the logarithmic stage. It listed 22 identifiable fatty acids, the smallest amount, 0.010994% weight of 6,9,12,15-docosatetraenoicacid, was produced in the 0ml $NaNO_3$ concentration while the largest amount 149.3107 004% weight of dodecanoic acid was made in the 0.05ml $NaNO_3$ concentration. The greatest amount of unknown fatty acids 86.35064336% weight was produced in the 0.05ml $NaNO_3$ concentration.

The effect of different concentrations of nitrate (0ml, 0.05ml, 0.5ml, 5ml and 10ml) on the fatty acid composition of *Chlorococcum humicola* was studied in the stationary stage. There were 20 identifiable fatty acids. The least amount produced was 0.055394967% weight of 7-hexadecenoic acid in the 0,5ml $NaNO_3$ concentration while the largest was 99.930168% weight of hexadecanoic acid in the 0ml $NaNO_3$ concentration. The biggest amount of unknown fatty acids 278.49202% weight was also produced in the same concentration.

The effect of different concentrations of nitrate (0g, 0.0125g, 0.125g, 1.25g and 2.5g) on the fatty acid composition of *Spirulina platensis* was studied in the logarithmic stage. Thirteen fatty acids were identified. The least amount of fatty acid produced was 0.15661787% weight of 7-hexadecenoic acid in the 2.5g $NaNO_3$ concentration. The greatest amount produced was 49.075793% weight of hexadecanoic acid in the 0g $NaNO_3$ concentration; this treatment was also the only one to produce palmitic (hexadecanoic) acid. The biggest amount of unknown fatty acids was 84.90756571% weight produced in the 1.25g $NaNO_3$ concentration.

The effect of different concentrations of nitrate (0g, 0.0125g, 0.125g, 1.25g and 2.5g) on the fatty acid composition of *Spirulina platensis* was studied in the stationary stage. Eighteen fatty acids were identified. The smallest amount of identifiable fatty acid was 0.001989951% weight of 9-hexadecenoic acid produced in the 0.0125g $NaNO_3$ concentration while the largest amount was 19.85219657% weight of hexadecatrienoic acid in the 2.5g $NaNO_3$ concentration. The largest amount of unknown fatty acids, 164.6206242% weight, was produced in the 0.125g $NaNO_3$ concentration.

According to Mata *et al*. (2009) and Deng *et al*. (2009) palmitolleic acid, linoleic acid, oleic acid, linolenic acid, stearic acid and palmitic acid are the main components in microalgal oil with the saturated fatty acids being present in less quantities than the unsaturated fatty acids. Only in *C. vulgaris* ADMU logarithmic phase were all the common fatty acids found. In *C. humicola* stationary phase, no trace of stearic acid and palmitic acid was found but in no other species or stage was such a clear dominance of unsaturated fatty acids found. For *C. vulgaris* UPLB, the fatty acids with the highest concentrations were in fact the saturated ones: stearic acid in stationary phase and palmitic in the logarithmic stage.

The ADMU strain, in both the logarithmic and the stationary stages, has a greater number of identifiable fatty acids (12 and 14 to 9 and 7 respectively). Yet the UPLB strain has produced greater amounts of those fatty acids, in either its logarithmic or stationary phase.

4 CONCLUSIONS AND RECOMMENDATIONS

The responses of the various species were studied under nitrogen starvation and corresponding amounts of lipids were analyzed in both the logarithmic and stationary phases. Based on this work, *Chlorella vulgaris* has been identified as the species that can produce lipids to support the biofuel program in the country.

It is recommended that the effects of other abiotic parameters such as iron (Fe), phosphorous (P), potassium (K) and carbon, on the growth and lipid production of the microalgae be examined both separately and in conjunction with nitrogen (N) in order to determine the best possible combination for the production of biodiesel. The use of red and blue light emitting diodes (LED) can be utilized with 12:12 light dark cycle at ambient temperature. Cost effective media should be considered and outdoor cultures can be set up. The effects of scaling-up the experimental setups should be tested as well, as well as that of outdoor conditions, in order to determine the best methods of dealing with any detriments that come up.

The work should not only consider the potential of the freshwater microalgae for biofuel production but also take into consideration the role of these species to support food security through

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aquaculture. These species should also be studied for pharmaceutical purposes.

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BIOFUELS DEVELOPMENT TRENDS IN THE PHILIPPINES

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ABSTRACT

The abundance of biomass wastes in the Philippines is not matched by utilization; consumption of this biofuel type is mainly on a household basis as fuel for cooking and at best for heating and drying purposes in small- to medium-scale industries. About 8,000 barangays (villages) in the Philippines are still unenergized and off-grid renewable energy such as biofuels can fill the needs of these rural communities. Historically, the reported biomass utilization having the largest percentage of use has been those of sugar mills primarily in their bagasse boilers. The country's other sources of biomass fuels include rice hulls, corn cobs, coconuts (shell, husk and coir dust), livestock manure, and urban wastes. The use of biomass resources will save the Philippines about 20 million barrels of fuel oil equivalent per year. Philippine demand for biofuels is expected to rise to 187 million gallons of ethanol and 54 gallons of biodiesel this year (2011). The potential for biomass utilization is great and factors for its effective use are being worked out. The Philippine government has now put in place the framework to support biofuel industry that is backed up by legal and institutional structures. However, there still issues that beset the Philippine biofuel industry that are currently being addressed by concerned sectors.

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1 INTRODUCTION

The Philippines is primarily an agricultural country with the agricultural sector contributing about one-third of the gross domestic product. The total land area is about 30 million hectares with about one-third classified as agricultural lands. The remaining portions are shrublands, wetlands and forestlands. Rice, corn, coconut and sugar cane are the most common crops that are planted to about 9 million hectares. Agricultural biomass resources are therefore abundant from the widespread agricultural activities throughout the archipelago.

The biomass resources include rice hull, coconut shell, coconut husk, coconut coir dust, sugarcane bagasse, forestry residues, livestock manure and domestic wastes. The use of biomass resources will save the Philippines about 20 million barrels of fuel oil equivalent per year.[2]

2 BIOMASS RESOURCES IN THE PHILIPPINES

2.1 Rice Hull

Rice is the staple crop of the country and the Filipinos are among the world's biggest rice consumers with a per capita consumption of 100 kilograms per year. Rice hull estimates based from production data uses a conservative value of 20% recovery, that is, for every ton of rice produced, about 200 kg of rice hull are generated. Rice hull is the by-product of rice milling and accounts for about 20% of the rice production. The country has more than 12,000 rice mills spread throughout the regions. These processing centers are the point sources of rice hulls. The utilization of rice hull wastes in the country is very minimal. There were no reported programs to utilize these wastes on a large-scale application. Burning of wastes by farmers is still the most common means of disposal followed by dumping of wastes in open field or private spaces.

2.2 Livestock Manure

The livestock and poultry industries in the Philippines continue to grow to address the demand for animal protein to support the increasing population. The estimated amount of wastes from the animal population alone was more than 30 million metric tons per year. The waste treatment processes in the country have been minimal and continue to pose serious problem of environmental pollution. The change is the pattern of livestock and poultry production from small to medium and large-scale operations and the increased concentration of livestock and poultry establishments in a few animal production areas resulted to environmental pollution problems.

2.3 Sugarcane Bagasse

The sugar industry in the Philippines is still thriving despite the decline in the price of raw sugar in the world market making the local production cost higher than those of imported sugar. The reason is still the vast tracts of land devoted to sugarcane areas (about 370,000 hectares). Bagasse is the major biomass waste produced and mostly utilized by the sugar mills for their boilers. Bagasse production is about 6 million metric tons per year. Cane trash is another biomass wastes from sugar production that have not been utilized in the same scale as that of bagasse. Based from the Industrial Research and Development Office of the Sugar Regulatory Administration, the estimated bagasse production from cane production was estimated at an average of about 29% having a moisture content of about 50%. A 20-25% recovery could be used as a conservative estimate value. The cane trash comprised about three-and-a half percent.[3]

In Negros Province, a bioethanol project operates as a cogeneration project, producing energy uploaded to the power grid. A private firm in Mindanao $92 million in sugarcane ethanol and a10-megawatt co-generation plant from bagasse. The power plant will be given pioneer status, which provides six years of tax-free status because the plant will use a renewable energy source. The ethanol plant received a four-year tax exemption.

2.4 Coconut Wastes

The Philippines has the largest number of coconut trees in the world. Half a billion of coconuts grow on three million hectares across the islands. The country produces most of the world market for coconut oil and copra meal. The major coconut wastes include coconut shell, coconut husks and coconut coir dust. Coconut husks and shells are the two most abundant biomass wastes from coconut oil processing in the country. Coconut shell is the most widely utilized as fuel for boilers and dryers and for making charcoal for household use. The heating value of coconut shell charcoal is comparable to coal. The coconut husk is further processed into coconut fiber whose by-product is the coir dust. The coconut fiber was extensively used as insulation and as car seat cushion in the past but with the use of synthetic foam for seat cushion, the market for coconut fiber became marginal. The coir dust is now marketed as

[2] Guidebook for Developing Sustainable Rural Renewable Energy Services, Department of Energy, Government of the Philippines, Manila, 2000.

[3] Industrial Research and Development Office, Sugar Regulatory Administration, Quezon City, 2005.

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soil conditioner but on a limited basis. For every 1,000 whole nuts of coconut, about 0.180 metric ton of coconut shells are produced, 0.400 metric ton of coconut husks and 0.280 metric ton of coir dust.[4] Of the country's energy sources, coco methyl ester (CME) production can only supply less than 1 percent.

2.5 Forestry Residues

Around 10 million hectares of land in the Philippines is classified as forestlands but the actual , remaining forest cover is only about 1 million hectares. The logging wastes and residues continually declined in consonance with the logging ban imposed by the government. However, if the area previously devoted to forest cover is to be replanted, this would be a significant addition to the country's sustainable energy resource from biomass.

3 POLICIES AND PROGRAMS TO PROMOTE BIO-FUELS

In a nutshell, the Philippine commitment on biofuels is anchored on the Biofuels Act of 2006[5] which mandates a gradual increase in the use of biofuels from 1 percent in 2006 to 2 percent in 2008, rising to 5 percent in 2010 and 10 percent from 2012 onwards. The government also plans to plant jatropha in about a million hectares, most of it in the poor Mindanao region. At the moment, there is a continuing debate on the provisions of biofuels law in response to issues about biofuel real contribution to the socioeconomic fabric and environmental concerns of the country.

The Philippine policy is to reduce dependence on imported fuels with due regard to the protection of public health, the environment, and the natural ecosystems consistent with the country's sustainable economic growth that would expand opportunities for livelihood by mandating the use of biofuels as a measure to:

- Develop and utilize indigenous renewable and sustainable-sources clean energy sources to reduce dependence on imported oil.
- Mitigate toxic and greenhouse gas (GSG) emissions
- Increase rural employment and income; and
- Ensure the availability of alternative and renewable clean energy without any detriment to the natural ecosystem, biodiversity and food reserves of the country.

- The salient provisions of the Biofuels Act include the following
- The Department of Energy (DOE), according to duly accepted international standards, shall gradually phase out the use of harmful gasoline additives such as, but not limited to Methyl Tertiary Butyl Ether (MTBE)
- Mandatory use of biofuels, that is, all liquid fuels for motors and engines sold in the Philippines shall contain locally-sourced biofuels.

To encourage investments in the production, distribution and use of locally-produced biofuels at and above the minimum mandated blends, the following incentives are provided:

- Specific tax - The specific tax on local or imported biofuels component, per liter of volume is zero (0). The gasoline and diesel fuel component, shall remain subject to the prevailing specific tax rate.
- Value Added Tax -The sale of raw material used in the production of biofuels are exempted from the value added tax.
- Water Effluents - All water effluents from the production of biofuels used as liquid fertilizer and for other agricultural purposes are considered "reuse", and are therefore, exempt from wastewater charges
- Financial Assistance - Government financial institutions provide financial services in activities involving production storage, handling and transport of biofuel feedstock, including the blending of biofuels with petroleum.

Organizationally, the National Biofuel Board (NBB) is created to monitor the implementation of the National Biofuel Program (NBP).

3 ISSUES ON BIOFUEL DEVELOPMENT IN THE PHILIPPINES

Since the enactment of the Philippine Biofuels Act of 2006 (R.A. 9367) in 2007, several issues have cropped up that is sharpening the focus of the biofuel industry.

3.1 Biofuels will displace food production

The biofuel program requires large amounts of crops e.g., sugar, cassava and jatropha, in order to produce the needed bioethanol and biodiesel. The land needed to grow such inputs could very well also be used to grow food crops. The proponents of biofuels point out that since bioethanol will be derived from sugarcane, there is no need to tap other arable land areas. Since sugar is only an additive, not an essential food staple like rice or

[4] United Coconut Association of the Philippines, 2010.

[5] Republic Act No. 9367, *The Biofuels Act of 2006*.

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corn, planting more sugar for bioethanol production will not sacrifice food security. New ways of combining food production with energy crop production have already been developed. Energy crops can be targeted for the more marginal lands, while food crops can be grown on more favorable lands.

In the case of jatropha, a tall bush with highly toxic fruit and bark that is already part of the rural farming landscape in the Philippines, can be planted in marginal lands where food crops are not or cannot be grown. Jathropa's seeds are increasingly seen as a more sustainable biodiesel raw material. Foreign investors have expressed interest in massive jatropha planting programs, encouraging local government units to offer large tracts of land. Over a million hectares will be tapped because jatropha needs vast areas to produce a substantial amount of oil. However, jatropha requires a lot of water – about 1,000 mm of rainfall a year. Growing concerns about the environmental sustainability of alternative fuels are forcing decisionmakers to rethink their commitment to such fuels. Is there enough land or water to produce all the crops needed to keep biofuel prices low?

3.2 It will not significantly reduce oil consumption

The biofuels program aims to displace 10% of imported gasoline and diesel by mixing these with bioethanol and biodiesel. But it overlooks the amount of oil products needed in the process of producing bioethanol. The raw materials need to be transported to the processing plant, then these would be transformed to ethanol in the processing plant, which will be followed by the blending of the oil product, and the distribution. All these steps would need oil products to run. These inputs would be equivalent to almost a half of the gasoline or diesel displaced by the biofuels. Thus, instead of the projected 10% reduction in imported diesel and gasoline, it would be only 5% at most.

3.3 Biofuels will be more costly

The whole process of producing biofuels and mixing these with gasoline and diesel would end up with a fuel that will cost more than the unmixed gasoline and diesel. The production of bioethanol and biodiesel is an expensive process. The whole cycle of producing biofuels from harvesting, to transport and processing it are all dependent on oil products, and thus the cost of producing these biofuels will be to a large extent dependent on the price of the oil products.

Biofuels would cost more than ordinary fuel oils; thus, the the government is exempting the biofuel component of the hybrid fuels (i.e., gasoline/diesel mixed with biofuels) from specific taxes in a bid to lower the overall price of the end product. Also, the government is granting an income tax holiday for companies setting up ethanol plants.

3.4 Extensive biofuel crop production will aggravate social tensions over land

The biofuel program and its need for large-scale production will mean that large tracts of land will be needed. This usually means that large plantations will need to be established for this purpose, or more likely big landlords will use the program as an excuse to avoid land redistribution. Biofuel advocates point out to large tracts of untilled lands, many of which are government owned, which would be used for planting the needed crops.

4 CONCLUSION

Biofuel production and utilization are opening new socioeconomic and scientific scenarios in the Philippines.

- Biofuels will play a significant role in the energy and agricultural sectors given the ever increasing oil prices and the need to strengthen the agroindustrial base
- Government biofuel policies and programs are in place but responsive policies should also be put in place to protect the poor farmers involved in biofuel production and ensure food security
- Technology advances on biofuels are being adopted and spearheaded by the private sector particularly in bioethanol production and processing

The large potential of biomass resources in the country is beginning to be tapped. The current low biomass resource utilization vis-à-vis biomass supply can be remedied through a concerted effort to maximize the industry's benefits. One approach is to enhance the population's level of awareness on the issues that beset the production and utilization of biofuels through objective information dissemination about their potential benefits, costs, and risks. Hand-in-hand with this approach would be the continuing research on biofuels impacts on the socioeconomic and biophysical systems, both at the farm, community and national levels.

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Appendix A: Questionnaires

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Questionnaire

International APEC Symposium on

"APEC-ATCWG Biofuels Network Annual Symposium and

Biotrade/Technical Training Workshop"

May 30th – June 4th, 2011, Chiang Mai, Thailand

<u>Instructions</u> The questionnaire is divided into three parts as following;

Part 1 Personal Information

Part 2 Questions

Part 3 Suggestions

Part 1 Personal Information

<u>Instructions</u> Please write ✓ sign in the box (☐) which is the most accurate

1. Gender ☐ 1. Male 54.17% ☐ 2. Female 45.83%
2. Age.............................years

20 - 30 years old	37.50%
31 - 40 years old	16.67%
41 - 50 years old	16.67%
51 - 60 years old	27.08%
61 - 70 years old	2.08%

3. Economy....................................

Indonesia	6.25%
Italy	2.08%
Korea	2.08%
Lao PDR	4.17%
Malaysia	4.17%
Mexico	4.17%
Papua New Guinea	4.17%
Philippines	6.25%
Thai	62.50%
United States of America	2.08%
Viet Nam	2.08%

4. Occupation

☐ Researcher 45.83% ☐ Scientist 14.58%

☐ Student 29.17% ☐ Trader/ Other 10.41%

5. Educational Level

☐ Below Bachelor Degree 2.08% ☐ Bachelor Degree 8.33%

☐ Master Degree 33.33% ☐ Doctorate Degree 56.25%

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Part 2 Questions

Instructions Please write ✓ sign in the box (☐) which is the most accurate

List of Questions	Evaluation Level				
	5 Most likely	4 Likely	3 Moderate	2 Low	1 Lowest
1. If situation permit, you will select the biofuels even though the price is higher than normal fuels	20.83 %	43.75 %	20.83 %	8.33 %	6.25 %
2. The biofuels development in your economy is not sufficient	14.58 %	33.33 %	31.25 %	14.58 %	6.25 %
3. Education plays important roles in the decision of selecting biofuels	52.08 %	31.25 %	16.67 %	0.00 %	0.00 %
4. Please rate the level of negative impact from fossil fuels to the environment	41.67 %	35.42 %	16.67 %	4.17 %	2.08 %
5. This symposium make you realize the Importance of biofuels in the future	60.42 %	31.25 %	8.33 %	0.00 %	0.00 %
6. This symposium helps you establish biofuels network	43.75 %	29.17 %	22.92 %	0.00 %	4.17 %
7. After the symposium you will transfer the gained knowledge to community, friends and/or student	54.17 %	39.58 %	6.25 %	0.00 %	0.00 %
8. How useful of this symposium to your organization	45.83 %	43.75 %	8.33 %	0.00 %	2.08 %
9. In your economies, what is the level of opportunities/roles of women in biofuels related agency/organization of biofuels	18.75 %	39.58 %	35.42 %	6.25 %	0.00 %

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List of Questions	Evaluation Level				
	5 Most likely	4 Likely	3 Moderate	2 Low	1 Lowest
10. Your level of biofuels understanding <u>before</u> participating in the biofuels symposium	14.58 %	33.33 %	39.58 %	8.33 %	4.17 %
11. Your level of biofuels understanding <u>after</u> participating in the biofuels symposium	50.00 %	39.58 %	10.42 %	0.00 %	0.00 %
12. Your satisfaction on the venue of symposium/workshop	54.17 %	41.67 %	4.17 %	0.00 %	0.00 %
13. Your satisfaction on lunch/break	50.00 %	37.50 %	12.50 %	0.00 %	0.00 %
14. Your satisfaction of materials/facilitating equipment	50.00 %	37.50 %	12.50 %	0.00 %	0.00 %
15. Your satisfaction of staff/APEC steering committee services	72.92 %	20.83 %	6.25 %	0.00 %	0.00 %
16. Your overall satisfaction of this symposium/workshop	58.33 %	39.58 %	2.08 %	0.00 %	0.00 %

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Part 3 Suggestions

Your recommendation of lecture/discussion topic(s) to be included in the future APEC funded symposium and activity which is related to biofuels

1. Management aspects or how those alternative fuels presented in this forum are produced.
2. Impact on the formulated and implemented goal and regulations on the usage of alternative fuels in APEC member economies.
3. Environmental and general communities impact on biofuel usage.
4. Proposal of small group discussion based on types of biofuels for economy report and/or recommendation to the steering committee.
5. Inclusion of discussion on other bioenergetics such as biogas and solid biofuels.
6. Method of government implementation among parties/economies on biomass chemistry generation/biofuels to force the bioenergy development.
7. Technology from Israel leading company or technology from Russia and Ukraine.
8. The effect of biofuels on the environments.
9. Impact of biofuels on economy and rural area.
10. Advances in utilization of lignocellulosic biomass for biofuels
11. Appropriate technology which implements biomass feedstock.
12. Future progress on microalgae.
13. Current situation on Jathopha biofuels feasibility.
14. Biofuels technology on microbial fuel cells (MFC) and bioethanol.
15. More discussion relevant to specific issues with invitation of all participants for comments/inputs.
16. Further information on financial and invesment feasibility with involvement of government sector such as Ministry of Energy with inclusion of lecture on thermochemical process.
17. The symposium should be set up on a yearly basis so that sustainable development can be achieved.

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Appendix B: Curriculum Vitae

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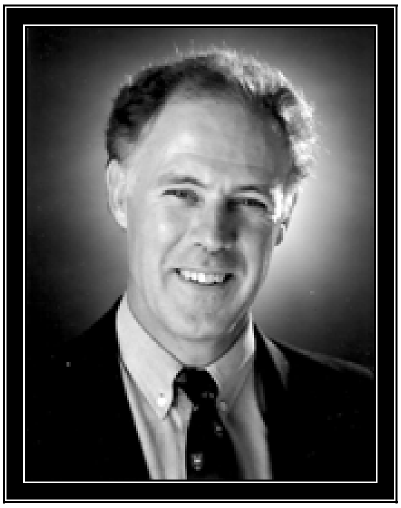
International APEC Symposium on
"APEC-ATCWG Biofuels Network Annual Symposium and
Biotrade/Technical Training Workshop"
May 30th – June 4th, 2011, Chiang Mai, Thailand

KEYNOTE SPEAKER CURRICULUM VITAE



Presenter	
APEC Economy	Australia
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	Manildra Starch P/L (Australia)
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Presentations:(2005 – 2010)	Numerous Conference Presentations both in Australia and internationally on biofuels and higher value fermentation products including pharmaceuticals by enzymatic processes
Publications:(2005 – 2010)	Total publications more than 200 in international journals, 6 international patents. Key recent publication/review on biofuels in Advances in Biochem Eng/Biotechnol. '*Zymomonas mobilis* for Fuel Ethanol and Higher Value Products 108,263-288 (2007). More than 20 peer-reviewed publications (2005-2010) on biofuels and higher value products.
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International APEC Symposium on
"APEC-ATCWG Biofuels Network Annual Symposium and Biotrade / Technical Training Workshop"
May 30th – June 4th, 2011, Chiang Mai, Thailand

ACTIVE PARTICIPANT CURRICULUM VITAE

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Research Skills	• Biofiltration • Bioenergy • Bioprocess Engineering
Presentations:(2005 – 2010)	• G. Aroca. Latest Developments of Biofuels in Chile. International Conference Biofuels Cooperation: Latin America and Europe. 13–14 July 2010, Brussels. Belgium. • G Aroca. Bioethanol from Lignocellulosic Biomass. Internationa Workshop ENERGY & FUELS FROM WASTE & BIOMASS .

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ACTIVE PARTICIPANT CURRICULUM VITAE

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Presentations:(2005 – 2010)	Selected presentations: • BBI International's 21st Annual International Fuel Ethanol Workshop & Expo, Kansas City, Missouri, USA, June 28-July 1, 2005 • 14th European Biomass Conference & Exhibition, Paris, France, Oct. 17-21, 2005 • 2005 Canadian Renewable Fuels Summit，Fraimont Royal York-Toronto, Ontario, Dec. 13-15, 2005 • World Ethanol 2006, Amsterdam, Netherlands, Nov. 7-9, 2006 • Biofuel Ttrade 2006, Shanghai, China, Dec. 5-6, 2006 • 2nd Annual World Renewable Energy Summit 2007, Kuala Lumpur, Malaysia, Mar. 27-28, 2007 • Bioenergy Outlook 2007, Singapore, Apr. 26-27, 2007 • International Conference on Natural Polymers, Bio-polymers, Bio-materials, Their Composites, Blends, IPNs and Gels: Macro to NANO CALES-2007, Kottayam, India, Nov. 19 -21, 2007 • Current Status of Research and Implementation of Bioenergy in China, 2009 • Outlook of Chinese Industrial Biotechnology, 2009 • A Demonstration on Biorefinery of Vegetable Oil: Integrated production for biodiesel and 1,3-propanediol, 2009 • Fuel ethanol development in China, 2009 • Cellulosic ethanol in China, F.O. Licht's Sugar and Ethanol Asia Conference, Philippines, Feb 2-3, 2010 • Bioenergy Korea Conference 2010 International Symposium, Korea, March 17-18, 2010 • Co-production of biodiesel & 1,3-propanediol from lipids with biological conversion, Nov. 8-10, Taiwan
Publications:(2005 – 2010)	So far, he has published more than 150 papers and applied more than 60 patents, among which 26 patents have been authorized. Following are selected publications. • Sun, T; Du, W; Liu, DeHua, Prospective and impacts of whole cell mediated alcoholysis of renewable oils for biodiesel production, BIOFUELS BIOPRODUCTS & BIOREFINING-BIOFPR. 3 (6):633-639 2009 (DOI 10.1002/bbb.180) • Xu, YZ; Guo, NN; Zheng, ZM; Ou, XJ; Liu, HJ; Liu, Dehua. Title Metabolism in 1,3-Propanediol Fed-Batch Fermentation by a D-Lactate Deficient Mutant of Klebsiella pneumonia, BIOTECHNOLOGY AND BIOENGINEERING 104 (5):965-972 2009, (DOI 10.1002/bit.22455) • Zhangqun Duan, Wei Du, Dehua Liu. The solvent influence on the positional selectivity of Novozym 435 during 1,3-diolein synthesis by esterification. BIORESOURCE TECHNOLOGY, DOI:10.1016/j.biortech.2009.11.087 • Zhangqun Duan, Wei Du, Dehua Liu. The pronounced effect

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Asia-Pacific Economic Cooperation

International APEC Symposium on
"APEC-ATCWG Biofuels Network Annual Symposium and Biotrade/Technical Training Workshop"
May 30th – June 4th, 2011, Chiang Mai, Thailand

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	America internationally on biofuels and higher value fermentation products by enzymatic processes

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Presentation:	
Title	**Integrated production for biodiesel and 1,3-propanediol**
Abstract	The 1st commercialized production line in the world which utilize A Novel bio-process to produce biodiesel with low-quality oil (such as waste cooking oil, PFAD, acidyfied oil etc) as feedstock, while utilizing lipase as catalyst. On the other hand, we can take crude glycerol, which is the main by-product of all kind of biodiesel plants, to produce 1,3-Propanediol(PDO) by directly fermentation, PDO can be used to polymerize with PTA to make PTT which is so-called "King of chemical fiber".

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Economic Cooperation

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Biotrade/Technical Training Workshop"
May 30th – June 4th, 2011, Chiang Mai, Thailand

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Academic Title or Position at the Place of Employment	Full Professor
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Fields of Interest	• Microalgae • Biofuels • Biochemical Engineering
Full biography such as experiences, etc.	• 08/1990-11/2000 Assistant prof. Institute of Process Engineering, Chinese Academy of Sciences • 11/2000-10/2003 Postdoctoral Research Fellow, the Birmingham University, UK • 03/2004-06/2008 Associate Prof. Ocean University of China • 06/2008-06/2010 Assistant Prof. Qingdao Institute of Bioenergy and Bioprocess Technology

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	(Qibebt), CAS • 06/2010-today Full Prof. Qingdao Institute of Bioenergy and Bioprocess Technology (Qibebt), CAS
Research Skills	• Mass cultivation of microalgae • Photobioreactor • Downstream processing technology • Fermentation
Publications and patents	• Liu Tianzhong, Liu Dehua, Xie Dongminand Ouyang Fan.(1999) Simulation of flow of liquid-jetting loop reactor under periodically oscillatory operation. Journal of Chemical Industry and Engineering(China), 50(3):411—416 • Liu Tianzhong, Su Ge, Li Jing, Qi Xiangming and Zhan Xiaobei.(2010) Numerical Simulation of Flow in Erlenmeyer Shaken Flask, "Computational Fluid Dynamics", Edited by Hyoung Woo OH，ISBN 978-953-7619-59-6，Intechwev.org. ，2010-1，1st Edition • Liu Tianzhong. (2008). A protential research hot point on energy microalgaec. *China Biodiesel*. 3:27-28. • Lu W., Xu J., Gao X. and Liu T.(2007)Preparation of calcium alginate fibre.Journal of Ocen University of China,37（addition）：73-76 • T. Liu, A. M. Donald* and Z. Zhang.(2005). Novel Manipulation in Environmental SEM for Measuring the Mechanical Properties of Single Nano-Particles. Material Sciences and Technology.21(3):290-294 • T. Liu, X. Qi, G. Su, J. Jin, and W. Cong. (2008) Computational fluid dynamics (CFD) simulation of flow in the rotary drum for pyrite bio-preoxidization. Chemical Engineering and Processing. 47(5):971-978 • T.Liu.(2005) Characterization of the Mechanical Properties of Urea-Formaldehyde Microcapsules. The Chinese Journal of Process Engineering.5(4):450-454 • Tianzhong Liu and Zhibing Zhang. (2003). Mechanical Properties of desiccated Ragweed Pollen Grains Determined by Micromanipulation and Theoretical Modelling. Biotechnology & Bioengineering,85(7)：770-775 • Chen L. and Liu T. (2010) Preparation of Microalgae biodiesel. Biomass Chemical Engineering, (accepted) • Chen M. and Liu T.,(2011) Subcritical Ethanol Extraction of Oil from Algae Wet Paste, • Chen Y., Liu T, *et al.*,(2010) Effects of initial population density (IPD) on growth and lipid composition of Nannochloropsis sp. • Chen X. and Liu T.(2010) Harvesting of Scenedesmus sp. using polyvinylidene Fluoride ultrafiltration membrane. • Chen M. ,Chen X. and Liu T.(2011) Subcritical Co-solvents Extraction Of Lipid from Wet Microalgae Pastes of Nannochloropsis sp. • Jiang H., Gao L.and Liu T.(2011) The influence of Nitrogen and Phosphor on the Growth and Lipid Accumulation of Chollera. Journal of Food Industrial Science.(accepted) • Gao L. and Liu T., (2010) Sidimentation and flotation characteristization for chollora, Oceanlogy Sceience., 34(12) • Gao L.Jiang H. and Liu T., (2010), Recovery Conditions of Porphyridium Cruentum Cells by Dissolved Air Flotation, Journal of Ocean University of China.40(11)

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- Li J., Zhan X., Liu T., Zheng Zhi. And Qi X.(2009) numerical Simulation of fluid flow in Erlenmeyer Shaken Flask with Computational Fluid dynamics, CIESC Journal.60(4): 878-885
- Li J., Zhan X., Zheng Zhi.Liu T., Qi X., Jiang Y. and Liu T.(2009) Numerical Simulation on Fluid Flow of Xanthan Gum Solution Stirred by Diferent Types of Impeller, The Chinese Journal of Process Engineering.(4): 634-640
- Dongming Xie, Dehua Liu and Tianzhong Liu. (2001). Modeling of glyverol production by fermentation in different reactor states. Process Biochemistry. 36:1225-1232.
- Ji Y., Gao L. Zhao H., Cheng J. and Liu T.(2009)Fed batch culture of Oenococcus., Agr-product Process.12:29-35
- Jiang Y., Ji Y. liu H. Li S. and Liu T., (2008)Optimization of Fermentaion of Lactic Acid by *Rhizopus Oryzae*, Agr-product Process.1:72-75
- Jiang Y., Ji Y.Gao L. Jiang H. Zhao H. and Liu T. （2009） Kinetics of Lactic Acid by Lactobacillus rhamnosus, Journal of Chinese Food (4):57-63
- Jiang Y.,Liu T.,Gao L. Ji. Y. and Qi X.(2009）Kinetics of Lactic Acid by Rhizopus Oryzae, Journal of Chinese Food 9(2):18-22
- Xie D., Liu D., Zhu, H and Liu T. (2001). Multipulse feed strategy for glycerol fed-batch fermentation- A steady-state nonlinear optimization approach, Applied Biochemistry and Biotechnology . 95(2):103-112
- Xie Dongming, Liu Dehua & Liu Tianzhong. (2001). Glycerol Production by Fermentation with Periodic Oscillating Airlift Loop Reactor (ARLR). China J. Appl. Environ. Biol. 2001,7(1):72—75
- Xie Dongming, Liu Dehua and Liu Tianzhong. (2000). Optimal Feed Strategy for Feb-batch Glycerol Fermentation Determined by Maximum Principle. Journal of Chemical Industry and Engineering 51:236-243

Patents：

- Liu Tianzhong and Zhang Wei. A method of microalgal culture in light-emission particles suspended in photo-bioreactor. Chinese patent CN200810167501.3.
- Liu Tianzhong, Jiang Huaizhen, Gao Lili, Zhang Wei and Chen Yu. A Method to extract both lipid and protein from microalgae. Chinese Patent CN2010071200033940
- Liu Tianzhong, Gao Lili, Jiang Huaizhen, Qi Xiangming and Xu Yuyan. A device of foam separation with multi-rods for the harvesting of microalgal cells. Chinese Patent CN200810159413.9
- Liu T., Zhang W. *et al*. A method for mass cultivation of microalgae. Chinese Patent CN2010101363004
- Liu T., Che M. *et al*., A method of lipid extraction by sub-critical ethanol from wet algal pastes. Chinese patent CN2010101363095

Awards and Honors:

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ACTIVE PARTICIPANT **CURRICULUM VITAE**

Presenter	
APEC Economy/Country	Indonesia
Full Name	DadanKusdiana
Academic Title or Position at the Place of Employment	Doctor
Affiliated Organization	Directorate General of New Renewable Energy and Energy Conservation, Ministry of Energy and Mineral Resources
Fields of Interest	• Small scale biofuel production • Sustainable biofuel development • Practical technology • Technical issue on biofuel • Rural empowerment
Full biography such as experiences,	**Dr. DadanKusdiana**was born in Sumedang, Indonesia on 29 December 1968. He was graduated from Bogor Agricultural University in 1992 majoring in agricultural engineering. In 1993, he then joined Ministry of Energy and Mineral Resources with first assignment on energy efficiency. From 1998 to 2004, he got a scholarship from Government of Indonesia to continue his master and doctoral degree in Kyoto University, Japan with research topic

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	on biodiesel production process. Currently, he is now Head for Rural Energy Division, Directorate General of Electricity and Energy Utilization. He is a member of Indonesian Renewable Energy Society and Indonesian Biofuel Forum.
Research Skills	• Biodiesel production process
Presentations:(2005 – 2010)	• Various presentations on biofuel and renewable energy have been made for mostly bilateral and multilateral cooperation.
Awards and Honors:	• Best young research on Energy from Japan Energy Society in 2004

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ACTIVE PARTICIPANT CURRICULUM VITAE

Presenter	
APEC Economy/Country	Indonesia
Full Name	Oberlin Sidjabat
Academic Title or Position at the Place of Employment	DR; Researcher, Head of Technology Research Group for Conversion Process & Catalyst
Affiliated Organization	R&D Centre for Oil and Gas Technology "LEMIGAS", Ministry of Energy and Mineral Resources
Fields of Interest	• Biofuel Production • Catalytic Processing for Energy
Full biography such as experiences, etc.	**<u>Education:</u>** • Academy of Analytical Chemistry, Bogor, Indonesia, B.Sc (1975) • Chemical Engineering, The University of New South Wales, Sydney, Australia, Ph.D (1995) **<u>Work Experiences:</u>** • 1975 – Present: LEMIGAS employee as researcher

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	• 2007 - Present: Head of Process Technology and Catalyst Group • 1995 – Present: Coordinator for Research on Biodiesel • 2001 – 2004: Coordinator for ARHDM Catalyst Selection in PERTAMINA Balongan • 2006 – 2007: Coordinator for Pilot Plant of Biodiesel • 2008: Development and Installation of Biodiesel Plant in Rokan Hulu, Province of Riau, with capacity 30 Ton per day (As Coordinator of Project)
Research Skills	• Biofuel • Catalytic Processing for Fuel and Environment
Presentations:(2005 – 2010)	• Oberlin Sidjabat, Abdul Haris and Cut Nanda, Production of Bioethanol and Compost From Organic Waste (Vegetable Garbage) of Traditional Market, *International APEC Symposium on "Biofuel from Agricultural and Argo-Industrial Wastes"*, Chiang Mai, Thailand, 24-27 May 2010 • Oberlin Sidjabat and Ego Syahrial, Carbon Capture And Storage–Enhancement Oil Recovery Potential in Indonesia, *Indonesia-UK Working Group on Environmental and Climate Change*, London, UK, 15-16 June 2009 • Oberlin Sidjabat, Abdul haris, and Hadi Purnomo, The Effect of Biodiesel Blending by Splash Technique in Engine Performance, *The 7th Asian Petroleum Technology Symposium*, Ho Chi Minh, Vietnam,18-19 February 2009 • Oberlin Sidjabat, Biofuel Production in Indonesia, *Asian Productivity Organization Forum*, Jakarta, Augustus 2007 • Oberlin Sidjabat, Yanni Kussuryani, and Hadi Purnomo, The Influence of Biodiesel Production for Fuel Quality, *International Biofuel Conference* - Bali, Indonesia, 10-11 December 2007
Publications:(2005 – 2010)	• Oberlin Sidjabat, Abdul Haris, and Cut Nanda, Production of Bioethanol and Compost From Organic Waste (Vegetable Garbage) of Traditional Market, *Proceeding of International APEC Symposium on "Biofuel from Agricultural and Argo-Industrial Wastes"*, Chiang Mai, Thailand, 24-27 May 2010 • Morina and Oberlin Sidjabat, The Effect of Ca and Cr in Production of Bio-gasoline from Metyl-Esther as feedstock, *Lembaran Publikasi Lemigas*, (2009), Vol. 43, (Indonesian Languange) • Dessy Yoediartiny, Oberlin Sidjabat, and Niken Atmi, Feasibility Study of Crude oil Refining from Sharing Contractor of Oil Company for Reducing Fuel Deficit, *Mineral & Energi,* (2009), Vol. 7 (Indonesian language) • Triyono and Oberlin Sidjabat, The Effect of Water and Ethanol in reactivity of iso-amyl alcohol hydrogenolysis with Ni/Zeolite catalyst, *Lembaran Publikasi Lemigas*, (2008), Vol. 42, (Indonesian language)
Awards and Honors:	

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KEYNOTE SPEAKER CURRICULUM VITAE

Presenter	
APEC Economy	
Full Name	Roberto De Philippis
Academic Title or Position at the Place of Employment	Professor, Microbial Biotechnology
Affiliated Organization	University of Florence, Italy
Fields of Interest	• Microbial biotechnology • Environmental microbiology
Full biography	• Degrees: MSc in Chemistry (1978); PhD in Microbial Biotechnology (1987) • International Appointments: - Secretary/Treasurer (1999-present) and member of Executive Committee (2005-present) of the International Society for Applied Phycology; - Italian Delegate in the IEA-HIA (International Energy Agency- Hydrogen Implementing Agreement) New

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	Annex 21 "Bio-inspired and Biological Hydrogen" (2010-present); - member of the Editorial board of the Journal of Applied Phycology (2005-present); - Guest Editor of Process Biochemistry for the Special Issue dedicated to IBS2010. • International Experience : responsible of several International Research Projects with China, Israel, Portugal, India, Mexico, USA.
Research Skills	• Extensive University experience in teaching and research/ research supervision (more than 10 PhD students). • Research skills in physiology and biochemistry of photosynthetic bacteria. In particular, he is studying the physiology and the possible biotechnological exploitation of phototrophic microorganisms in the production of biopolymers of industrial interest or in processes related to the production of energy from renewable resources or for the treatment of polluted waters.
Presentations:(2005 – 2010)	• More than 40 Conference Presentations both in international and in Italian Congresses on the main topics of his studies.
Publications:(2005 – 2010)	• Total publications: more than 160 scientific papers, mainly on international peer reviewed journals, three chapters in books. • Recent publications on photobiological hydrogen production with *Rhodopseudomonas palustris* in Int J Hydrogen Energy 2008 (33: 6525) and 2010 (35: 12216). • Two chapters on microbial hydrogen production in the book "Microbial Technologies in Advanced Biofuels Production", Patrick C. Hallenbeck, Springer Verlag (in press).

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ACTIVE PARTICIPANT CURRICULUM VITAE

Presenter	
APEC Economy	Papua New Guinea
Full Name	Andrew Puy
Academic Title or Position at the Place of Employment	Lecturer
Affiliated Organization	The Papua New Guinea University of Technology
Fields of Interest	• Renewable Energy • Biofuels • Appropriate Technology
Full biography such as experiences,	• Bachelor of Engineering in Mechanical Engineering, PNG University of Technology, 1989 • Master of Engineering Science in Development Technologies, The University of Melbourne, Australia, 1996
Research Skills	• University experience in teaching, research and community development projects in renewable energy, appropriate technology and biofuels.

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Presentations:(2005 – 2010)	• Sumanasiri, K.E.D. and Puy A., *'Status of Renewable Energy Development in Papua New Guinea and Potential for Utilisation in Agriculture and Other End-use Applications'*, International Agricultural Engineering Conference, Asian Institute of Technology, Bangkok, Thailand, 3 - 6 December 2007 • Puy, A., '*Coconut Biofuel Activities in Unitech'*, Power point presentation presented at the Inaugural Coconut Oil Biofuel Symposium, National Fisheries College, Kavieng, New Ireland, 18 March 2008
Awards and Honors:	• John Crawford Scholarship for Post Graduate Study in Australia, 1995.

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ACTIVE PARTICIPANT **CURRICULUM VITAE**

Active Participant	
APEC Economy	Papua New Guinea
Full Name	Reilly Nigo
Academic Title or Position at the Place of Employment	Lecturer
Affiliated Organization	PNG University of Technology, Lae, Papua New Guinea
Fields of Interest	• Renewable Energy • Biofuels • Appropriate Technology
Full biography	• Bachelor of Science in Food Technology, The Papua New Guinea University of Technology, 1993 • Master of Science in Process Engineering (Food Engineering), The University of Reading, UK • Reading towards Phd Chemical Engineering (pending correction - current), University of Cambridge, UK • The author has briefly worked in 1994-1995 manager Anderson Foodland and Nestles (PNG) both in food process engineering/production. • Currently working as a University lecturer, PNG University of Technology

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	• The author plays key advisory roles in Food Security, food manufacturing and downstream processing agricultural commodities.
Research Skills	• University teaching experience including research and community development projects. • Community development work experience in renewable energy and rural and urban settlements development mostly through student-centered projects, consultancies including small scale bio-diesel processing and quality tests , portable biogas production using household wastes, design and manufacture fuel efficient wood stoves and small scale food industry in rural, urban settlements and households in reduction of poverty and moving towards green energy and reduction of environmental pollution.
Presentations:(2005 – 2010)	• Nigo, R.Y., '*Move Towards Clean and Renewable Energy in PNG: An Over View*, PNG University of Technology, Seminar Presentation Series, Lae, Papua New Guinea, 31 May, 2010.
Publications:(2005 – 2010)	• R.Y.Nigo, R.Paterson and I.Wilson, " *Novel Design – Spinning Disc Apparatus*": Fouling Behaviour of Food Fats, Food Fouling and Cleaning Conference, November, 2006, University of Cambridge**.** • R.Y.Nigo, Y.M.J.Chew, R.Paterson and I.Wilson, " Experimental Studies of Freeze Fouling of Model Food Fat Solutions using a Novel Spinning Disc Apparatus", *Energy and Fuels*, October, 2006. • Alu.E. and Nigo,R. *'Development of Solar Sorption Freezer, A Preliminary Investigation using Activated Carbon and Methanol Absorption Pairs*, PNG University of Technology, Lae, 29 October, 2009. • Saki.B. and Nigo,R. *'Production of Biodiesel from Coconut,Palm Oil and Waste Vegetable Oils*, PNG University of Technology, Lae, 29 October, 2009. • Carseldine.Craig and Nigo.R." Biogas Production from Household Wastes, *PNG University of Technology, Lae, 29 October, 2010.*
Awards and Honors:	• PNG Government Higher Education Scholarship to do Master of Science in Process Engineering, Reading University, UK, 1996-1997 • UK Government Scholarship through Association of Commonwealth Universities to do PhD in Chemical Engineering, Cambridge University, 2004-2008

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KEYNOTE SPEAKER CURRICULUM VITAE

Presenter	
APEC Economy	Japan
Full Name	Takashi Watanabe
Academic Title or Position at the Place of Employment	Professor, Biomass Conversion Vice director of Research Institute for Sustainable Humanosphere
Affiliated Organization	Kyoto University
Fields of Interest	• Biomass Conversion • Microbial lignin degradation
Full biography	• Degrees :Bachelor degree in Agric. (Kyoto Univ,1982); • Master degree in Agric. (Kyoto Univ,1984) (Scholarship from Japan Scholarship Foundation) • Doctor degree in Agric. (Kyoto Univ,1989) • Research scientist of Sanyo-Kokusaku Pulp Co., Ltd. (Present name: Nippon Paper Industries Co., Ltd.) (1984-86). (Ninomiya Award, Exellent Prize, 1985) • Research Associate (1987-93), Assoc. Prof. (1993-2002) and Prof. of Wood Research Institute, Kyoto University (2002-04)

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	• Prof. of Research Institute for Sustainable Humanosphere (RISH), Kyoto University (2004-) • Head of Center for Exploratory Research on Humanosphere, Research Institute for Sustainable Humanosphre (RISH), Kyoto University (2008-2010) • Vice director of Research Institute for Sustainable Humanosphre (RISH), Kyoto University (2010-) • International Experience : Visiting Scientist of Institute of Biochemical Technology and Microbiology, University of Technology, Vienna (1996-97) , Visiting Professor of South China University of Technology (2001) , External Assessor of Department of Chemistry, Universiti Sains Malaysia (2003) • Editor, advisory board and editorial board of international journals such as • Applied Microbiology and Biotechnology, Holzforschung and Journal of Wood Science. • Chairs of the thematic working group in Finnish–Japanese Workshop on functional materials, JST (2009). • Coordinator of "Fostering Program of Leading Young Scientists toward the Establishment of Humanosphere Science in East Asia", JSPS (2010-11). • Project leader of biofuel production from lignocellulosics including "Development of Bioethanol Production Process from Woody Biomass Based on High Efficient Solvolysis and Ethanologenic Bacteria", NEDO (Total budget > 1 billion yen). • Fellow of International Academy of Wood Science (2007-)
Research Skills	• Extensive University experience in teaching and research/ research supervision. • Research skills in chemistry, biochemistry and applied mycology related to lignin biodegradation, biomass conversion and structural analysis of plant cell wall components.
Presentations:(2005 – 2010)	Numerous presentations in international conference including keynote , plenary and invited talks on biomass conversion and lignin biodegradation, such as "An International Forum ASEAN-Korea Symposium and Workshop on Biorefinery Technology for Sustainable Production of Biofuel and Industrial Biochemicals" (2010).
Publications:(2005 – 2010)	Total publications more than 100 in international journals, 30 books and 30 patents.
Awards and Honors:	

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"Biofuels from Agricultural and Agro-Industrial Wastes"
May 24th – 27th, 2010, Chiang Mai, Thailand

KEYNOTE SPEAKER CURRICULUM VITAE

Presenter	
APEC Economy	Korea
Full Name	Choul-Gyun LEE
Academic Title or Position at the Place of Employment	Professor, Biotechnology
Affiliated Organization	Inha Unviersity, Incheon, Korea
Fields of Interest	• Microalgal Biotechnology • Marine Bioenergy • Systems Biology
Full biography such as experiences,	• Degrees : BS (Seoul Nat'l Univ. 1985); MSc (Seoul Nat'l Univ. 1988); M.S.E. (Univ. of Michigan, Ann Arbor, MI, USA, 1992) Ph.D. Eng (Univ. of Michigan, 1994) • Inha Fellowship Professor, Inha University, Incheon, Korea, 2010. 3. – present. • Leader (PI), National Project on Marine Bioenergy, Ministry of Land, Transportation and Maritime Affairs, Korea, 2009.12. – present. • Head, Institute of Industrial Biotechnology, Inha University, Korea, 2007. 3. – present. • Director, Technology Transfer Office, Inha University, Incheon, Korea, 2007. 3. – 2008.1. • Vice Dean, Division of Research, Inha University, Incheon, Korea, 2006. 4. – 2008.1.

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	• Professor at Inha University, Incheon, Korea, 1997. 3. – present. • Visiting Scholar at University of California, San Diego, USA, 2003. 2. – 2004. 2. • Resident Research Associate at NASA, Kennedy Space Center, USA, 1996. 1. – 1997. 1. • Post-Doctoral Research Fellow at University of Michigan, 1995. 1. – 1995. 12. • Engineer (part-time), Aastrom Bioscience Inc., Ann Arbor, USA, 1995. 1. – 1995. 6. • Teaching Assistant at University of Michigan (1992 – 1994). • Scientific Researcher at KRIBB, Taejon, Korea, 1989. 8. – 1990. 8. • Research Assistant at Seoul National Univ. (1986-1988) and U. of Michigan (1990-1994).
Research Skills	• Extensive University experience in teaching and research/ research supervision • Numerous of research planning and consulting for government • Research skills in photobioreactor engineering, biochemical engineering, biotechnology, systems biology, process designs and analysis
Presentations:(2005 – 2010)	Numerous conference presentations in all over the world including a number of keynote speaks and several plenary talks on marine microalgal bioenergy production
Publications:(2005 – 2010)	Over 40 peer reviewed publications (2005-2010) on microalgal biotechnology including microalgal biofuels and over 10 international patents
Awards and Honors:	• Korean National Scholarship, Korean Government (1990-1994) • National Research Council Fellowship (NRC, USA, 1995-1997) • New Scholar Award (The Korea Society for Biotechnology and Bioengineering, 2000) • Young Scholar Award (The Korean Society for Microbiology and Biotechnology, 2007) • Best Research Award (Inha University, 2009) • Best Teaching Award (Inha University, 2010)

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ACTIVE PARTICIPANT CURRICULUM VITAE

Presenter	
APEC Economy	Russia
Full Name	Alex Ablaev
Academic Title or Position at the Place of Employment	President of Russian Biofuels Association
Affiliated Organization	Russian Biofuels Association
Fields of Interest	• Bioprocesses, fermentation • Biobutanol & ethanol production • Second generation biofuels
Full biography such as experiences, etc.	• 2006-current Russian Biofuels Association, President • 2004-2009 Genencor, a Danisco division, Senior Manager, Marketing and Business Analyst
Research Skills	• Enzymes • Fermentation processes
Presentations:(2005 – 2010)	• Alex Ablaev (2010), master-class «Management of innovation business in biotechnology», Institute of Economic Strategy Russian Academy of Science, Moscow, Russia • Alex Ablaev (2010), «Launch of the innovation future", plenary presentation at the International conference

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	«Ecology of big city", Moscow, Russia • Alex Ablaev (2010), master-class «Innovations in biotechnology», Institute of Economic Strategy Russian Academy of Science, Moscow, Russia • Alex Ablaev (2009), «Biofuels and food security", in McGill Conference on Global Food Security, Montreal, Canada • Alex Ablaev (2009), «Biofuel: New market opportunities for agriculture", in VIII Russian Agri-Food Forum by Adam Smith Institute, Moscow, Russia • Alex Ablaev (2009), «Russia's Emerging Bio-economy: New Developments in Biofuels and Bionanotechnology», in the BIO International Convention, Atlanta, USA • Alex Ablaev (2008), «Prospect of Biofuels in Russia and Former Soviet Union", APEC Task Force meeting, Denver, USA • Alex Ablaev (2007), "Bioethanol for Russian market", International Congress "Fuel Bioethanol-2007", Moscow, Russia • Alex Ablaev (2006), "Biodiesel production and application", international Congress "Biodiesel-2006", Moscow, Russia
Publications:(2005 – 2010)	• Alex Ablaev (2010), Grain processing in bioproducts – base for regional development in Russia, in Agrotechnology, 11-2010 • Alex Ablaev (2010), High-tech grain, in Independent Newspaper, 25.01.2010 • Alex Ablaev (2008), Russia: necessary conditions for biofuel industry, in Energopolis, 2-2008 • Alex Ablaev (2007), Biodiesel for Russia, Oil & Fats, 10-2007 • Alex Ablaev (2007), Biofuels, think outside of oil pipe, in Russian Expert Review, 1–2 (20) 2007 • Alex Ablaev (2006), Production and application of biodiesel, book • Alex Ablaev (2005), Catalysis of creation: enzymes for safe and healthy food, in Food Ingredients, raw materials & additives, 1-2005, 26-28 • Alex Ablaev (2005), Enzymes for new types of beer and beverages, in Beer and Beverages, 2-2005, 78-79
Awards and Honors:	

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ACTIVE PARTICIPANT CURRICULUM VITAE

Presenter	
APEC Economy	Malaysia
Full Name	Dr. Lee Keat Teong
Academic Title or Position at the Place of Employment	Associate Professor (Deputy Dean)
Affiliated Organization	Universiti Sains Malaysia
Fields of Interest	• Energy Sustainability and Biofuel from Biomass
Full biography such as experiences, etc.	• 2004-2007 Lecturer Universiti Sains Malaysia • 2007-2009 Senior Lecturer Universiti Sains Malaysia • 2009-present Assoc. Professor Universiti Sains Malaysia
Research Skills	• Biodiesel and Bioethanol Production • Supercritical Technology • Catalytic Technology
Presentations:(2005 – 2010)	**ONLY list 2009 onwards** • "A Comparative Study on the Energy Policies in Japan and Malaysia in Fulfilling Their Nations' Obligation Towards Kyoto Protocol for CO_2 Reduction", **1st Regional Conference on Geo-Disaster Mitigation and Waste Management in**

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Publications:(2005 – 2010)	**ONLY list 2010 onwards** • Hossein Mazaheri, **Keat Teong Lee**, Subhash Bhatia and Abdul Rahman Mohamed, "Subcritical Water Liquefaction of Oil Palm Fruit Press Fiber for the Production of Bio-oil: Effect of Catalysts", **Bioresourse Technology**, Elsevier, Vol. 101 (2) **2010**, pp 745-751. • Siew Hoong Shuit, **Keat Teong Lee**, Azlina Harun Kamaruddin, Suzana Yusup, "Reactive Extraction and In Situ Esterification of Jatropha curcas L. Seeds for the Production of Biodiesel", **Fuel**, Elsevier, Vol. 89 (2) **2010**, pp 527-530. • Kok Tat Tan, **Keat Teong Lee**, Abdul Rahman Mohamed, "A Glycerol-free Process to Produce Biodiesel by Supercritical Methyl Acetate Technology: An Optimization Study via Response Surface Methodology, **Bioresource Technology,** Elsevier, Vol. 101 (3) **2010**, pp 965-969. • Goh Chun Sheng, **Lee Keat Teong**, "A Visionary and Conceptual Macroalgae-based Third Generation Bioethanol (TGB) Biorefinery in Sabah, Malaysia as an Underlay for Renewable and Sustainable Development", **Renewable and Sustainable Energy Reviews**, Elsevier, Vol. 14 (2) **2010**, pp 842-848.

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	optimisation study using response surface methodology", **Carbohydrate Polymer**, Elsevier (In Press). • Man Kee Lam, **Keat Teong Lee**, "Renewable and sustainable bioenergies production from palm oil mill effluent (POME): Win–win strategies toward better environmental protection", **Biotechnology Advances**, Elsevier (In Press). • Steven Lim, **Keat Teong Lee**, "Parallel production of biodiesel and bioethanol in palm oil based biorefinery: Life cycle assessment on the energy and greenhouse gases emissions", **Biofuels, Bioproducts & Biorefinery**, Wiley-Blackwell (In Press). • Kok Tat Tan, **Keat Teong Lee**, Abdul Rahman Mohamed, "Response to "Comment on A Glycerol-Free Process to Produce Biodiesel by Supercritical Methyl Acetate Technology: An Optimization Study via Response Surface Methodology, **Bioresource Technology**, Elsevier (In Press).
Awards and Honors:	• National Science Fellowship (PhD program) from July 2000 to Jun 2003 Ministry of Science, Technology and Environment, Malaysia • Best Thesis Award 2004 Universiti Sains Malaysia • Excellence Service Award 2006 – Universiti Sains Malaysia

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Economic Cooperation

International APEC Symposium on
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May 30th – June 4th, 2011, Chiang Mai, Thailand

ACTIVE PARTICIPANT CURRICULUM VITAE

Presenter	
APEC Economy	Malaysia
Full Name	Wan Hasamudin Wan Hassan
Academic Title or Position at the Place of Employment	Senior Research Officer/Group Leader
Affiliated Organization	Malaysian Palm Oil Board (MPOB)
Fields of Interest	• Bioprocess • Fermentation • Cellulose Derivatives • Biopolymer/Degradable Plastics • Biofuel/Bio-energy
Full biography such as experiences, etc.	• 1992 – 2000 Res. Officer Palm Oil Res. Inst. Of Malaysia (PORIM) • 2000 – 2001 Res. Officer Malaysian Palm Oil Board (MPOB) • 2001 - Senior Res. Officer Malaysian Palm Oil Board (MPOB) • 2009 - Group Leader Bio-composite Research Group (MPOB)

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	• 2010 - Team Leader (I) B5 (Biofuel) Working Group (MPOB) • 2006 - Advisory Committee for Skill Development (Biotech.), Ministry of Human Resource (Malaysia)
Other Appointments	• COP Auditor (Palm Oil Industry, Malaysia: 2010 -) • Internal Auditor ISO9001:2008 (MPOB; 2010 -) • Industry Expert [(Biotechnology), Malaysia: 2009 -] • Bio-energy Committee (MPOB) • B5 Logistic Sub-Committee (2008 -) • B5 Std & Quality Sub-Committee (2008 -) • Secretary for B5 Implementation (2008-2010) • Biomass Sub-Group: IE Paper for Alternative Energy in Malaysia (2010 -) • Active Representative/Participant from Malaysia for Biomass/Bio-energy Asia/ASEAN Workshop • Advisor for Science Project (Science Secondary School, SHAH Pekan, Pahang, Malaysia): ***Bio-ethanol from Overripe/Rotten Banana***, 2006-2007; National Champion (2006) and represented Malaysia for INTEL-ISF Competition at New Mexico, USA (2007)
Research Skills	• Bio-processing • Bio-fuel/Bio-energy • Cellulose Derivatives • Biopolymer/Degradable Plastics
Presentations: (2005 – 2010) (International)	• Syamsiah Abu Bakar, Rosma Mohd Dom, Ajab Bai Akhbarally and Wan Hasamudin Wan Hassan* (2010) Prediction of Physical Properties of Degradable Plastics: A Fuzzy Approach. In: *The 2010 International Conference on Science and Social Research (CSSR 2010): Universiti Teknologi MARA*, December 5-7, 2010, Kuala Lumpur, Malaysia. (Oral Presentation) • Wan Hasamudin Wan Hassan*, Choo Yuen May, Lim Weng Soon, Ropandi Mamat, Astimar Abdul Aziz, Harrison Lau Lik Nang and Yung Chee Liang (2010) Malaysian Palm Oil Industry for Sustainable Energy. *In: 3rd Asia Biomass Energy Workshop (Bio-fuel Database Workshop) / 7th Biomass-Asia Workshop*, November 29 - December 1, 2010, Jakarta, Indonesia. (Oral Presentation) • Wan Hasamudin Wan Hassan*, Choo Yuen May, Lim Weng Soon, Ropandi Mamat, Astimar Abdul Aziz, Harrison Lau Lik Nang and Yung Chee Liang (2010) Developments of Palm Biofuel in Malaysia: Advances & Challenges. In: *2nd Seminar on Catalysis Science and Technology, Pusat Kecemerlangan Sains dan Tekonologi Katalisis (PutraCAT), Fakulti Sains, Universiti Putra Malaysia,* November 24, 2010, Serdang, Selangor Malaysia. (Oral Presentation) • Wan Hasamudin Wan Hassan*, Mohd Basri Wahid, Choo Yuen May, N. Ravi Menon, Lim Weng Soon, Astimar Abdul Aziz and Nasrin Abu Bakar (2010) SREP Development in Sabah from MPOB Perspective. In: *Sabah Electricity Sdn Bhd (SESB) Open Day*, July 17, 2010, Kota Kinabalu, Sabah, Malaysia. (Oral Presentation) • Wan Hasamudin Wan Hassan*, Ropandi Mamat, Astimar Abdul Aziz, Mohd Basri Wahid, Choo Yuen May and Lim Weng Soon (2010) Renewable Energy Outlook in Malaysia: *Workshop on Industrialisation of Seedling Production for*

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Participation in APEC Meeting (latest)	• 34th APEC Expert Group on New and Renewable Technologies (EGNRET), Kuala Lumpur, April 26-27 2010. • 6th APEC Bio-fuels Task Force and Related Meetings, Kuala Lumpur, April 28-29 2010.
Publications: (2005 – 2010)	• Rosnah Mat Soom, Astimar Abdul Aziz, Wan Hasamudin Wan Hassan*, Ab Gapor Md Top. Solid-State Characteristics of Microcrystalline Cellulose from Oil Palm Empty Fruit Bunch Fibre. *Journal of Oil Palm Research*, 21(June 2009): 613-620. • Rosnah Mat Soom, Wan Hasamudin Wan Hassan*, Ab Gapor Md Top and Kamarudin Hassan. Thermal Properties of Oil Palm Fibre, Cellulose and Its Derivatives *Journal of Oil Palm Research*, 18(December 2006): 272-277. • Chantara Thavy Ratnam, A. Min Min, T.G. Chuah, A. R. Suraya, Thomas S.Y.Choong and Wan Hasamudin Wan Hassan*. Physical Properties of Polyethylene Modified with CPO *Polymer-Plastics Technology and Engineering, 45(8) 2006: 917-922*. • Min Min A., Medyan R., Wan Hasamudin Wan Hassan*, Chuah T.G., Robiah Y. and Thomas C.S.Y (2005) Application of DSC on Modified Polyethylene using CPO as Bioactive Component *Jurnal Teknologi Terpakai (Journal of Applied Technology)*, 3(1) Mei 2005: 7-13.
Awards and Honors:	• 2004: Silver Medal ITEX – NoSKIN Moulded Particleboard from Oil Palm Biomass • 2005: Bronze Medal ITEX – Carboxymethylcellulose (CMC) from Oil Palm Biomass • 2005: MPOB Director General Annual Research Award – Carboxymethylcellulose (CMC) from Oil Palm Biomass • 2005: MPOB Gold Medal Research Award – Biofuel from Palm Oil

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May 30th – June 4th, 2011, Chiang Mai, Thailand

ACTIVEPARTICIPANTCURRICULUM VITAE

Presenter	
APEC Economy	Mexico
Full Name	José Luis Arvizu Fernández
Academic Title or Position at the Place of Employment	Manager ofbiomass project
Affiliated Organization	Electrical Research Institute
Fields of Interest	• Biological and thermal biomass to energy conversion
Full biography such as experiences, etc.	Chemical Engineer from National University Autonomous of Me Conventional Energy Resources Department of the Electrical Research Institute of Mexico from 1983 until today. I have developed systems, processes, methodologies in the field of biomass to energy from solid, liquid, municipal, agricultural and cattle wastes, by biogas generation and thermal conversion. Some as: • Family Rural Digesters. • Small Farms Type Digesters. • Anaerobic Treatment of Market Waste. • Anaerobic Acidogenic/Methanogenic Treatment of Waste water from Citric Acid Industry. • Anaerobic/Aerobic/Anoxic Treatment of Municipal Waste

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	water. • Laboratory Assays of Urban Solid Waste Samples for Methane Potential of Landfills. • Site FeasibilityAssessment in Landfills and Open Dumps. • National Inventory of Methane Emissions from Waste as a Green House Gas. • Demonstration Process for Electricity Generation withLandfills Methane (in progress). • Gasification of Municipal Solid Waste Conversion.
Research Skills	• Technical Manager of Biomass Projects in the field of Electricity generation from biogas produced in landfills, and Methane Emissions from Waste in the National Green House Gas Emissions Inventory. Model for biofuels development in Mexico.
Presentations:(2005 – 2010)	• Presentations in International and National Workshops on Biomass energy
Publications:(2005 – 2010)	• Book Coauthor “Maize Book” with the subject“Biofuels from maize (2010)”. In Spanish. • Book CoauthorAssociated Coordinator of “Bioenergy in Mexico”. (2005). In Spanish. • Book Coauthor of “On Climatic Change a Vision from Mexico with the subject Historical Registry of the Main Emitting Countries”. In Spanish.

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Presenter	
APEC Economy	Mexico
Full Name	Julián Vega Gregg
Academic Title or Position at the Place of Employment	Secretary General, Project Manager, Consultant
Affiliated Organization	Mexican Bioenergy Network (REMBIO), ETEISA, ECOSYSTEMS
Fields of Interest	• Bioenergy • Biofuels (biogas, bioethanol, biodiesel) • Waste to energy • Sustainability of biofuels
Full biography such as experiences, etc.	• Ing. Julián Vega Gregg is responsible as Secretary General of the REMBIO, to promote and encourage the development of Bioenergy in Mexico. At the same time he represents the Mexican Bioenergy Network in Mexico City. Further activities here involve the promotion of research and technological development on bioenergy. • He has been project leader in projects of renewable energies and energy efficiency helping to reduce thousands of dioxide carbon emissions to the atmosphere.

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	• He participated in the development of a biogas upgrading plant in Germany through membrane technology • Ing. Julián Vega Gregg has also experience in environmental, economic and social sustainability • Ing. Julián Vega Gregg has worked in the development of traditional Clean Development Mechanism projects as well as in the Programme of Activities (PoA) modality • Regarding biogas experience, he has worked in many turnkey biogas projects, elaborating technical and economic feasibility studies, designing and optimizing plants and selecting the best regional substrates for an optimal biogas yield
Research Skills	• Biogas upgrading to Biomethane • Optimization of biogas production • Biogas yield from different feedstock: agricultural substrates, • Energetic plantations • Agro-industial waste
Presentations:(2005 – 2010)	• Julián Vega Gregg - National Meeting of Bioenergy Alternatives: Case of Biofuels, April 2011, Chapingo, Mexico. (Presidium Member and panelist) • Julian Vega Gregg - Biogas: Process, Technology, Applications and potential in Mexico in: "Treatment and Sustainable Management of Natural Resources" May 2011, Veracruz, Mexico. (speaker)
Publications:(2005 – 2010)	• Vega Gregg, J., (2009) Development of a low cost Biogas Plant using livestock waste in Mexico and its Potential, Munich University of Applied Sciences • Vega Gregg, J., (2011) Interview for magazine QUO "Feasability of using biogas of sludge from Wastewater Treatment Plants and potential of its utilization in Mexico.
Awards and Honors:	• Honorific mention for achieving the best grade for the Thesis: Biogas in Mexico - Case Study: Dimensioning of a Biogas Plant in Guanajuato, Mexico

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Presenter	
APEC Economy	Thailand
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Academic Title or Position at the Place of Employment	Senior Vice President – Corporate Planning
Affiliated Organization	Electricity Generating Public Co.Ltd. (EGCO)
Fields of Interest	• Power Generation • Energy
Full biography such as experiences, etc.	Present Positions: • Senior Vice President - Corporate Planning Division, Electricity Generating Public Co.; Ltd. (EGCO) • Director, EGCO Green Co.; Ltd. • Director, Egcomp Tara Co.; Ltd. Experiences: • Managing Director, EGCO Mining Co.; Ltd. • Senior Vice President - Energy Business Division, EGCO • Project Development Manager - Business Development Division, EGCO

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Presenter	
APEC Economy	Thailand
Full Name	Mallika Boonmee
Academic Title or Position at the Place of Employment	Assistant Professor
Affiliated Organization	Khon Kaen University
Fields of Interest	• Bioprocess • Fermentation (lactic acid and ethanol)
Full biography such as experiences, etc.	• 2003-2009 Lecturer Khon Kaen University • 2009 Assistant Professor Khon Kaen University
Research Skills	Analysis of bacterial kinetics
Presentations:(2005 – 2010)	• Prawphan Yuvadetkun, Kunjana Plengsiri, Sutthawan Intarapanich and Mallika Boonmee* (2009) Effects of inoculum concentration and yeast extract for ethanol production from Sweet Sorghum Grain by *Saccharomyces cerevisiae* and *Zymomonas mobilis*. In: *The 19th Annual Conference of Thailand's Chemical Engineering and Applied*

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	Chemistry Society, October 26-27, 2009, Kanchanaburi, Thailand. (Poster presentation) • Buasai Akachart, Narin Kaabbuatong and Mallika Boonmee* (2009) Optimization of enzymatic hydrolysis of cassava starch and lactic acid production from the hydrolyzate by *Lactococcus lactis* NZ133. In: *The 19th Annual Conference of Thailand's Chemical Engineering and Applied Chemistry Society*, October 26-27, 2009, Kanchanaburi, Thailand. (Poster presentation) • Onanong Cotano, Sittipong Amnuaypanich, Nurak Grisadanurak and Mallika Boonmee* (2009) Selection of amine extractant/ diluent system for use in liquid-liquid extraction of lactic acid. In: *The 19th Annual Conference of Thailand's Chemical Engineering and Applied Chemistry Society*, October 26-27, 2009, Kanchanaburi, Thailand. (Poster presentation) • Mallika Boonmee* and Nurak Gridsadanurak (2009) Selection of Extractant/Diluent System to Be Used for Partial Purification of Lactic Acid from Ion Exchange Extractive Fermentation. In: *The 9th Thailand Research Fund Annual Meeting*, October 15-17, 2009, Petchaburi, Thailand. (Poster presentation) • Mallika Boonmee* (2009) Possible Synegistic Effect Between High Lactate and Depletion of Essential Peptides Caused Biomass Reduction During High-Cell Starter Culture Production. In: *The 5th Asian Conference on Lactic Acid Bacteria: Microbes in Disease Prevention & Treatment*, 1-3 July 2009, National University of Singapore, Singapore. (Oral presentation) • Mallika Boonmee* and Sootharwan Intarapanich (2008) Initial Investigation on Acetic Acid Production as Commodity Chemical. In: *The 2nd International Conference on Science and Technology*, 12-13 December 2008, Universiti Tekno-logi MARA, Pulau Pinang, Malaysia. (Oral presentation) • Mallika Boonmee* and Sootharwan Intarapanich (2006) Evaluation of 6 *Acetobacter* spp. for Acetic Acid Production Capabilities. In: *The Proceedings of 18th Annual Meeting of the Thai Society for Biotechnology*. 2–3 November 2006 The Montien Riverside Hotel, Bangkok, Thailand. (Poster presentation) • Mallika Boonmee*, Sootharwan Intarapanich (2006) Significance of Substrate Loss during Fermentation on Product Yield Calculation: a Case Study of Acetic Acid Production, *In: The 16th Annual Conference of Thailand's Chemical Engineering and Applied Chemistry Society*, 26-27 October 2006. Bangkok, Thailand. (Poster presentation) • Mallika Boonmee*, Paweena Pakping and Prapatsorn Polyorat (2006) Effect of shaking conditions on the growth and ethanol production by *Saccharomyces cerevisiae* and *Kluyveromyces marxianus*, *In: The Proceedings of 44th Kasetsart University Annual Conference*. 30 January - 2 February 2006. Bangkok, Thailand. (Poster presentation)
Publications:(2005 – 2010)	• Mallika Boonmee* (2010) Possible synergistic effect between high lactate and insufficient intake of peptides caused biomass reduction during high-cell starter culture production. *Beneficial Microbes*, 1(2): 175-182. • Mallika Boonmee*, Paweena Pakping and Montree

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	Promkudtum (2007) Cultivation in Series for Sequential Conversion of Mixed Glucose/Xylose to Ethanol by *Zymomonas mobilis* and *Candida shehatae*. *Chiang Mai University Journal of Agro-Industry*. 1,29-36. (Thai content) • Mallika Boonmee*, Sootharwan Intarapanich and Onanong Cotano (2008) Effect of Temperature Control on Acetic Acid Production by *Acetobacter* spp. *Naresuan Agriculture Journal*, 11(2), 99-106. (Thai content) • Mallika Boonmee, Noppol Leksawasdi, Wallace Bridge and Peter L. Rogers* (2006) Electrodialysis for Lactate Removal in the Production of the Dairy Starter Culture *Lactococcus lactis* NZ133. International Journal of Food Science and Technology, 42(5), 567–572.
Awards and Honors:	• *Outstanding Poster Presentation*: The 9th Thailand Research Fund Annual Meeting, October 15-17, 2009, Petchaburi, Thailand.

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May 30th – June 4th, 2011, Chiang Mai, Thailand

ACTIVE PARTICIPANT CURRICULUM VITAE

Presenter	
APEC Economy	Thailand
Full Name	Natanee Vorayos
Academic Title or Position at the Place of Employment	Lecturer, Dr.
Affiliated Organization	Chiang Mai University
Fields of Interest	• Life Cycle Assessment & Management • Life Cyle Costing, Renewable Energy • Design of Thermal Systems
Full biography such as experiences, etc.	• 2003-2009 Lecturer Khon Kaen University • 2009 Assistant Professor Khon Kaen University
Research Skills	• Life Cycle Assessment • Design of Thermal Systems
International/Regional Conference: (2005 – 2010)	• Piamdee, J., Vorayos, N., Arjharn, W. and Vorayos, N., 2010, “Potentialities and Environmental Burdens of an Electricity Generated from Dendrothermal Power Plant”, **International Symposium on Low Carbon & Renewable Energy Technology**, November, 2010, Jeju, Korea. • Piamdee, J., Vorayos, N., Kiatsiriroat, T., Arjharn, W. and

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	Vorayos, N., 2009, "Comparison Study on Life Cycle Assessment of Small-Scale Dedrothermal Power Generation for in Thailand including Economic Feasibility Analysis: A case Study", **The World Renewable Energy Congress 2009 – Asia**, May 2009, Bangkok, Thailand. • Vorayos, N., Malawanno, N. and Kiatsiriroat, T., 2009, "Experimental Test of Ethanol Distillation by Direct Boiling in Solar Collector", **The World Renewable Energy Congress 2009 – Asia**, May 2009, Bangkok, Thailand. • Sawangwong, J. and Vorayos, N., 2008, "Feasibility Analysis of Waste – to –Energy Facility in Chiang Mai, Thailand", **The 15th Tri-University International Joint Seminar and Symposium 2008**, October 2008, Zhenjiang, China. • Vorayos, N., Vorayos, N. and Kiatsiriroat, T., 2007, " Life cycle assessment of dendrothermal power generation in Thailand including economic feasibility analysis: A case study", **The International Life Cycle Assessment and Life Cycle Management Conference**, October, 2007, Portland, Oregon, USA. • Chamsilpa, M., Vorayos, N. and Kiatsiriroat, T., 2007, "Energy and environmental assessment over the entire life of amorphous silicon solar module for a solar cell power plant", **Asian-Pacific Regional Conference on Practical Environmental Technologies: APRC 2007**, August, 2007, Kon Kaen. • Chamsilpa M., Vorayos N., and Kiatsiriroat T., 2006, "Life Cycle Inventory of Amorphous Silicon Solar Module Manufacturing Using Activity-Based Approach", **Int. Conf. on Green and Sustainable Innovation ICGSI 2006**, November 2006**,** Chiang Mai.
International/Regional Journal: (2005 – 2010)	• Chamsilpa, M., Vorayos, N. and Kiatsiriroat, T., 2010, "Environmental impact analysis of solar cell power plant compared with fossil fuel power plants in Thailand", **Asian Journal on Energy and Environment**, 11(2), pp. 103 – 117. • Nuntaphan, A., Vithayasai, S., Vorayos, N., Vorayos, N. and Kiatsiriroat, T., 2010, "Use of Oscillating Heat Pipe Technique as Extended Surface in Wire-on-Tube Heat Exchanger for Heat Transfer Enhancement", **International Communications in Heat and Mass Transfer**, 37, pp. 287 – 292. • Thongwik, S. , Vorayos , N., Kiatsiriroat, T. and Nuntaphan, A., 2008, "Thermal Analysis of Slurry Ice Production System Using Direct Contact Heat Transfer of Carbon Dioxide and Water Mixture", **International Communications in Heat and Mass Transfer,** 35 (6), pp. 756-761. • Vorayos, N., Kiatsiriroat, T. and Vorayos, N., 2006, "Performance Analysis of Solar Ethanol Distillation", **International Journal of Renewable Energy**, 31 (15), December, pp. 2543 – 2554. • Vorayos, N., Kiatsiriroat, T. and Vorayos, N., 2006, "Performance Analysis and Economic Consideration of Solar Ethanol Distillation with Different Types of Solar Collectors", **International Journal of Ambient Energy**, 27 (1), January, pp. 3 -14.
Awards and Honors:	• *Best Paper Award*: International Symposium on Low Carbon & Renewable Energy Technology, November, 2010, Jeju, Korea.

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KEYNOTE SPEAKER CURRICULUM VITAE

Presenter	
APEC Economy	Thailand
Full Name	Wongkot Wongsapai
Academic Title or Position at the Place of Employment	Assistant Professor
Affiliated Organization	Chiang Mai University
Fields of Interest	• Renewable Energy • Energy Efficiency • Energy Policy and Modeling • Greenhouse Gases management
Full biography such as experiences, etc.	• 1995 Lecturer Chiang Mai University • 2001 Assistant Professor Chiang Mai University
Research Skills	• 2001 -2006 Head School of Biochemical Engineering School *P. Universidad Católica de Valparaíso* • Energy Modeling, Renewable energy management, Energy Policy, Greenhouse gas management

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Presentations:(2005 – 2010)	• Wongkot Wongsapai, "THAILAND ENERGY SITUATION: PROSPECTIVE FOR FUTURE", *International Conference on Green and Sustainable Innovation 2006*, November 29-December 1, 2006, Chiang Mai, Thailand. • Wongkot Wongsapai and Pongsiri Jaruyanon, "STRATEGIC PROVINCIAL ENERGY PLANNING: THE CASE OF LOWER NORTHERN REGION OF THAILAND", *International Conference on Green and Sustainable Innovation 2006*, November 29- December 1, 2006, Chiang Mai, Thailand. • Wongkot Wongsapai and Ram M.Shrestha, "INTERFUEL SUBSTITUTION RESPONSES IN THAI ECONOMY, " *International Conference on Green and Sustainable Innovation 2006*, November 29- December 1, 2006, Chiang Mai, Thailand. • Wongkot Wongsapai and Pongsiri Jaruyanon , THAILAND MUNICIPAL ENERGY PLANNING: THE CASE OF KHANU CITY, *PSU-UNS International Conference on Engineering and Environment - ICEE-2007*, Phuket May10-11, 2007, Prince of Songkla University, Faculty of Engineering, Hat Yai, Songkhla, Thailand • Wongkot Wongsapai and Pongsiri Jaruyanon , EVALUATION OF ENERGY EFFICIENCY IMPROVEMENT IN SIX MANUFACTURING SECTORS: THE CASE OF SMEs IN THAILAND, *PSU-UNS International Conference on Engineering and Environment - ICEE-2007*, Phuket May10-11, 2007, Prince of Songkla University, Faculty of Engineering, Hat Yai, Songkhla, Thailand • Wongkot Wongsapai and Pongsiri Jaruyanon , EVALUATION OF BOILER EFFICIENCY IMPROVEMENT PROGRAMS FOR SMALL AND MEDIUM FACTORIES; *The 6th Asia Pacific Conference on Sustainable Energy and Environmental Technologies*, Bangkok, Thailand. May 6-11, 2007 • Songwut Penphanussak and Wongkot Wongsapai, 2008, The Demand for Energy in Manufacturing Sector of Thailand, Paper presented in "*Technology and Innovation for Sustainable Development Conference (TISD2008)",* 28-29 January 2008, Khon Kaen, Thailand. • Sarawut Polvongsri and Wongkot Wongsapai, 2008, The Long-Term Energy Demand For Thailand Under MAED, Paper presented in *"Technology and Innovation for Sustainable Development Conference (TISD2008)",* 28-29 January 2008, Khon Kaen, Thailand. • Auswin Keawsing and Wongkot Wongsapai, 2008, The Rebound Effect Estimation and Interfactor Responses in Thai Manufacturing Sector, Paper presented in *"Technology and Innovation for Sustainable Development Conference (TISD2008)",* 28-29 January 2008, Khon Kaen, Thailand. • Wongkot Wongsapai, Poon Thienburanathum and Prasert Rerkkriengkrai, 2008, Biogas Situation and Development in Thai Swine Farm, Paper presented in "*International Conference on Renewable Energy and Power Quality (ICREPQ'08)",* 12-14 March 2008,, Santander, Spain. • Pongsiri Jaruyanon and Wongkot Wongsapai, 2008, Biodiesel Technology and Management From Used Cooking Oil in Thailand Rural Areas, Paper presented in *"International Conference on Renewable Energy and Power Quality (ICREPQ'08)",* 12-14 March 2008,, Santander, Spain.

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	• Wongkot Wongsapai, 2008, Energy Policy and Situation under the Applications of the Sustainable Economy Principle: The Case of Thailand, Paper presented in *"The 13th IIES International Oil & Gas Conference Energy Management; Policies and Experiences"*, November 30 - December 1, 2008, Tehran, I.R. Iran. • Waraporn Eakpaopan and Wongkot Wongsapai, "Renewable Energy Demand and Supply Forecast Under Long-term National Renewable Energy Development Plan: The case of Thailand", *World Renewable Energy Congress 2009 – Asia, The 3rd International Conference on "Sustainable Energy and Environment (SEE 2009)"*, 18-23 May 2009, Bangkok, Thailand. • Thirayu Pinthong and Wongkot Wongsapai, "Evaluation of Energy Demand and Supply Under Electricity Generation Scenarios of Thailand", *World Renewable Energy Congress 2009 – Asia, The 3rd International Conference on "Sustainable Energy and Environment (SEE 2009)",* 18-23 May 2009, Bangkok, Thailand • Yasintinee Aimyuak and Wongkot Wongsapai, "The Analysis of Energy Saving and Carbon Dioxide Emissions in Thai Manufacturing Sector by Decomposition Technique", *World Renewable Energy Congress 2009 – Asia, The 3rd International Conference on "Sustainable Energy and Environment (SEE 2009)",* 18-23 May 2009, Bangkok, Thailand • Wongkot Wongsapai and Pruk Aggarangsi, "The Economic Evaluation of the Performance-Base Tax Incentive for Energy Efficiency and Renewable Energy Project in Thailand" *International Conference on Green and Sustainable Innovation 2009*, December 2 – 4, 2009, Chiang Rai, Thailand. • Krissana Romchaiyaphruk,Wongkot Wongsapai and Itthichai Preechawuttipong**,** "The Assessment of Carbon Dioxide Elasticity by Using the Input-output Method: The case of Thailand", *The 3rd Technology and Innovation for Sustainable Development International Conference (TISD2010),* Faculty of Engineering, Khon Kaen University, Thailand, 4-6 March 2010.
Publications:(2005 – 2010)	Wongkot Wongsapai, Yasintinee Aimyuak and Chatchawan Chaichana, "The Analysis of Energy Saving and Carbon Dioxide Emissions in Thai Manufacturing Sector by Decomposition Technique"**,** *Journal of Sustainable Energy & Environment (JSEE)* (to be printed)
Awards and Honors:	• National Committee and Working Group on Thailand Renewable Energy Development Plant: Bioenergy section • National Committee on Thailand Atomic Policy and Strategy Plan Development • National Committee on Thailand-IAEA collaboration project

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KEYNOTE SPEAKER CURRICULUM VITAE

Presenter	
APEC Economy	U.S.A
Full Name	Dr. Kulinda Davis, PhD
Academic Title or Position at the Place of Employment	Director – Grain Processing Enzymes
Affiliated Organization	Verenium Corporation
Fields of Interest	• Ethanol Production
Full biography such as experiences,	• Dr. Kulinda Davis is responsible for the grain processing enzyme segment of Verenium Corporation – a major player in the enzymes industry. • Dr. Kulinda Davis has a track record in the initiation of biofuel projects. She secured competitive multi –million U.S. government grants and developing strategic partnerships to execute biofuel project development. She is currently a Product Director at Verenium Corporation with direct oversight for a multimillion dollar enzyme product targeted at the biofuels industry.

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	• Two projects she initiated at Sapphire Energy ($135M)and Pacific Ethanol ($25M) are on-going and are proudly profiled in the DOE funded portfolio of projects aimed at achieving the U.S. national goals for biofuel development. She is in the U.S under the prestigious National Interest Waiver status recognized for her contributions to securing America's energy future.
Research Skills	• Management and technical experience working in cellulose- and algae –based fuel development processes spanning feasibility studies, planning and design of advanced biofuel projects; development and implementation of corporate strategy and R&D and operational strategies • Project management skills applied in the renewable fuel space including managing cross-disciplinary projects involving microbiology, engineering, construction , operations and business development to ensure effective integration of work teams • Identification of unit operation performance indicators and managing optimization efforts that facilitate proactive technical and business analysis • Comprehensive experience of a wide variety of microbiological and analytical techniques as applied to fermentation systems including , bioreactor operations, HPLC and GC operations
Presentations:(2005 – 2010)	• Kulinda Davis, PhD. "Demonstration scale cellulosic ethanol production technology" Harvesting Clean Energy Conference VIII – Portland, OR. January 2008. • Kulinda Davis, Panelist Optimizing Ethanol Plants "Next Generation Technologies: What are the Opportunities for Plant Optimization Projects". May 2008, Minneapolis, MN.
Publications:(2005 – 2010)	• Davis, L., Svenson, C., Pearce, J., Rogers, P. "Evaluation of *Zymomonas*-based ethanol production from a hydrolysed waste starch stream." Biomass and Bioenergy, 30 Issues 8/9 (2006) pp. 809-814. • Davis, L., Y-J, Jeon, Svenson, C., Pearce, J., Rogers, P. "Evaluation of wheat stillage for ethanol production by recombinant *Zymomonasmobilis*." Biomass and Bioenergy, 29 (2005) pp. 49-59.
Awards and Honors:	$ 75 million secured funding from the U.S. Department of Energy for biofuel projects Responsible for $ 20M in revenue in current position

Presentation:	
Title	A Novel Thermostable Broad pH Alpha -Amylasefor Ethanol Production
Abstract (up to 200 characters)	Verenium Corporation's Fuelzyme® alpha-amylase is an archaeal α-amylase which possesses unique characteristics that make this enzyme extremely useful for the ethanol industry. The uniqueness of this enzyme resides in its ability to function in acidic pHs and high temperatures, causing a fast breakdown of the starch substrate that results in a sharp reduction in viscosity. Fuelzyme® provides an opportunity to increase the solids loading in production, resulting in higher ethanol yields. Fuelzyme® remains active in hotter liquefaction conditions of 194°F-212°F (90-100°C)

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	and at broad pH range 4.9-5.4, at which Bacillus α-amylases are inactive. Fuelzyme® can be used in processes utilizing a wide range of starchy grains such as corn, milo, wheat or blends. Using high throughput, multi-step screening techniques, genomic libraries prepared from environmental DNA, enrichments, and isolates, were surveyed for the production of high temperature, low pH α-amylases. Three clones likely to be from the archaeal order *Thermococales* were obtained which had the desired characteristics. A chimerical gene produced from a reassembly of restriction fragments from these original clones provided improved expression and activity. The reassembled gene has been used for industrial production of Fuelzyme®.

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ACTIVE PARTICIPANT CURRICULUM VITAE

Presenter	
APEC Economy	Perú
Full Name	Ari Loebl
Academic Title	Mechanical-Electrical Engineer
Affiliated Organization	Biofuels Committee at the National Society of Industries of Peru
Fields of Interest	• Development of Biofuels
Full biography such as experiences, etc.	• 1966 -1976 Lecturer – Physics and Science University of Engineering, Lima-Peru • 1976 to present Presentations and Conferences in different Universities and Organizations in Perú • President of the Biofuels Special Committee of the National Society of Industries of Perú • President of various Companies and Organizations in Perú and in the USA
Research Skills	• Power generation
Presentations:(2008 – 2010)	• Various presentations, Lectures related to Biofuels in Perú and worldwide in numerous Universities and organizations throughout the country and Articles and Press Conferences regarding Biofuels
Publications:(2008 – 2010)	• Various Articles and essays published in Perú and in some worldwide distributed specialized press releases.

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ACTIVE PARTICIPANT CURRICULUM VITAE

Presenter	
APEC Economy	Peru
Full Name	Pedro Gamio
Academic Title or Position at the Place of Employment	Lawyer / Regional Manager GVEP International
Fields of Interest	• Energy • Biofuels • Hydrocarbons • Renewals
Full biography such as experiences, etc.	• 2007-2008 ViceMinister of Energy PERU • 2009- to date Regional Manager GVEP Intl • 2000 – to date Main Coordinator and Teacher Universidad Católica del Perú
Research Skills	• Petroleum and Renewal Energy
Presentations:(2005 – 2010)	Presentation 2010: • National Forum of Renewable Energy, Lima • Forum: Energies In Peru: Where Are We going, Organized by The Ebert Foundation Arequipa, Lima

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	• Renewable Energies, fundación Ebert,Lima • Development Cooperation ITDG, Lima • Macro Regions, Cusco • Renewable Energies Fundación Ebert, Piura • Regional Hydroelectrics, Cusco • Society and Energy Strategy World Bank, Lima Miraflores • IV COBER, Lima • Simposium : Solar Energy – APES, Cusco Presentation 2009: • VIII Cientific International Encounter • Clean Air and Sulfure Reduction in Diesel • More Clean Energy, Finland Embassy, Lima • Participación en el - XV Simposium: Solar Energy Cajamarca, Perú Presentation 2008: • 46º Annual Conference CADE • Hydroelectric Development • VI International Congress of Exploration of Petroleum and Gas– VI INGEPET • Security and Diversification of Energetic Matrix- New Opportunities, Latin America • Latest Technologies by General Electric Energy. • Transparency in the Extractive Industry– EITI • "EXPOPERÚ 2008" – Entrepreneur Encounter, Brasil • Energetic Crisis. Opportunities and Challenges for for Latin America • I Forum of Investment ALC/UE, Latin America, Caribbean and European Union Countries • APERC Annual Conference 2008, Japan • Technology Scenarios for Clena Energy in Americas IDB International Development Bank Washington, EEUU • International Seminarium: Natural Resources and Democracy in Latin America
Publications:(1995 – 2011)	• Book: "Petroleum, Environment and Indian Communities" Edit 2001 • Book: "Energy: Where are we going?" Edit 2010 / Friederich Ebert Foundation • Publications, Articles and Essays during the last 15 years in local media: El Comercio, Gestión, Claridad and other.
Awards and Honors:	• Outsanding Citizen and Keys of the City Cajamarca – Perú • Outsanding Citizen and Keys of the City La Matanza, Piura - Perú

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ACTIVE PARTICIPANT CURRICULUM VITAE

Presenter	
APEC Economy	Viet Nam
Full Name	Doan Thai Hoa
Academic Title or Position at the Place of Employment	Assoc. Prof. Dr.,
Affiliated Organization	Ha Noi University of Technology
Fields of Interest	Bioefinery
Full biography such as experiences, etc.	• Teaching and doing research on Pulp and Paper Technology and Biorefinery
Research Skills	• Pulp and Paper Technology • Bio-ethanol production from biomass • Waste water treatment
Presentations:(2005 – 2010)	• Presentations in 4 International and National Workshops on Biomass • 5th National Conference on Chemistry "Vietnam Chemistry for Sustainable Development"
Publications:(2005 – 2010)	**2010 onwards on biofuels**: • Doan Thai Hoa, Nguyen ThiThanhNga, Tran Dinh Man and Nguyen The Trang, J. Chemistry, Vietnam Academy of Science and Technology, *Study on Acid Pretreatment of Acacia mangium for Improving Enzymatic Hydrolysis in Bioethanol Production*, Vol. 48, No.4A, pp.534-539 (2010).

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	• Nguyen Hoang Chung and Doan Thai Hoa, J. Science &Technology Technical Universities, *Study on Pretreatment of Woody* (*Acacia mangium)Waste by Liquid Hot Water and Ball-Milling* (in press, 2011). • Le Mai Oanh and Doan Thai Hoa, J. Science & Technology Technical Universities, *Study on Alkaline Pretreatment of Woody* (*Eucalyptus urophylla) Wastefor Enzymatic Hydrolysis in Bioethanol Production* (in press, 2011).
Awards and Honors:	

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ACTIVE PARTICIPANT CURRICULUM VITAE

Presenter	
APEC Economy	Vietnam
Full Name	Nguyen Duc Cuong
Academic Title or Position at the Place of Employment	Director of Center for Renewable Energy and Clean Development Mechanism
Affiliated Organization	Institute of Energy, Ministry of Industry and Trade
Fields of Interest	Renewable Energy (biomass Technologies)
Full biography such as experiences, etc.	• 1983 - 1997 Energy Researcher, Institute of Energy • 1998 - 2004 Director of Biomass energy and Rural Energy Department, Institute of Energy • 2005 -2011 Director on Center for Renewable Energy & CDM, Institute of Energy
Research Skills	• Design and analysis on rural energy (technologies, planning, policies)
Presentations:(2010 – 2011)	• Renewable Energy and Environment of Vietnam, the 11th Meeting of Head ASEAN Power Utilities/Authorities (HAPUA) Working Committee 9-11 November 2009 in Thailand • Status of Renewable Energy base on International Cooperation Projects and it’s analysis, the Eighth Asia Biomass Seminar, February 3 to 9, 2010 in Tokyo, Japan

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	• EAS Asia Biomass Seminar Follow Up Workshop Mach 8, 2010 in Hanoi, Vietnam • Strategy, Master Plan on Renewable Energy Development of Vietnam up to the year 2015, with outlook to 2025, Hanoi, May, 2010 • Development and Demonstration of Biomass Multi Fuel Supply Chain for Power Plants and Industrial Boilers in Vietnam, • ADB RETA 7575: Determining the Potential of Carbon Capture and Storage in Southeast Asia, 17 Jan. 2011, Hanoi, Vietnam • Review and update of the mobilization of GEF projects in Viet Nam and REVIEW OF CLIMATE CHANGE PORTFOLIO FOR VIET NAM, 18, August 2010, Hanoi, Vietnam
National Publications:(2009 – 2010)	• Strategy, Master Plan on Renewable Energy Development of Vietnam up to the year 2015, with outlook to 2025, Hanoi, March 2010 • Impacts Assessment of Climate Change on Energy Sector of Vietnam and Proposal Adaptation, Hanoi, may 2010. • Development and establishment of incentive mechanism for Wind energy development in Vietnam (Feed-in tariff), Hanoi, September 2010
International Publications	• N.D.Cuong, P.K.Toan, et.al. (2002), “Improvement and Development of Biomass Briquetting Systems and Briquette Stoves in Vietnam”, GLOW- Vol. 26, The Asia Regional Cookstove Program, Indonesia. • N.D.Cuong, et.al. (2001) “Application of LEAP for Wood Energy Planning in Vietnam”, Wood Energy News, Vol.16 No2 Regional Wood Energy Development Program in Asia, Bangkok, Thailand.
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ACTIVE PARTICIPANT **CURRICULUM VITAE**

Presenter	
APEC Economy	Philippines
Full Name	Carmelita Garcia Hansel
Academic Title or Position at the Place of Employment	Acting Director of Research; Professor
Affiliated Organization	Mamitua Saber Research Center/ Department of Biology, Mindanao State University
Fields of Interest	• Environmental and natural resource management • Plant biodiversity • Limnology • Microbiology
Full biography such as experiences, etc.	Present Positions: • Officer-in-Charge – Director of Research, Mamitua Saber Research Center (from 2006) • Professor, Department of Biology (from 1992) Past Positions: • College Research Coordinator, College of Natural Sciences and Mathematics, 2003-2006 • Dean, College of Natural Sciences and Mathematics, 1993-1996 • Chair, Department of Biology

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Research Skills (from 2005)	• Supervisor of undergraduate theses-directed projects • Project Director, *Development of alternative energy, food and livelihood sources towards sustainable management of the Bayug River Watershed, Brgy. Rogongon, Iligan City*, Dec. 2006 – June 2008 • Member of Research Team, *Investigation into the reported unusual greening of Lake Lanao*, October – December 2006 • Study Leader, *Assessment of the headwaters of Layawan River: linkage between the terrestrial and aquatic ecosystems in the Oroquieta watershed of Mt. Malindang, Misamis Occidental*, October 2004-May 2005 • Research collaborator, *Soil ecological diversity and relevant interrelationships of critical resources in Mt. Malindang, Misamis Occidental*, January 2004 – May 2005 • Researcher, *Comparative assessment of Langaran and Layawan Rivers*, September 2003 – May 2005
Selected Presentations: (2005 – 2010)	• Carmelita G. Hansel (2009) No-tillage perennial alternative food crops. In: *The Sixteenth Annual ECHO (Educational Concerns for Hunger Organization) Agricultural Conference*, 8-10 December 2009, Crowne Plaza Hotel, Fort Myers, Florida, U.S.A. (Oral presentation) • Carmelita G. Hansel (2009) Alternative energy, food and livelihood sources for forest conservation and climate change mitigation. In: *The 2nd World Congress of Agroforestry*, 23-28 August 2009, Nairobi, Kenya. (Poster presentation) • Julieta P. Lagmay, Carmelita G. Hansel* *et al*. (2007) Hydrobiological assessment of Lake Lanao subsequent to its unusual greening. In: *The 39th Annual Convention of the Federation of Institutions for Marine and Freshwater Sciences (FIMFS)*, 29-31 October 2007, Visayas State University, Baybay City, Leyte, Philippines. (Oral presentation) • Carmelita G. Hansel* *et al*. (2005) Assessment of the headwaters of Layawan River. In: *The Joint 37th Annual Convention of FIMFS and 10th Annual Zonal Research Development & Extension (RD&E) Review and Science and Technology (S&T) Planning Workshop of the Philippine Council for Aquatic and Marine Resources Research and Development (PCAMRD) Zonal Center IV*, 13-15 October 2005, Mindanao State University at Naawan, Naawan, Misamis Oriental, Philippines. (Oral presentation)
Publications: (2005 – 2010)	• Lagmay, Juliet P., C.G. Hansel, *et al*. 2006. Hydrobiological assessment of Lake Lanao subsequent to its unusual greening in 2006. *Mindanao Journal* 29: 85-100.Hansel, C.G., *et al*. 2006. Assessing the headwaters of • Layawan River: linkage between the terrestrial and aquatic ecosystems in Mt. Malindang, Misamis Occidental. BRP Monograph Series No. 14. BRP, SEAMEO SEARCA, College, Laguna. 62 pp. • Boniao, R.D., RV B. Estoista, R. De Goede, C.G. Hansel, et al. 2006. Soil ecological diversity and relevant interrela tionships of critical resources in Mt. Malindang, Misamis Occidental. BRP Monograph Series No. 11. BRP, SEAMEO SEARCA, College, Laguna. 77 pp. • Gorospe-Villarino, A., DG Bacaltos, E. Roa, C.G. Hansel, *et al*.

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