PROJECT COMPLETION REPORT FOR THE

EEP 3-V-053: Biodiesel Production from closed-algae growing systems using wastewater of Ethanol Plant in Vietnam

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ABBREVIATIONS AND ACRONYMS

NLU: Nong Lam University of Ho Chi Minh city
GFC: Dong Xanh company
SYKE: Finish Environment Institute
ULB: Universite Libre de Bruxelles
EEP: Energy and Environment Partnership-Mekong
LCP: Low cost Photobioreactor
PBR: Photobioreactor
ChEPro: Chemical Engineering and Processing Department, NLU

BASIC PROJECT DATA:

Project Title:	Biodiesel Production from closed-algae growing systems using wastewater of Ethanol Plant in Vietnam
Contract number:	

Partners and budget contributions:

	Partners	**Euro**	**%**
1	Lead Applicant (NLU)	18,000	10
2	Dong Xanh company (GFC)	34,200	19
3	Finish Environment Institute (SYKE)	9,000	5
4	Universite Libre de Bruxelles (ULB)	1,800	1
5	EEP	117,000	65
	Total cost	**180,000**	100%

Country (ies)	***Technical focus***	***Main activity***
Vietnam	*Bio-fuels*	*Pilot Project*

Intended beneficiaries / target groups:		*No. Male*	*No. Female*	*Total*
1	The people who use biodiesel for transport in Viet nam in the future			Vietnamese people
2	The poor who work in the fields of algae growing or in biodiesel processing plants (700 m^2 land)	15	10	25
3	Researchers, policymakers, Cassava Ethanol plants, and service providers in this area			

Project Purpose:	Building a pilot biodiesel producing system including low cost photobioreactors for growing of microalgae using wastewater and CO_2source producing from cassava ethanol plant as well as biodiesel processing facilities for process analysis and scale-up.
Project Overall Objective:	Development a technique for producing of biodiesel from microalgae using wastewater and photobioreactor as well as to assess the cost of biodiesel and potential of using microalgae as an alternative source of vegetal oil for biodiesel.
Project Duration :	**Start Date:** 9/2011 **End Date**: 11/2012 **Total number of months:** 15
	Revised end date: 9/2013

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EXECUTIVE SUMMARY

Since the project commencement date of Sept 2011, the low cost tubular photobioreactors (LCP) of the volume 40 to 400 liters were started to design and manufactured at NLU. The LCP40L-6 photobioreactor was developed for outdoor stress treatment experiments. It consisted of 6 columns with the volume of 40 liters each. The photobioreactor (PBR) of 170 liter volume was designed for tube size 70mm, the other PBRs of tube sizes 140, 170 and 210mm were designed with volume of 400 liter at NLU.

The PBRs were tested at NLU for comparison of PBR performance between tube sizes in outdoor condition. The comparison of PBR performance is the base to select the best system to be scaled up to the higher volume PBRs (LCP2500, LCP6000 and LCP8000) and then the 500 m^2 system using wastewater of ethanol plant. The tests were carried for at least the dry and wet seasons of the year.

The experiments of PBRs with volume of 400 liter at NLU were conducted outdoor condition. In the previous tests (Jan-July 2012), the performance of PBRs of tube size of 140mm and 170mm were better than the tube size of 210mm, i.e., oil capacities (g/m^2) were 3.27, 5.75, and 5.6 for 210, 170 and 140 mm tube size, respectively. Therefore, continuation to find the methodology of algae growth in PBRs of tube size 140 and 170mm was carried out during the period of August to December 2012. The comparison between PVC, PP and PE materials of the tube was also carried out for 210mm tube size. Finally, the PBRs with tube size of 170mm using PP plastic material was selected for scaling up to 500m2 growing system.

During Aug 2012 Dec 2012, the strain selection was conducted for waste water using both marine and fresh *Chlorella* algae. The marine *Chlorella* was found to produce high content of triglycerides suitable for transesterification to convert them to methyl esters for biodiesel. However, for the growing of algae, adding of some salts to the waste water has been done to approach the sea water. This modification led to high salinity of water after growing affecting the environment. The fresh *Chlorella vulgaris* was then tested with waste water. This strain grown well in wastewater producing high content of hydrocarbons rather than triglycerides. These hydrocarbons were found to be used directly as fuel without transesterifiaction to convert to methyl esters as required for triglycerides.

Some trials of combination of methodology of growth in PBR with stress treatment were conducted to see that oil content can be improved by 40% outdoor but low productivity. This result recorded for the period of August to December 2012 and was found to be similar to the results recorded for the period of January to July 2012. In other words, for the climate condition in Ho Chi Minh city of Vietnam, the current PBRs systems got low productivity during stress treatment. The main reasons for this low productivity were low light/dark circle and the transparency of the PP plastic material used for the tube. Some reports in the literatures said that the best condition for algae to grow is 24h supply of light with a constant light intensity (Eduardo Jascob-Lopes et al, 2009). The period of light in our system was less than 10h/day (7AM to 17PM). The total energy provided by sun in a day is more than the requirement for algal growth, but the duration of the light period is not suitable. Therefore, further modification of the PBR technology should be considered in terms of tube material, supply of light period, and growing methodology.

By the end of 2012, the PBR was scaled-up from 400 liters to 2500 liters of the tube volume or solar receiver, so called LCP2500 in which the volume of degas tank was 500 liters. The LCP2500 had total volume of 3000 liters and total energy used for pumping and degas was 262W. This step is important before scaling up to 500m^2 system.

The fluorescent spectrophotometer FS8000 funded by EEP-Mekong has been installed in April 2012 for mainly determination of the crude oil in algal cell. Staring Jan 2013, the treatment of algae by spectral light using fluorescent spectrophotometer FS8000 was conducted to isolate a new strain of *Chlorella vulgaris* with better oil and biomass. This new strain has been tested for small volume of 3 liters indoor resulted initially an improvement of oil content by a factor of 1.4-1.5 times compared to the control. However, this improvement was not significant for outdoor. This situation is similar to what we have met with stress treatment by nitrogen deprivation for outdoor condition. The problem may come from that the growing condition outdoor for this strain under current PBR system is not good enough as indoor.

Scaling up to industrial scale of PBR was carried out for 6000-28000 liter system. The LCP6000 and 8000 worked properly to obtain biomass of 0.5g/L. Testing of different growing methodologies in PBR under outdoor conditions to optimise the growing process using LCP6000 and 8000 systems was conducted. From this, a pilot algal growing system of 500m2 land was designed including 2 LCP6000, 2 LCP8000 for growing and 2 LCP400 for nursery. The PBRs for 500m2 land was manufactured during April-June 2013 and installed at the experimental site of ChEPro (NLU) during June-July 2013.

It has been reported previously that our collaborator (GFC) got the difficulty in their business and could not effort to complete the downstream system as planned in the proposal. However, GFC has done something such as preparation of land, purchase the dryer, centrifuge, and oil press. We have already asked other ethanol plant - the Xuan Loc plant- in Dong Nai province to overcome this situation. We have come to Xuan Loc company to collect the wastewater for analysis and for algal growing experiments. The results were given in the previous semi-annual reports. However, the process for replacing the GFC by Xuan Loc company was not simple. Therefore, we decided to install the 500m^2 pilot PBR at the experimental site of ChEPro (NLU) for growing experiments using wastewater model (not real wastewater) developed in this project. So far, for large scale, we have already installed the growing system, harvesting by centrifuge or sedimentation containers, and drying facilities. Comparison to the original plan, we didn't have the extraction and refining equipments for large scale system but we did have these facilities for lab scale. In addition, through out of the project, we have considered either wet extraction or dry extraction for reduction of the energy. The original plan designed the extraction equipment based on the dry extraction method, however, the wet extraction appeared to be more efficient due to less energy. Even though the selection of wet or dry extraction method is still under consideration, without the dry extraction equipment would be purchased by GFC, the progress on extraction activities was running well.

Trials of algae growing in LED light resulted higher oil content (by a factor of 2.87) and less energy consumption compared to normal fluorescent light for indoor condition. This is a good result contributed to the methodology of algal growing for high oil content in biodiesel production. The application of LED light producing from waste for outdoor growing of algae should be considered in the future.

Trials of harvesting for large quantity of cultured broth were conducted. The flocculation in combination to centrifuge was tested successfully to reduce energy consumption of harvesting. The sedimentation of algae for 2 days under pH11 reduced the electrical energy consumption of centrifuge by a factor of 50 times and increased oil content by a factor of 2.60.

Testing of biodiesel using small engine was tried initially for some batches with good results. More experiments on this area should be conducted to conclude about the use of algal biodiesel.

Treatment of the spent medium for discharging to the environment was carried out. The treated water can be removed to the environment safely.

Cost analysis was carried out for the current production system and for the assumed system. By sedimentation method, harvesting has been reduced to very low contribution 0.2-0.6% of total cost. Application of technology such as wet extraction, using waste for nutrients, free CO2 source, stress treatment under LED light, can reduce contribution cost of growing part from 79.8% to 48.7% of total resulted in cost reduction from USD129/kg oil to USD14.3/kg oil. At this production cost (USD14.3/kg oil), extraction and refining part occupied 50.7% of total become a technical to be considered in the future to reduce further the production cost.

Proposal on the production process to produce biodiesel from algae was carryout based on the results obtained in this project. The necessary modifications were highlighted for the next research in this area.

The main activities carried out of this project are listed below:

- Analysis of wastewater chemical composition
- Analysis of composition of Chlorella vulgaris algae grown in normal condition
- Design and manufacture of low cost PBRs (Sep 2011-Jan 2012)
- Testing the performance of 400 liter PBRs (Jan-July 2012)
- Stress treatment indoor condition and outdoor in combination with growth methodology in 400 liter PBRs during dry season (Jan-July 2012)

- Wet extraction trials for reducing of energy from removing of drying in dry extraction method
- Cost analysis for comparison between tube sizes of small scale PBRs
- Installation of Fluorescence Spectrophotometer (FS-meter) in April 2012
- Trip of Spilling Kristian, a scientist of SYKE to NLU in Feb 2012
- Trip to Finland of Vietnamese project leaders and partners in June 2012
- Planning with GFC for land preparation and CO_2 supply system
- Monitoring of EEP Mekong in Dec 2011 and Feb 2012
- Finland Ambassador in Hanoi, Mr. Kimmo Lahdevirta and his wife visited the experimental site for EEP-3-V-053 of Chemical Engineering Department, NLU on 18/9/2012
- Testing of different growing methodologies in PBR under outdoor conditions to optimise the growing process for the wet season. Continue to conduct stress treatment outdoor in combination with growth methodology in PBRs (Aug-Dec 2012).
- Some initial results of this project has been presented at the 5th Algae World Asia, Singapore, 7-9 Nov 2012
- Extraction trials for reducing of energy using maceration and wet extraction method
- Refinery of oil/hydrocarbons in crude oil produced from algae and analysis of chemical composition of refined oil
- Testing of algae growth in wastewater and wastewater model
- Design and manufacture of PBRs of 2500 liters, and growing test of algae in PBRs of 2500 liters (Jan-March 2013)
- Installation of the centrifuge 200L/h and 20L/h for harvesting
- Installation of the tank for sedimentation of algae (20 units x 50 liter/unit = 1000liter). Harvesting trials using flocculation and sedimentation to save centrifuge energy (May 2013)
- Design and manufacture of PBRs of 6000 liters, and growing test of algae in PBRs of 6000 liters (Apr-July 2013)
- Scaling up to 500m^2 system (25000-30000L) in Aug-Sep 2013.
- Environmental issues of the spent medium: spent media can be discharged to the environment safely.
- Capacity building: Training of 12 Bachelors (Sep 2011 to July 2012) and 10 Bachelors (Aug 2012 to Sep 2013) and 3 Master students of NLU on related area.
- Report writing: the semi-annual reports and final reports were submitted.

The technical team of EEP-Mekong including Mr. Christer Nyman and Mr. Mikael Forss has visited our experimental site on 29 June 2013 and discussed about the progress of the project. Based on the actual situation of GFC, we have agreed to adjust the budget of this project to fit the actual contribution of each partners.

For summary, the following results have been achieved from this project:

1. Development of low cost photobioreactor simple in construction, easy to operate with low energy consumption. The 6000 liter PBR worked properly produced algae without contamination. The systems can be scaled up easily to 500m2 system and is the base for higher area.
2. Finding three new technologies for oil enhancement in algae, i.e., spectral light treatment the algae before growing, nutrient stress under LED light, and sedimentation stress at pH11 during harvesting, with oil improved factors of 1.45, 2.87 and 2.6, respectively.
3. Development of method and equipments for large scale harvesting by sedimentation to reduce energy of centrifuge by a factor of 50 times and oil improvement by 2.6 times.
4. Discussion on possible oil production cost of current PBR system of USD14.3/kg refined oil ($12/L). The biomass yield and oil yield of 1ha/year are 21630kg and 10815kg, respectively, which are 27 times of yield produced from soybean. Analysis to show that further improvement of the PBR system as well as extraction technology will reduce the oil production cost to a suitable level. In terms of meeting the demand of transportation and saving land, algae is a potential vegetal source for biodiesel.

1 BACKGROUND

Fossil fuels are the major source of energy in the world but their combustion resulted in an increase in the atmospheric CO_2 concentration leading to global warming. If the fossil burning were substituted by biomass burning, the global CO_2 emission of 5.4 Gt/year of carbon produced could be brought down to the emission levels of 1985 by the year 2050 (Fjerdingstad, 1971). In addition, it is expected that fossil oil fuels are to be depleted by the years 2050. Moreover, oil price has increased from $30/barrel in 2000 to $80-100/barrel in 2008, thus imported fuels become a big problem at present. With the need to reduce carbon emissions, and the dwindling reserves of crude oil, liquid fuels derived from plant material – biofuels – are an attractive source of energy. Beside, in comparison with other forms of renewable energy such as wind, tidal, and solar, liquid biofuels allow solar energy to be stored, and also to be used directly in existing engines and transport infrastructure.

Current sources of commercial biodiesel are primarily soybean oil, rapeseed oil, palm oil, corn oil, waste cooking oil, and animal fat, but these sources of biodiesel cannot meet even a slight fraction of present demand for fuels (Chisti, 2007). However, a major criticism against large-scale fuel production from agricultural crops is that it will consume a lot of farmland, native habitats and drive up food prices. For example, satisfaction of half diesel demand for transport in 2008 using biodiesel from oilseed rape, it would require 17.5 Mha, more than half the land area of the UK (Scott, 2010). Another example, meeting only half the existing US transport fuel by biodiesel would require unsustainably 54% and 24% of the US cropping land using coconut and oil palm, respectively (Chisti, 2007). This requirement would be only between 1 and 3% of the total U.S. cropping area if using algae. In terms of environment, 1MJ of energy produced during the growing, harvest to burning, biodiesel from soybeans will produce 83 kg CO_2 to the atmostphere, whereas, using algae will absorb 183 kg CO_2 from atmostphere (Thomas, 2009). With an extreme rapid growing rate and high oil content without competion the arable land or biodiversity in biofuel production, microalgae appear to be the only suistainable source of biodiesel that has the potential to completely displace fossil diesel (Chisti, 2007). Moreover, biodiesel from algae is lead-free, almost sulfur-free, biodegradable, and can run any modern diesel engine (Mcintyre, 2009).

Growing of algae may be either by raceway ponds or photobioreactors. Raceways are perceived to be less expensive than photobioreactors, because they cost less to build and operate. Although raceways are low-cost, they have a low biomass productivity compared with photobioreactors because raceways are poorly mixed and cannot sustain an optically dark zone. In addition, raceways use carbon dioxide much less efficiently than photobioreactors due to significant water evaporative losses. Productivity of raceway is affected by contamination with unwanted algae and microorganisms that feed on algae (Chisti, 2007). Photobioreactors, in contrast to ponds, offer the opportunity to optimize the light path, the extent to which the incoming light is diluted, and also the frequency of the light–dark cycle seen by an algal cell as it travels from deep in the culture to the illuminated surface (Scott, 2010). Closed photobioreactors have attracted much interest because they allow a better control of the cultivation conditions than open systems. With closed photobioreactors, higher biomass productivities are obtained and contamination can be easily prevented (Ugwu, 2008; Briassoulis, 2010). Typical configurations tested at either laboratory or pilot scale have included vertical, flat plate reactors, annular reactors, or arrangements of plastic bags operated as batches, and various forms of tubular reactor, either pumped mechanically or by air-lift.

The coil-type systems were made of flexible plastic and coiled around a supporting frame to form a helical coil tubular photobioreactors (Chisti, 2007; Briassoulis, 2010). The scale up of these systems is easy. However, building of these systems seemed to be costly because of a supporting frame. Therefore, the coil-type systems are potentially useful for growing a small volume of microalgal broth, for example, for inoculating the larger tubular photobioreactors (Chisti, 2007). Flat-plate systems are also developed for the production of algae (Evens et al., 2000). Light is evenly emitted from a flat surface screen or from lamps above the culture. Their scale up potential seems to be difficult (Briassoulis, 2010). Biomass output may be limited by photo-inhibition and problems have been reported with temperature control (Moholkar, 2008). Tubular systems are the most widely used commercial systems. Usually they are made of polypropylene acrylic or polyvinylchloride pipes having small internal diameters. Mixing and agitation of the culture is maintained by an air-pump forming bubbles. The scale up of these systems is reasonable. Tubular photobioreactor are very suitable for outdoor mass cultures of algae since they have large illumination surface area (Ugwu et al., 2008). However, controversy surrounds the cost of scale-up, energy consumption, sources of CO2, with estimates of capital and production costs varying widely for different types of photobioreactors (Scott, 2010). Only a few of these designs can be practically used for mass production of algae (Lerh, 2009).

The main disadvantages of photobioreactors, varying however among the individual systems, concern the relatively high space requirements, high energy requirements, with temperature control, cleaning problems and low efficiency in terms of mass production per unit of space. Their operational difficulties may include: growth of algae in tube walls blocking light; high oxygen concentration that can inhibit photosynthesis; limit on the length of the tube in single run (Moholkar, 2008; Briassoulis, 2010). On the other hand, the length of the tube can be kept as short as possible while a tubular photobioreactor is scaled up by increasing the diameter of the tubes. In this case, the cells at the lower part of the tube will not receive enough light for cell growth (due to light shading effect) unless there is a good mixing system (Ugwu et al., 2008). Regarding the effect of weather on the production of algae, tropical developing countries might be potential cultivation sites for commercial production of algal products.

There is one demand from Vietnamese government on development of biofuel for the next 10-15 years. According to the governmental official document dated on May 27th 2008 that 50 000 ton per year of B5 fuel containing 5% biodiesel and 100 000 ton/year of E5 fuel containing 5% ethanol are the target of Vietnamese government by the year 2010. But upto now in 2011, according to Ministry of Industry and Trade, Vietnam has just started to use E5 commercially without any planning of using B5 fuel. It meant that there is not available technology selected for biodiesel production in Vietnam. Technologies for biodiesel production from oil crops, waste cooking oil and animal fat are available but not ready for microalgae. Unfortunately, biodiesel from oil crops, waste cooking oil and animal fat cannot realistically satisfy even a small fraction of the existing demand for transport fuels and the cost of biodiesel from microalgae is still high compared to fossil diesel. According to Chisti (2007), the cost of petrodiesel, palm oil and algae oil biodiesel was 0.5, 0.66 and 1.4 USD/L respectively for the case of 30% oil in algae. Therefore, the cost of algae oil biodiesel would be equal to petroldiesel only if oil content in algae was 57%. This condition leads to the difficulty for the policymakers to select the technology for biodiesel production in Vietnam.

As shown in the above analysis, the biodiesel cost from microalgae should be reduced for commercial application. It can be decreased only by an integration of the engineering with discoveries in algal biology in order to increase the triglycerides content in algae, to reduce specific energy used in each step of processing such as growing, harvesting, extraction, and refining.

There was a Ministry level project on biodiesel production from microalgae run by Chemical Enginnering Department of Nong lam University Ho Chi Minh city Vietnam - lead applicant of this project - in between the years 2008-2010. Oil content in algae has been doubled by some stress treatments, i.e., from 10% to 22% . Oil extracted from *Chlorella Vulgaris* has been studied. This composition was suitable for biodiesel production. Lab scale tubular photobioreactor of 170L/batch has been developed for growing of *Chlorella Vulgaris* using solar energy resulted in cell concentration of 125×10^6/mL of culture with a dry matter of 0.67g/L. Wet extraction method had also been tested to give an improvement of 8% of crude oil fraction in comparison to dry extraction method. Furthermore, wet extraction method reduced energy use for biodiesel processing as no drying was required. Some trials of algae growing in wastewater of ethanol plant of Dong Xanh Company in laboratory showed that available nutrient in wastewater with additional nutrient produced normal algae biomass with oil content of 25% (Truong Vinh, 2010).

Beside, the group from the Finnish Environment Institute (SYKE) has experience with municipal water waste treatment, methodology for optimizing oil content in algae and dewatering. These activities have been carried out on a small, laboratory scale and the proposed project will enable the SYKE team to test and help implement techniques on a larger scale.

Starting from the above results, this project will concern with the development of large scale tubular photobioreactor for growing of microalgae for biodiesel production. Design of this industrial scale tubular photobioreactor will focus on the following engineering targets: easy in construction and operation, simple temperature control, moderate space requirements, enhancement of efficiency of CO_2 use, avoidance of photosynthesis inhibition, and easy in scale-up. The economical targets will be low cost of construction, growing in wastewater to use available nutrient sources, using CO_2 produced from ethanol plant. The method of large scale harvesting will be developed. Other technologies to reduce biodiesel cost will be considered. These include maximising the rate of production of oil, reducing of energy used in downstream processing such as flocculation harvesting, wet extraction.

The project is expected to contribute to the improvement of environment as well as the demand of fuel for transport in Viet nam in the future. The project results will also aim to create more jobs for the poor to work in the fields or in biodiesel processing plants such as handling of algae, running of extract equipments. The research institutions will benefit by continuous improvement of their knowledge and R & D capabilities. Cassava Ethanol plants will get benefit from their knowledge improvement on algae biodiesel processing in both the economical and technological point of view. In terms of economic, the managers will have the actual data for easier making decision in the future about investment of larger systems. In terms of technology, the technical peoples ethanol plant will have a chance to practice the whole actual algae biodiesel processing. The policymakers are expected to get suitable information for their future plan of biofuel program.

2 PROJECT DESIGN

- Briefly discuss the appropriateness of the project strategy and design (internal logic) including the management set-up and institutional framework, and the changes made during implementation.

Project rationale, principles, strategies

Biofuels are renewable resources of energy that could be sustainably supplied to replace the petroleum reserves which are to be depleted by the years 2050. In comparison to fossil oil fuel, biofuel can represent an environmental improvement in terms of less emissions of dangergous gases such as COx and SOx. If microalgae are used to produce biodiesel, there is about 3-8% of solar energy can be converted to biomass whereas observed yields for terrestrial plants are about 0.5%. These interesting properties lead to potential productivities which are far higher than those of palm oil or sunflower. This high productivity combined with both the less competition with feed crop and the possibility to uptake industrial sources of CO_2 has motivated studies depicting microalgae as renewable source for biodiesel.

Vietnamese government has developed biofuel program for the next 20 years. In this program, ethanol plants from cassava have been planned to build along the country. One of these plants- the Dong Xanh Company- was started to produce ethanol in 2009. With the capacity of 400,000 liters of ethanol/day, it produces 60 tons CO_2/day and 4000 m^3 wastewater/day. The experiments at laboratory of Chemical Engineering and Processing Department of Nong Lam University in Ho Chi Minh city, Viet nam, found that the algae *Chlorella Vulgaris* grew well in this wastewater with additional nutrition producing oil content of 25%. The BOD and COD of wastewater were reduced from 86 and 297 to 8 and 78 after growing of algae. Low cost tubular photobioreactor of 170 litter volume has also been developed and worked well.

Therefore, this project will develop an industrial scale photobioreactor for growing of microalgae using high nutrient wastewater of ethanol plant. The system will use CO_2 source produced by this ethanol plant. The facilities for processing of biodiesel including harvesting, oil extracting and refining will also be built. The project will study the improvement of algae growing, oil extract and refining, cost, and environmental issues from microalgae biodiesel.

The SYKE team will assist and share their knowledge on growing algae in wastewater systems and how to improve the lipid content. The SYKE will help us the methods of how to measure bulk lipid concentration using Nile Red staining. This is a simple, low-cost method that the SYKE team has long experience with. We are also planning Finnish scientist going for some period to the pilot plant, providing help setting things up and also assisting in testing different set-up of the system. SYKE will also provide assistance in testing the water before and after the algae growth, i.e. finding out how much nutrients and potentially also other contaminants have been taken up by algae. This information will be further used for analysis of the environmental benefits of the project.

The ULB team has experience on extraction and transesterification will also help the project to train the students in these areas during the project implementation.

This project will start from building of 400 litter volume photobioreactor for experiment of optimum growing condition including stress treatments. The system will be scaled-up to 2.5-8m^3 or higher for industrial scale photobioreactor for study of large scale growing condition. The protocol from lab scale will be applied to this industrial system to optimise oil production. Down stream processing will also be built and tested to evaluate the biodiesel production cost. This pilot algae biodiesel system will be used for consideration of application to larger land in Dong Xanh company and other ethanol plants as well as demonstration to policymakers and any related agencies.

Management set-up

This project will be implemented through the project leader in Nong Lam University. The project leader will coordinate the various collaborators of this project in Vietnam. He will also be responsible to manage the team and appoint the required personnel in the project. The training and research component will also be supervised directly by the project leader in conjunction with other collaborators. The project leader will be in direct touch with the Dong Xanh company and Finnish Environment Institute (SYKE). The project leader and collaborators from the Dong Xanh company and SYKE will supervise the staff members visiting Europe for the training or research. The laboratory facilities of NLU and equipments purchased by Dong Xanh company will be used for some parts of related activities of the project. The photobioreactors will be designed and manufactured by NLU and installed at NLU for small scale units and at Dong Xanh Company for large scale units. Dong Xanh company will be in charge of providing land, housing, CO2, waste water and other facilities for growing experiments. They will also purchase the equipments for handling of biomass, extraction and transesterification under the supervision of NLU.

Institutional framework:

NLU: most of PBR design and experiments, planning for installation of 500m^2 system at GFC, for visiting, training carried out by NLU staff. The available laboratories of NLU were used for related activities such as growing, harvesting, drying of algae and extraction of oil from algae, storage of algae in cold storage facility, or chromatographic column for separation of the components from crude oil. The algae strains and outdoor PBR of 70mm tube size with volulme of 170-250 liters was provided by NLU for algae growing experiments. Training of 14 students of Chemical Engineering Department via their Bachelor thesis and 6 staffs of NLU as well as parts of some Master students on algal biodiesel was involved to EEP-3-V-053 project.

EEP: The fund of EEP-3-V-053 for installation of Fluorescense Spectrophotometer (FS-8000) was usefull for strain selection. This fund was helpfull in building of different algal growing facilities such as seed growing system, outdoor PBRs of 210mm, 170mm, 140mm tube sizes with volume of 400 liters and the outdoor stress streatment systems. EEP-3-V-053 provided fund for expense of experiments at NLU such as chemicals, glassware, support analysis tools for Bachelor training of 14 students of Chemical Engineering Department and some Master students realated to algal biodiesel topics. The trip to Europe of Vietnamese project leaders was organised using EEP fund to visit the SYKE, the partner from Finland, and the AlgaePARC of Wageningen University of The Netherlands. This trip helped the partners to exchange the knowledge each others and discuss more details on the algal biodiesel as well as to strengthen our collaboration.

SYKE: discussion on the growing methodology, helping in the Nile Red method using the FS-8000 equipment, planning for visiting, preparing for the training of Vietnamese young staff at SYKE on algal biodiesel. Dr. Kristian, the scientist of SYKE, has presented a seminar on algal biodiesel for staff and students of NLU and related peoples at NLU during his trip to Vietnam on 28 Feb to 04 Mar, 2012. The exchange of knowledge between SYKE and NLU in algal biodiesel was usefull in this project.

GFC: providing waste water, planing for experiments of PBR 500m^2 at GFC
ULB: discussion on the enzymatic extraction methodology

Changes made during implementation

Marine algae and fresh algae:

Original issues: Growth experiments initially using marine algae. However, when conducting experiments with wastewater, the fresh water, some salts was added to approach the sea water for a suitable condition of algal growth. This adding of salts led to a high salinity of the spent medium which was not satisfied the standard of waste to be discharged to the environment. The marine algae produced mainly the triglycerides to be converted to biodiesel via transesterification process.

Solution: Growth experiments was conducted with fresh Chlorella vulgaris to avoid the environmental problem mentioned above. However, fresh Chlorella vulgaris produced maily the hydrocarbons rather than fatty acids. This hydrocarbon can be burnt directly in the engine. Therefore, the transesterification process is not required any more.

Development of modelled wastewater:

The distance from NLU to ethanol plant is very far. Handling of large amount wastewater from ethanol plant to NLU is complicated and costly. Therefore, the procedures are modified by inserting an intermediate step between growing in fresh water and wastewater in PBR experiments. This intermediate step is to grow the algae in a modelled wastewater in PBR at NLU before going to the ethanol plant.

Change of the place for installing of 500m2 system:

In the proposal, the work at GFC will be the final step to analysis the prodution cost of a pilot system. However, we are not sure that the system can be installed at GFC for experiments. Therefore, additional ethanol plant was considered to test the growing system. The Xuan Loc ethanol plant at Dong Nai province was selected. The wastewater of Xuan Loc ethanol plant was collected and conducted growing test with *Chlorella vulgaris*. Later on, the GFC got problem with their business could not collaborate any more. We also could not collaborate with the Xuan loc company due to limiting of time. Finally, the 500m2 system was installed at NLU for experiments.

So far, for large scale, we have already installed the growing system, harvesting by centrifuge or sedimentation containers, and drying facilities. Comparison to the original plan, we didn't have the extraction and refining equipments for large scale system but we did have these facilities for lab scale.

Financial change:

Due to the problem of GFC, adjustment of budget has been done. The original contribution of Euro 34,200 of GFC is now adjusted to actual contribution of this company of Euro 15,400. Up-dated partners and budget contributions of EEP-3-V-053 is given in Table 1.

Table 1: Up-dated partners and budget contributions of EEP-3-V-053

	Partners	**Euro**	**%**
1	Lead Applicant (NLU)	18,000	14.25
2	Dong Xanh company (GFC)	15,400	12.19
3	Finish Environment Institute (SYKE)	9,000	7.13
4	Universite Libre de Bruxelles (ULB)	1,800	1.43
5	EEP	82,085	65.00
	Total cost	**126,285**	**100%**

End date: the end date of Nov 2012 is changed to Sep 2013 to fit the progress of this project.

3 INPUTS

1. Financial resources:

Error! Reference source not found.a: Financial Report Summary Table (see in the annexes)

Assessment of main constraints and issues faced, and explanations for of major differences between total actual expenditure and budget:

<u>Output 5.1</u>: Installation of pilot biodiesel processing plant

This output, the actual expenditure was only 33% due to the fact that GFC has purchased only Eur8800 for the oil press, centrifuge and dryer. Whereas, in the plan, the contribution of GFC for this part should be Eur27000 for equipments to complete the pilot biodiesel processing plant.

<u>Output 6:</u> the actual expenditure was only 56% of the target because less transportation of the partners from SYKE and ULB.

<u>Output 8.1:</u> the training of staff in Vietnam on related issues was organised for training of 22 Bachelor and 3 Master students of NLU. The expenditure for this part was 29% of the total. We haven't got enough condition to organise the training class for related staff in the provincial areas.

<u>Output 8.2:</u> the workshop and demonstration materials were not yet organised. This part took 11% of the total mainly for materials to support the students.

2. The Involvement / contributions of the different partners in terms of human resources, expertise, physical resources, etc. Assessment of the operation of the partnership

NLU: most of design and experiments of stress, harvesting, photobioreactors, scaling up and planning for installation of 500m^2 system at GFC, for visiting, training carried out by NLU staff. The available laboratories of NLU were used for related activities such as growing, harvesting, drying of algae and extraction of oil from algae, storage of algae in cold storage facility, or chromatographic column for separation of the components from crude oil. The algae strains and outdoor PBR of 70mm tube size with volulme of 170-250 liters was provided by NLU for algae growing experiments. Training of 22 students of Chemical Engineering Department via their Bachelor thesis and 6 staffs of NLU as well as parts of 3 Master students on algal biodiesel involved to EEP-3-V-053 project.

EEP: The fund of EEP-3-V-053 for installation of Fluorescense Spectrophotometer (FS-8000) was usefull for strain selection. This fund was helpfull in building of different algal growing facilities such as seed growing system, outdoor PBRs of 210mm, 170mm, 140mm tube sizes with volume of 400 liters and the outdoor stress streatment systems. EEP-3-V-053 provided fund for expense of experiments at NLU such as chemicals, glassware, support analysis tools for Bachelor training of 22 students of Chemical Engineering Department and some Master students realated to algal biodiesel topics. The trip to Europe of Vietnamese project leaders was organised using EEP fund to visit the SYKE, the partner from Finland, and the AlgaePARC of Wageningen University of The Netherlands. This trip helped the partners to exchange the knowledge each others and discuss more details on the algal biodiesel as well as to strengthen our collaboration.

SYKE: discussion on the methodology, helping in the Nile Red method in using the FS-8000 equipment for quick determination of oil in algal cell, planning for visiting, preparing for the training of Vietnamese young staff at SYKE on algal biodiesel. Dr. Kristian, the scientist of SYKE, has presented a seminar on algal biodiesel for staff and students of NLU and related peoples at NLU during his trip to Vietnam on 28 Feb to 04 Mar, 2012. The exchange of knowledge between SYKE and NLU in algal biodiesel was usefull in this project.

GFC: providing waste water, CO2, installation of some equipments for biodiesel processing, planning for experiments of PBR 500m^2 at GFC

3. Brief overview of the use of equipment

The fluorescent spectrophotometer FS8200S funded by EEP-Mekong has been installed in April 2012 for mainly determination of the crude oil in algal cell. Staring Jan 2013, the treatment of algae by spectral light using fluorescent spectrophotometer FS8000 was conducted to isolate a new strain of *Chlorella vulgaris* with better oil and biomass. The use of FS8000 was efficient in this project.

The centrifuges installed by NLU and GFC were helpful in development of the sedimentation method of harvesting which saved 50 times of electrical energy for centrifuge. This method contributed to reducing production cost of algal biodiesel.

The PBRs manufactured in this project using EEP fund were useful in testing the performance of PBR and studying the growing methodology. Testing of these PBRs helped us to improve the design of PBR working properly for algal growing in Vietnam climate condition without using cooling equipments. This also helped us to design a low cost PBR with low energy consumption with best distribution of CO2 in the medium for algae to grow. We also got experiences with the effect of the transparency of plastic materials on algal growing rate, the durability of this plastic under sunlight.

4. List of contracts (works, supplies, services) above 10.000€ awarded during the reporting period, giving for each contract the amount, the award procedure followed and the name of the contractor

Table 2: List of contracts above 10.000€ awarded during the project

No	Contract	Amount, EUR	Award procedure	Contractor

1	Fluorescent Spectrophotometer FS-8000	22802	• The competive tendering process was applied • The Nguyen Anh company was selected based on the lowest price provided by this company	Nguyen Anh Limited Liability Company (Supplier: JASCO International Co. LTD.)

5. Brief review of the adequacy and timeliness of the inputs:

The finance, equipments and human resources are adequate for this project to implement. In the plan, the GFC would like to invest their own the plastic making machines helped to make the tube for this project. Even though this plan has not been included in the project activities, but via discussion, we found the condition for development of the photobioreactor using plastic material for tube. However, the company got the difficulty in financial issues affected our design of the tube. We had to select the available plastic tube in the market to install in the PBRs. Therefore, the transparency, the durability of the materials could not be adjusted to our design. The PP plastic material currently is not good enough for light penetration reducing the biomass a factor of 1.95 times compared to glass leading to low biomass of our PBR. However, this available plastic tube is the cheapest material for building of solar receiver of a PBR compared to others such as Acrylic Plexiglass. For instance, the price was USD 0.268 and 85.7 per meter of PP plastic (D=170mm) and Acrylic Plexiglass (D=145mm, thickness = 3mm), i.e., the Acrylic Plexiglass is 320 times more expensive than the PP plastic. The PP plastic material used in our PBR was not UV protective, therefore, its shell life under sunlight was short revealed the disadvantages of this material. It's should be considered to ask the collaboration of plastic companies for this issue.

The GFC has carried out some activities such as providing of wastewater, purchased of the dryer, oil press and the centrifuge. Due to the business problem, this company has stopped collaboration so we could not install the algal growing system of 500m2 to conduct experiments at this company. This led to the delay of the project to change the end date from Nov 2012 to Sep 2013.

4 IMPLEMENTATION OF ACTIVITIES

1. Experiments of optimal growth methodology for indoor condition

The optimization of the growth methodology indoor was studied for Chlorella vulgaris. The main purpose was to find condition for enhancement of oil productivity and saving energy. In the first period of study (Sep 2011-July 2012), Basal medium was used for experiments. In general, nutrient stress treatment increased oil content but decreased biomass. However, oil productivity was increased by a factor of 2.2-2.6 times. Using 30% of Basal medium was good for cell density of 8-15 million/mL. Maximum oil productivity of 0.47g/L was obtained when adding 0.675 gMgSO4.7H2O/L during nutrient stress treatment. It was found that growing of algae in glass container increased biomass productivity by 1.95 times in comparison to the plastic container.

In the second period of study (Aug 2012-Dec 2012) wastewater was used for experiments. Physiochemical characteristics of wastewater of GFC and XuanLoc companies were analysed. The nitrogen and potassium were 8-10 times of the Basal medium. The magnesium was 17-18 times higher than the Basal. Therefore, wastewater was diluted to 20-30% for algal growing resulted biomass of 1.5-2g/L under fluorescent light. Similar biomass was also obtained under LED light in which required only half energy consumption. In the third period of study (Jan 2013-June 2013), nutrient stress treatments were conducted under LED light increased oil by factor 2.6 to produce oil productivity of 0.47g/L. The improvement of oil by nutrient stress under LED light is a new result has not been published elsewhere.

2. *Development of small scale PBR for growing of algae outdoor condition. Study the methodology of algal growing in PBR with stress treatment to enhance oil content for biodiesel*:

The tubular photobioreactors (PBR) of different tube sizes (70-210mm) were developed for algal growing test. The main objectives of this part was to design a low cost PBR with an acceptable performance for algal growing outdoor condition. The tube was constructed by plastic with the thickness in between 140-180 μm. Different types of plastic materials such as PP, PE and

PVC were tested for the performance of PBR. The air-lift pump was used for pumping of the liquid. No cooling system was used in this PBR. The 400 liters systems were designed in Sep 2011 to Dec 2011 and manufactured in early 2012 in which the LCP400-D140 is denoted for 400liter volume with tube size of 140mm. The growing tests were performed for LCP400s in the period of April 2012 to Dec 2012. Algae grown well in the medium temperature of 28-43^{o}C. High temperature up to 47^{o}C killed the algae. Different growing methodologies were conducted to determine best tube size. The tube size of 170mm using PP plastic was selected because it had highest oil capacity (5.75g/m2), stable in algal growing and lowest production cost among the tube sizes. The biomass was in the low range of 0.4-0.74 g/L. This result is similar to Eduardo Jacob-Lopes et al. (2009) where the maximum biomass was 0.944g/L in laboratory for the case photoperiod of 14:10 (night:day, h). The main reasons come from less photoperiod and low transparency of plastic material for tubing of the PBR. The period of light in our system was less than 10h/day (7AM to 17PM). The total energy provided by sun in a day is more than the requirement for algal growth, but the duration of the light period is not suitable. In addition, as noted in our experiments, plastic material reduced biomass by a factor of 1.95 times compared to glass material. Therefore, in order to get 2g/L, the photoperiod of 12:12 should be applied with an improvement of the transparency of plastic material for PBR.

Application of the stress treatment for outdoor condition using information of indoor condition was conducted for LCP400s during January to July 2012 (dry season) and August to December 2012 (wet season). Different nutrient deprivation treatments were applied such as using the spent medium, or 30-60% Basal medium for stress treatments of the algae grown 7-24 days in PBR. Oil content was improved dramatically to 69% compared to the control (7-23%). However, most of the cases oil productivity were very low (0.02-0.04 g/L) and were about 1/10 of indoor condition (0.22-0.47 g/L). The stress treatments conducted in the 40L column PBR outdoor got the same situation with growing in LCP400 resulting low biomass compared to indoor discussed above. Therefore, further modification of the PBR used for stress treatment should be considered such as tubing material, photoperiod, and growing methodology.

3. *Strain selection for biodiesel*:

The strain selection was conducted for waste water using both marine and fresh *Chlorella* algae. The marine *Chlorella* was found to produce high content of triglycerides suitable for transesterification to convert them to methyl esters for biodiesel. However, for the growing of algae, adding of some salts to the waste water has been done to approach the sea water. This modification led to high salinity of water after growing affecting the environment. The fresh *Chlorella vulgaris* was then tested with waste water. This strain grown well in wastewater producing high content of hydrocarbons rather than triglycerides. These hydrocarbons were found to be used directly as fuel without transesterifiaction to convert to methyl esters as required for triglycerides.

The fluorescent spectrophotometer FS8200 funded by EEP Mekong was installed in April 2012. It was used mainly for quick determination of the crude oil content in the algal cell. From Jan 2013 to Sep 2013, the treatment of algae by spectral light using fluorescent spectrophotometer FS8200 was conducted to isolate a new strain of *Chlorella vulgaris* with better oil and biomass. This new strain has been tested for small volume of 3 liters indoor resulted initially an improvement of oil content by a factor of 1.4-1.5 times compared to the control. However, this improvement was not significant for outdoor. This situation is similar to what we have met with stress treatment by nitrogen deprivation for outdoor condition. The problem may come from that the growing condition outdoor for current PBR system is not good enough as indoor.

4. *Scaling up to industrial scale PBR: 2500, 6000, 8000 liters and applied to 500m2 system*

The scaling up of the PBR to higher volume was based on the strategy of low cost and low energy consumption..The PBR was scaled-up from 400 liters to 2500 liters of the tube volume or solar receiver, so called LCP2500 in which the volume of degas tank was 500 liters. The LCP2500 had total volume of 3000 liters and total energy used for pumping and degas was 262W. This step is important before scaling up to 500m^2 system.

Scaling up to industrial scale of PBR was carried out for 6000-28000 liter system. The LCP6000 and 8000 worked properly to obtain biomass of 0.5g/L. The specific energy reduced from 354W/m^3 (400L) to 53W/m^3 (6000L). Testing of different growing methodologies in PBR under outdoor conditions to optimise the growing process using LCP6000 and 8000 systems was conducted. From this, a pilot algal growing system of 500m2 land was designed including 2 LCP6000, 2 LCP8000 for growing and 2 LCP400 for nursery. The PBRs for 500m2 land was manufactured during April-June 2013 and installed at the experimental site of ChEPro (NLU) during June-July 2013.

It has been reported previously that our collaborator (GFC) got the difficulty in their business and could not effort to complete the downstream system as planned in the proposal. However, GFC has done something such as preparation of land, purchase the dryer, centrifuge, and oil press. We have already asked other ethanol plant - the Xuan Loc plant- in Dong Nai province to overcome this situation. We have come to Xuan Loc company to collect the wastewater for analysis and for algal growing experiments. The results were given in the previous semi-annual reports. However, the process for replacing the GFC by Xuan Loc company was not simple. Therefore, we decided to install the 500m^2 pilot PBR at the experimental site of ChEPro (NLU) for growing experiments using wastewater model (not real wastewater) developed in this project. So far, for large scale, we have already installed the growing system, harvesting by centrifuge or sedimentation containers, and drying facilities. Comparison to the original plan, we didn't have the extraction and refining equipments for large scale system but we did have these facilities for lab scale. In addition, through out of the project, we have considered either wet extraction or dry extraction for reduction of the energy. The original plan designed the extraction equipment based on the dry extraction method, however, the wet extraction appeared to be more efficient due to less energy. Even though the selection of wet or dry extraction method is still under consideration, without the dry extraction equipment would be purchased by GFC, the progress on extraction activities was running well.

5. *Development of harvesting method for low energy consumption and high oil content*:

Both sedimentation and flocculation by pH change were applied. The method of Zechen Wu et al. (2012) and Vandamme et al (2012) was modified. The efficiency and oil content were checked with respect to sedimentation time and pH3 and pH11. A 2 days sedimentation under pH11 was the best with oil content improvement by a factor of 2.6. Biomass recovery was 85% for liquid depth of 10cm. Biomass recovery can be improved to higher than 90% by liquid depth of 8cm. This long sedimentation time was considered as a stress treatment on algae. The flocculation by adjustment of pH to 11 and adding of Mg^{2+} resulted in very fast sedimentation of algae in 10 min, but no oil improvement. Therefore, sedimentation for 2 days under pH11 was selected. The procedure of harvesting by sedimentation under pH11 for 2 days reducing 50 times of energy for centrifuge and improvement of oil content by a factor 2.6 is a new method developed in this project.

6. *Study the downstream processing methodologies*:

Chemical composition of wastewater, algal oil of marine and fresh algae have been analysed. The oil of marine algae was found to contain mainly triglycerides suitable for biodiesel after transesterification. In contract, the fresh algae synthesized mainly hydrocarbons (77% of $C_{22}H_{46}$) that can be burnt directly in the engine without transesterification step.

Wet extraction method is useful for extraction of oil from algae in comparison to dry method in terms of saving of energy for drying. The wet extraction using Chloroform-methanol-water followed Folch (1957) and Bligh and Dyer (1959) procedure was tested. The Folch procedure was found to be better than Bligh and Dyer. The extraction efficiency was about 72% of Soxhlet by hexane solvent. Extraction and refining are the two important steps contributing high percentage in the production cost of algal biodiesel as shown in the cost analysis section. Reducing of the cost in this step will reduce the biodiesel cost dramatically.

7. Testing of B5 in engine

The B5 was prepared by mixing of 5% refined oil of fresh algae and 95% of petrol diesel. The Diesel Engine RK125-2X-GE and generator KUBOTA-ASK– R150 was used. The smoke concentration of the flue gas was measured by Diesel OP-120 smoking meter and related software. The fuel consumption of petrol diesel was lower than that of B5, especially for both methyl and ethyl esters at high loading. The smoke concentration of biodiesel was lower than that of petrol diesel indicated the contribution of biodiesel to reducing the contamination of air due to petrol diesel. The smoke concentration of B5 from algal biodiesel was similar to B5-B20 of biodiesel of soybean.

8. Environment impact of this project

Testing of the medium composition after growing of algae showed that the BOD and COD were 9.6 and 33 mgO_2/L, respectively, which is below the critical values that can be discharged to the environment. Most of the nutrients in the medium after growing were in a low range compared to the original Basal medium. The values of spent medium after 7 days of growing satisfied the level A of Vietnamese standard of industrial wastewater for aquaculture processing (QCVN 11: 2008/BTNMT) that can be discharged to the water resources used for domestic water.

The spent media after harvesting was treated by HCl to convert NaOH into NaCl that is safely to discharge to the environment.

The smoke concentration of the flue gas when using B5 was lower than that of petrol diesel as reported in the above section implied the impact of this project on the reduction of air contamination.

9. Cost analysis

Cost analysis was carried out for the current PBR system and assumed systems based on the experimental data in this project. The calculation was based on the actual results from the PBR 6000 liter (LCP6000).

For the system using Basal medium with CO2 rate of 0.8L CO2/L medium continuous run in 24 days with biomass of 0.5g/L, the biodiesel cost is USD129/kg oil. ($110/L oil). By using waste for nutrient, this cost reduces to $112.4/kg oil ($95.5/L). Further application of wet extraction, the cost reduces to $76.7/kg oil ($65.2/L). If the rate of CO2 is 1.4 L/L, the growing time is reduced to 14 days with a biomass of 0.88g/L. The cost is now reduced to $31/kg oil ($24.4/L).

Assuming that the stress treatment under LED light can be applied as indoor case to produce oil content of 50%. In this situation, supplementation of electricity from agricultural waste should be applied to provide artificial light source as well as some parts of power to the PBR. The biomass is assumed to be 1g/L and the cost is now become $14.3/kg oil ($12/L). For a better material of tube for PBR, the biomass can be higher, say, 2-3 g/L, and the cost may be reduced further.

The analysis was useful to see that harvesting cost is reduced to very low of 0.2-0.6% of the total by mean of sedimentation method. When growing methodologies were applied, the growing cost occupied only 49.3% whereas the extraction and refining part increased dramatically to 50.7% of total. This is to remind that improvement of the extraction and refining steps will reduce dramatically the cost of biodiesel.

10. Proposal of algal biodiesel production system based on the integration of different techniques and potential of algae for biodiesel:

<u>Algal biodiesel production process:</u>

From the results obtained in this project, figure 1 is the proposal of algal biodiesel production process.

Algae => Spectral light treatment => Growing in ethanol wastewater for 8-10 days (normal light) => Nutrient deprivation under LED light for 5 days => Sedimentation harvesting for 2 days

Figure 1: Proposal for algal biodiesel production process

The above steps have done separately indoor and outdoor condition through out the experiments of this project. However, a combination of these steps has not been carried out. The oil content is expected to be 50%.

Potential of algae for biodiesel was analysed by comparison on the biomass productivity of algae grown in different conditions and systems. A possible oil production cost of our current PBR system is USD14.3/kg refined oil ($12/L). Further improvement of the PBR system as well as extraction technology will reduce the oil production cost to a suitable level. Even if the production cost of algae system is still high, the most advantage of algal diesel system is its yield. The biomass yield and oil yield of 1ha/year are 21630kg and 10815kg, respectively, which are 27 times of yield produced from soybean. In terms of meeting the demand of transportation and saving land, algae is a potential vegetal source for biodiesel.

11. Capacity building

Training of 22 Bachelors, 3 Master students and 6 staff of NLU on related area.

5 ACHIEVEMENT OF PROJECT OUTPUTS

- List the main outputs produced and compare to those planned and as specified by the various output indicators and targets. Explain the main constraints and issues and any major differences between the planed and actual achievements.

Table 3: List the main outputs produced and compare to those planned of the project

PLANNED OUTPUTS	PRODUCED OUTPUTS	Indicators
1. Development of low cost tubular photobioreactor 250L	• Development of low cost tubular photobioreactor 400L with tube size 170mm	• The arrangement with maximum oil productivity is identified • Stable in algal growing and lowest production cost
Notes: *the volume of PBR was changed from 250L to 400L.*		
2. Optimisation of growing process of algae in 250L photobioreactor using waste water	• Creation of new strain using spectral light treatment • Optimisation of growing process of algae indoor condition using waste water • Suitable growth methodology of algae in 400L PBR • Methodology of stress treatment outdoor	• Selection of best strain for waste water: improved oil content a factor of 1.45 times • Identification of condition for maximum biomass and oil content: oil content improved by 2.6 times by nutrient stress treatment at 30% wastewater under LED light • Harvesting of 10-20% of volume is easy for handling of the cultured broth in stress treatment, sedimentation and centrifuge and reducing cost of investment for harvesting • Oil content increased to 69%, however oil productivity decreased due to less photoperiod and low tube transparency
Notes: *a wastewater model was developed similar to wastewater of ethanol plant. The optimisation was conducted for wastewater indoor condition, and for wastewater model and Basal medium outdoor condition*		
3. Improvement of downstream processing to reduce biodiesel cost	• Development of harvesting method by sedimentation under pH11 to reduce energy of centrifuge and improve oil content • Wet extraction using Folch procedure	• Identification of processes with less energy use and high recovery : reducing of centrifuge energy 50 times and increasing oil content by 2.6 times • Oil recovery was of 72%. Less energy compared to the dry extraction.
Notes: *In wet extraction, oil recovery should be improved in the future, enzymatic method should be considered.*		
4. Development of industrial low cost photobioreactor with surface area of 500m^2 for growing of microalgae	• Scaling up from 400L to 6000L and 8000L PBR • Development of 500m2 system including 2 LCP6000, 2 LCP8000 for growing and 2 LCP400 for nursery.	• The pilot scale (80-160m^2) is tested: algae grown well in these PBRs. The specific energy reduced from 354W/m^3 (400L) to 53W/m^3 (6000L) with the same biomass. the total electricity for growing of 1 ha is estimated to be 37kW/ha • Scaled up to 500m^2 system worked properly based on the performance of 6000L PBR.
Notes: *In the plan, the 500m2 system should be installed at GFC for testing using wastewater of this plant. However, in the middle of the project operation, this company could not effort to support any more, therefore, the system was installed at NLU for testing using wastewater model.*		
5. Building of pilot biodiesel processing	• Installing of some related equipments such as centrifuge, dryer, oil press	• Some equipments for biodiesel processing and housing are ready
Notes: *In the plan, the pilot biodiesel processing plant should be installed at GFC for testing. However, in the middle of the project operation, this company could not effort to support any more, therefore, only some of the related equipments were installed. We used these equipments to conduct experiments at NLU*		
6. Biodiesel production from 500m^2 land system and its	• Growth performance of algae in 6000L PBR	• Identification of best condition for producing of high biomass and oil

trials in engine	• Chemical composition of oil from marine algae and hydrocarbons from fresh algae. • The B5 from algal biodiesel • Testing of B5 in the engine showed that flue gas of B5 was cleaner than that of petrol oil.	• Properties of biodiesel and its use in engine are acceptable
Notes: *In the plan, the marine algae was selected, during experiments with waste water, we found that the marine algae required an amount of salt for stable growth. This modification led to high salinity of water after growing affecting the environment. The fresh Chlorella vulgaris was then tested with waste water*.		
7. Environment impact of this project	• Chemical composition of the spent media • Method of treatment of the spent media using pH increase for harvesting • The test results of flue gas using B5 for engine	• The BOD and COD were 9.6 and 33 mgO_2/L, respectively, which is below the critical values that can be discharged to the environment. • Using HCl to convert NaOH into NaCl that is safely to discharge to the environment. • The smoke concentration of flue gas burnt by B5 of algae-biodiesel was less than that compared to fossil diesel
8. Capacity building (visiting of project leaders, training of 3 staff), dissemination (training of 40 participants), workshop (60 participants) and demonstration	• Visiting of project leaders • Training of 22 Bachelors, 3 Master students and 6 staff of NLU on related area.	• Staff knowledge in algae biodiesel processing improved • Producing of 22 Bachelors in the Chemical Engineering in two years. Contribution to some parts of the thesis of 3 Master students and 6 staff of NLU
Notes: *In the plan, the young staff will be sent to overseas for training. The workshop will be organised in the related areas. However, during the project running, we got the difficulty in management due the change of the Board of Rector, the business issues of GFC, so we could not arrange for these. However, the NLU got benefit from training of 22 Bachelor and 3 Master students via this project.*		
9. Report writing	• Report produced	• Report produced

- Brief assessment of the overall efficiency of the project in its use of resources to produce the project outputs.

Photobioreactor development: In terms of low cost, the PBR has been developed successfully using plastic for tube construction. The system worked properly without cooling equipments reducing running cost described by low specific energy of 53 W/m^3. From this, the total electricity for growing of 1 ha is estimated to be 37kW/ha. The efficiency of this part is considered to be 100%.

Technique for biodiesel production from microalgae growing in photobioreactor using wastewater and CO_2 of ethanol plant: The strain for wastewater was obtained. The algal growth in wastewater has been optimised for indoor condition. Suitable growth methodology of algae in 400L PBR has also developed. The enhancement of oil productivity has not been developed successfully due to less photoperiod and low tube transparency. Photoperiod can be increased by installation of artificial light system using electricity produced from agricultural waste for energy saving. The tube transparency can be improved from plastic technology. These resources didn't include in the project. Therefore, this part is considered to be 90%.

Building of pilot biodiesel processing: only some related equipments have been installed. This part was not successful due to management issues and out of our expectation. This is our of the risk that we have evaluated in the proposal. This part is is considered to be 70%.

In overall, from the available resources, overall efficiency of the project in its use of resources to produce the project outputs is estimated to be 87%.

6 ACHIEVEMENT OF PROJECT PURPOSE, OVERALL OBJECTIVES AND IMPACT

- Asses the achievement of the project purpose (should be achieved by the end of the project) making reference to the appropriate indicators and targets.
- Assess the likely achievement or contribution of the project to its overall objective insofar as this can be determined at the end of the project making reference to the appropriate indicators and targets (the overall objective may not be fully achieved until some time after the completion of the project).
- Assess the likely achievement of the impact of the project insofar as this can be determined by the end of the project. This may require a separate study to be carried out by the project towards the end of the project, in order to determine the major change or difference caused by the project.

6.1 Asses the achievement of the project purpose (should be achieved by the end of the project) making reference to the appropriate indicators and targets

The main purpose of this project is to build a pilot biodiesel producing system from algae occupying about 700 m^2 land for growing and processing in which the growing system occupies 500m2 to grow algae using wastewater and CO2 of ethanol plant. In fact, the 500m2 land for algal growing has been built together with developed growing conditions. The effect of wastewater and CO2 on the cost was identified. However, the biodiesel processing plant in equivalent to 500m^2 land growing system is not ready due to the problem of partners collaboration mentioned previously.

6.2 Assess the likely achievement or contribution of the project to its overall objective insofar as this can be determined at the end of the project making reference to the appropriate indicators and targets (the overall objective may not be fully achieved until some time after the completion of the project).

For summary, the following results have been achieved from this project:

1. Development of low cost photobioreactor simple in construction, easy to operate with low energy consumption. The 6000 liter PBR worked properly produced algae without contamination. The systems can be scaled up easily to 500m2 system and is the base for higher area.
2. Finding three new technologies for oil enhancement in algae, i.e., spectral light treatment the algae before growing, nutrient stress under LED light, and sedimentation stress at pH11 during harvesting, with oil improved factors of 1.45, 2.87 and 2.6, respectively.
3. Development of method and equipments for large scale harvesting by sedimentation to reduce energy of centrifuge by a factor of 50 times and oil improvement by 2.6 times.
4. Discussion on possible oil production cost of current PBR system of USD14.3/kg refined oil ($12/L). The biomass yield and oil yield of 1ha/year are 21630kg and 10815kg, respectively, which are 27 times of yield produced from soybean. Analysis to show that further improvement of the PBR system as well as extraction technology will reduce the oil production cost to a suitable level. Performance of the engine using this biodiesel is similar to other vegetal oils. In terms of meeting the demand of transportation and saving land, algae is a potential vegetal source for biodiesel.

Therefore, it can be said that, this project has contributed to the achievement of the overall objective that to develop a technique for biodiesel production from microalgae growing in photobioreactor using wastewater and CO_2 of ethanol plant and assess the cost of biodiesel and potential of using microalgae as an alternative source of vegetal oil for biodiesel.

6.3 Assess the likely achievement of the impact of the project insofar as this can be determined by the end of the project. This may require a separate study to be carried out by the project towards the end of the project, in order to determine the major change or difference caused by the project

It is expected that if this project applied in the field, the waste discharged from the operation of the plan will be safely to the environment. The BOD and COD were 9.6 and 33 mgO_2/L, respectively, which is below the critical values. The spent media after harvesting by sedimentation will be treated by using HCl to convert NaOH into NaCl that is safely to discharge to the environment.

The smoke concentration of flue gas burnt by B5 of algae-biodiesel was less than that compared to fossil diesel. If algal biodiesel used in replacement of petrol diesel, the contamination of air will be decreased mean that it will contribute to the improvement of environment as well as the demand of fuel for transport in Viet nam in the future. The project results will also aim to create more jobs for the poor to work in the fields or in biodiesel processing plants such as handling of algae, running of extract equipments.

However, this project got a problem with collaboration to the partners for application to the field. Therefore, it is still early to say about the impact of this project without the chance of application in the field.

7 RELEVANCE, SUSTAINABILITY AND SPREAD

- Relevance: how far are the outputs of the project consistent with the interests of government policy, the sector, local communities, etc. Has this changed during the life of the project?

Biodiesel is still no answer to government from the policy makers. However, the local communities are very interesting in this issue, especially this project. But we have not received any consideration from the government up to now.

- Sustainability: What is the likelihood that the things developed by the project which should continue will actually continue as required.

With an extreme rapid growing rate and high oil content without competion the arable land or biodiversity in biofuel production, microalgae appear to be the only suistainable source of biodiesel that has the potential to completely displace fossil diesel. However, solution for the improvement of the technologies to produce algal biodiesel economically required step by step and could not go quickly. This type of project should be under a long term program of the government. This project has developed some useful techniques contributing to the lowering of the biodiesel cost from algae. The technical issues to be solved were also found from this project for a future better solution. However, it is not a profitable area at this stay for the company to invest. The government should recognize this problem to have a policy in helping the company to involve in this type of product. The cost we did analyzed haven't taken into account the co-products which required more time to study. The co-products will contribute to the reduction of biodiesel cost.

In our opinion, this type of project should be continued in accordance with the policy and plan of the government.

- Spread: What are the possibilities for the things produced by the project to be replicated through similar activities or spread by themselves (i.e. through private initiatives rather than further external investment).

It was expected to be applied this project at GFC for further replicated to other similar ethanol plants in Vietnam. However, we can not say anything now because this project hasn't run in the field as planned.

8 IDENTIFICATION OF ALTERNATIVES

- Is there, or would have been, a more efficient, effective or relevant way to approach the problem addressed by the project?

Biodiesel can be produced from other sources that people through out the world are finding. Other methods of growing algae such as using pond can also applied to convert algae to biofuel.

9 FOLLOW ON ACTIVITIES OR INVESTMENT

- What follow on activities or further investment are needed to advance the developments supported by the project. How far has funding support been obtained for these?

Algal biodiesel is still not profitable at this stay. However, compare to other sources of agricultural products such as soybean, rapeseed, palm oil tree, etc, in terms of land saving for food security, sustainability, algae is one of the potential candidate for biofuel. This project have developed low cost photobioreactor and found some techniques that can be applied to improve the production cost. An integration of technologies should be combined such as improvement of plastic materials, supplementation of artificial light and power from agricultural waste via electricity, stress treatment, and co-products such as converting of the residual to ethanol by fermentation or to fertilizer, etc, to improve the production cost.

The further investment can be focused on the following issues:

1. Plastic material is cheap contributed to low cost of the PBR. Improvement of plastic material for better transparency, strength, and UV protection is recommended. This required a collaboration from plastic companies.
2. Sun energy stored in agricultural waste should be converted to electricity to supplement light source to PBR during cloudy, raining, and night time. Building a system such gasification from rice husk or boiler using saw dust to run generator for electricity.
3. Converting by-products into ethanol or fertilizer.

Initially we got support from GFC for funding and our own university for facilities. But this GFC could not effort anymore due to their own business problem. We haven't got any other support up to now.

10 LESSONS LEARNED

- "Lessons learned" are important generalizations based on project experiences (may be positive or negative) which are useful in broader situations for other projects, development of the sector, policy development, etc.
- Briefly present the main lessons learned as distilled from the broad range of project experiences.

From the project, we got the lessons learned as following:

1. *Project management*: we are the researchers who working in the middle between the authorised offices such as EEP-Mekong and NLU's administration offices. During working process, we need to transfer the things such as the money and documents, we need to ask the admin peoples. The researchers work in this project because of their interest. However, the system need the admin peoples so they are involved with this project but not of their interest or responsibility. Therefore, they will work in the free time leading to slow the process. We propose that should have some source of money in the form of management fee supporting the University (not to anyone) so the admin officers will work as their responsibility for the project. This will shorten the process time and helping the project to handle the budget for operation specially when we need to borrow the money in advance from the University.
2. *Commercialization of biofuel*: Biofuel is still the new products in our society and not familiar with the peoples. The policy and support from government are important for the company to invest their money to this area, especially biodiesel. From our point of view, we need some more times to improve the technology before commercialization of biodiesel. Energy is the responsibility of the government. More funding should be considered from the government at this stage.

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ANNEXES

- Supporting information as required.