Jap. J. Phycol. (Sôrui) 30: 207-212. September 20, 1982

Biofuel-gas production from marine algae

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SIVALINGAM, P. M. 1982. Biofuel-gas production from marine algal. Jap. J. Phycol. 30: 207-212.

The viability in production of biofuel-gas from the marine alga, *Ulva reticulata* FORSSKÅL, is presented. It is found that the rate of biogenic methane gas production by marine strain methanogenic bacteria at 50% wet algal thalli amendment is greater by 33.4% on comparison with results of freshwater cattle manure strain methanogenic bacteria under similar experimental conditions. The proportion of methane gas content in this biofuel-gas is ca. 58%, while the remaining gases are CO_2 (major portion), H_2S, NH_3, N_2 and O_2.

In this study, a simple family unit, in the form of a semi-continuous integrated algal digester and gas collector system, is proposed for possible usage in isolated fishing communities.

Key Index Words: *Biofuel-gas*; Ulva reticulata; *marine algae*.

The utilization of marine algae as a substrate for biofuel-gas production is not well practised probably due to the availability of other important economic avenues. However, in countries where algae are not utilized to the fullest extent as in Japan, it is better that this aspect be exploited instead of letting this potential energy resource rot away. This is true in India and there is already a report by RAO *et al.* (1978) supporting such possibilities. Lately, an identical appoarch has been reported by RANTENBACH (1981).

In Malaysia, this aspect of utilizing marine algae is not normally practised. Hence, one can frequently notice large amounts of marine algae going to waste on the shores after rough weather (Fig. 1). As an initial step, the author has studied the possible utilization of the Malaysian sea lettuce, *Ulva reticulata* FORSSKÅL, (SIVALINGAM 1978a, b, 1980). In this connection, the contamination levels of environmental pollutants, such as trace metals (SIVALINGAM 1978a, b, 1978a, b, SIVALINGAM and ZAKARIAH 1979) PCBs and persistent pesticides (SIVALINGAM 1981), have also been clarified.

Here, as another perspective and in view of the energy crisis, the author has further attempted to convert the presently wasted

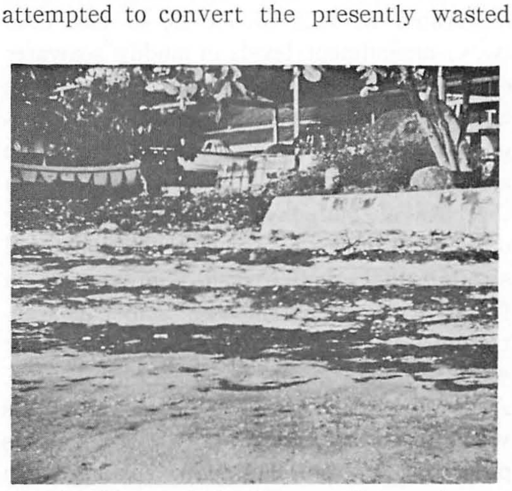

Fig. 1. The washed ashore sea lettuce during rough weather conditions.

Malaysian sea lettuce natural resource into biofuel-gas via biogenic processes. Also from the stance of technological feasibility, a simple semi-continuous household unit algal digester and gas collector for rural fishing communities has been designed. The results of this investigation are presented in this report.

Materials and Methods

Thalli of the Malaysian sea lettuce, *Ulva reticulata* FORSSKÅL, were harvested from the mud-flats of the Marine Depot, Penang, Malaysia, prior to experimentation. The harvested thalli were immediately brought to the laboratory and cleaned off epiphytes and other external contaminants by washing thoroughly either with seawater (a) or freshwater (b) depending on the mode of biofuel-gas (=methane) production; a) biofuel-gas production relying on marine methanogenic strain bacteria and b) biofuel-gas production relying on cattle manure methanogenic strain bacteria. The washed thalli were rid-off surface moisture by pressurizing them against Whatman No. 1 filter paper for five times until negligible weight fluctuation was observed.

For experimental biofuel-gas production from marine methanogenic bacterial strains the algal thalli washed with seawater were macerated at 1, 5, 15, 25, 30, 40 and 50% (w/v) amendment levels in muddy seawater obtained from the algal bed site because it is a well established fact that such methnogenic bacterial forms prevail in these areas (MECHALAS 1974, WHELAN 1974). In the case of biofuel-gas production relying on cattle manure methanogenic bacterial strain, algal thalli washed previously in freshwater were amended at similar concentration levels in freshwater media together with a teaspoon of fresh cattle manure. All amended seawater and freshwater series were then placed in 250 cm^3 vacuum flasks painted black on the outer surface with their mouths sealed airtight, after addition of samples to be digested, with a rubber stopper. The auxillary arm of the flasks were joined individually with polyvinyl tubing which ended directly at the mouth of inverted graduated measuring cylinders filled completely with water and sitting over a trough of water. Under such anaerobic conditions the fermentation process of producing biofuel-gas (=methane) was followed daily over a period of 40 days at a room temperature of 28°C. In order to obtain precise results, three replicates of each experimental lot were carried out. As blanks for both series, one fermentation flask containing 100 cm^3 muddy seawater was used in the marine methanogenic bacterial strain lot while the other containing 100 cm^3 freshwater and a teaspoon of cattle manure was used for the freshwater methanogenic bacterial series. The results were averaged and plotted as methane gas production with time for each experimental lot. Further, suitable methanogenic bacterial activity conditions were maintained by constant daily monitoring of the pH of the digest and adjusting it to lie between 7.0–7.5, where activity is highest.

The evaluation of methane composition of the evolved gas was performed by routing a known amount of the gas through a 15 cm activated charcoal U-tube column trap at −70° C, achieved by mixing acetone with dry ice, which specifically absorbs CH_4 (SWINNERTON and LINNENBAUM 1967), and then through a liquid nitrogen trap at −190° C to condense CO_2. The U-tube trap of activated charcoal was then heated to 90° C with a hot-water bath to release the absorbed methane gas which was calibrated in inverted measuring cylinders filled with water, as mentioned previously. The purity of this methane gas was verified by gas chromatography against authentic methane samples at an isothermal temperature of 35° C on 6 ft×1/8 inch stainless steel column packed with Chromosorb 102 attached to a thermal conductivity detector. In the case of the condensed CO_2 gas, it was determined manometrically in a Warburg-respiratory apparatus with special attachments after bringing the nitrogen trap to normal room temperature.

With the above data and on viewing the prospective usage of biofuel-gas production as energy source from marine algae in rural fishing communities, a simple semi-continuous integrated household unit digester and gas collector system model was designed.

Results

Figs. 2 and 3 demonstrate the rates of biofuel-gas production by both marine and freshwater methanogenic bacterial strains, respectively, over a period of 40 days. The patterns of maximal biofuel-gas production by both strains of bacteria are quite identical except that the maximal production of gas by the freshwater strain was delayed by ca. 4-5 days. It is also noticable that both strains had a small peak at ca. 3 and 6 days, respectively, after initiation of fermentation, which could be attributed to volatile gases (mainly acetic acid) produced by the symbiotic bacteria involved in the degradation of organic matter during the nonmethanogenic phase (liquefaction). Obviously, the shorter period required for decomposition of the organic matter by marine methanogenic bacteria, facilitates rapid methane production processes indicating the high activity of these bacterial forms in relation to the suitability of both substrates and optimal conditions prevalent in the medium. Further, the ratio of maximal biofuel-gas production at 50% *Ulva reticulata* amendment indicates the marine methanogenic bacterial strain's capacity to be greater by 33.4% than the other strains under the present experimental conditions. For both strains, it is obvious that 50% amendment of substrate generally provides a longer period in high production of the gas as compared to the other percentages. Higher amendment levels were found to cause too much frothing and inconvenience in substrate feeding and experimentation, which culminates in inefficiency of the process. Hence, it is concluded that the optimal concentration of algal thalli amendment seems to be in the region of 50% (w/v).

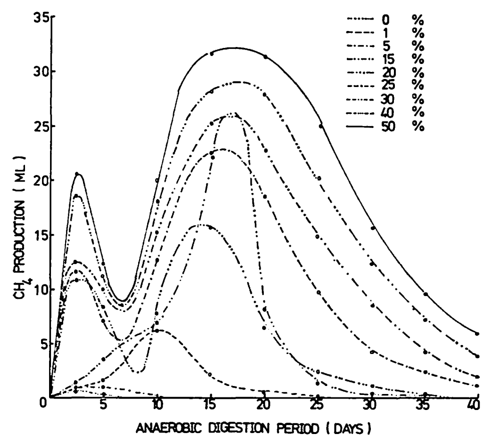

Fig. 2. Rates of methane gas production by marine strain methanogenic bacteria at various amendment levels of *Ulva reticulata* as substrate material.

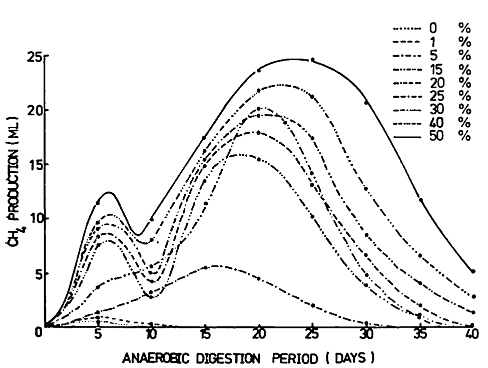

Fig. 3. Rates of methane gas production by cow dung strain methanogenic bacteria at various amendment levels of *Ulva reticulata* as substrate material.

pH calibrations of the marine methanogenic bacterial strain system indicated that for the first ten days there was a drop in pH from 8.2 to 6.5 for most of the percentages except for the control which remained at 8.2. Thereafter until the 40th day the pH fluctuated only between 7.3–7.8, requiring no further buffering with alkaline solution to maintain optimal pH conditions for biofuel-gas production. This self-buffering mechanism of regulating the pH in the marine methanogenic bacterial strain system can be attributed primarily to the inherent properties of seawater arising from the silicate and calcium-carbonate buffering systems and the Iron Paradox. In the case of the freshwater cattle manure methanogenic bacterial strains system, the pH kept dropping from ca. 8.0 to between 5.6-6.3, most of the time, and all these systems required daily adjustments in pH to 7.5-7.8 with 0.1N NaOH solution.

Gas chromatographic analysis of the evolved gas indicated the methane gas content to be 58%, while CO_2 was ca. 35% and the remaining fractions were assumed to be H_2S, NH_3, N_2 and O_2, as illustrated by MECHALAS (1974) and WHELAN (1974).

Fig. 4 is a practical simple semi-continuous integrated household unit biofuel-gas digester and gas collector model derived from a working system examined in the laboratory. The 50% (w/v) marine algal substrate is fed down the inlet pipe to the bottom of the first compartment of the double compartment digester. The level of the liquid substrate in the digester is controlled by the overflow level of the outlet pipe. In the process of digestion, the liquid travels through the compartments and out of the outlet pipe. Here, when assuming a digester 60 cm in diameter and 60 cm in height (=fluid level), the system would theoretically

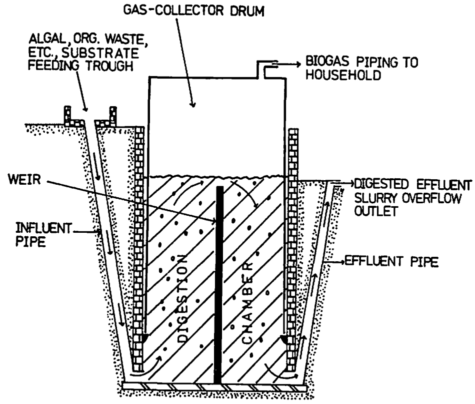

Fig. 4. A practical simple semi-continuous integrated household unit biofuel-gas digester and gas-collector for algal and other household organic waste substrates.

produce 60 l of gas daily from 88.7 kg wet algal thalli. Since the normal optimal detention time of the substrate in the digester is ca. 40 days, one canhave a working semi-continuous integrated system by feeding ca. 23 kg wet algal thalli every ten days at 50% (w/v). Att his feeding rate the detention time of each batch would be 40 days, i. e. before final dischage in the outlet pipe, which is the time required by the substrate to travel from one compartment to the other over the weir and out.

The produced biofuel-gas is trapped by a floating drum, which acts as a gas storage chamber. This accumulated biofuel-gas can then be drawn off as needed for purposes of lighting and cooking.

Discussion

It is obvious that the feasibility of biofuel-gas production from marine algae is quite promising and could be utilized efficiently in countries where this natural marine resource majorly goes into waste. The proposed digester could be an economical asset to rural fishing communities during this period of the energy crisis. Further, besides employing marine algae as a substrate source, the auxillary feeding of organic household vegetable waste, poultry and human excreta could also be incorporated into this system. In such instances, due to the higher content of nitrogen in excreta, the yield of biofuel-gas could be increased (MCGARRY 1980).

Comparison of the methane constitution of the biofuel-gas produced from marine algae (58%) to that of human and poultry excreta (60-65%) indicates not a very drastic difference. In countries where the concept of excreta digesters are not readily accepted, this proposal for the utilization of wasted marine algae could possibly play the function of a forerunner for its acceptance.

The question of having either a marine or freshwater methanogenic bacterial strain digester system depends on the requirements in the further utilization of the effluent slurry as nutrient recycling. Normally, owing to the convertion of the digested slurry into nutrients in available forms for plant life, it is employed as soil conditioners, or as enrichments in fishculture ponds. However, the marine methanogenic bacterial strains system slurry is not viable for the former use because of the high content of salts, which are detrimental to plant life. Nevertheless, the usage of marine algal substrates in digester systems should be encouraged, due to the fact of the simplicity in operational technology and the higher biofuel-gas yield. The situation should be otherwise when poultry and human excreta are introduced into the digester as auxillary feedings, which seems worthwhile to investigate in the future.

As a concluding remark, it can be suggested that until the acceptance of marine algae in the dietary habits of the Malaysian community or its exploitation from the standpoint of pharmaceutical drugs is permissable, algae of both economical and noneconomical significance should be tapped by the rural fishing communities as a substrate in the production of biofuel-gas for cooking and lighting purposes.

Acknowledgements

The author wishes to express his gratitude to the School of Biological Sciences, University of Sciences Malaysia, Penang, Malaysia, for all the aid rendered during the course of this investigation.

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P.M. シバリンガム: 海藻基質からの生起原ガスの生産

海藻類, *Ulva reticulata* FORSSKÅL を基質とした生燃料ガス生産の生産能力についてここに説明されている。上述の湿った海藻の 50% 添加した場合の海産株メタン細菌による生起原メタンガスの生産率を, 同条件下で淡水きゅう肥メタン細菌の生産率と比較してみたところ, 海産株メタンの細菌による生産率は 33.4% も高いことがわかった。この生起原ガスのメタンガス（量）の割り合いを調べたところだいたい 58% で, 残りのガスはおもに炭酸ガス, H_2S, NH_3, N_2 と O_2 からなっていた。したがって, この研究では簡単な単位家族的な完全半連続藻類発酵器とガス収集器からなる系統が離れたところにある漁師農村で利用されることを提案している。

新 刊 紹 介

Lee, R.E.: **Phycology.** xii+478 PP. Cambridge University Press. London, New York, New Rochelle, Melbourne, Sydney, Hardcover 邦価 16,500円. Paper back 邦価 5,900円.

従来, 藻類の教科書といえば, G.M. SMITH (1955) の Cryptogamic Botany とか Manual of Phycology とか数えるほどしかなかった。ところが, 最近あいついで新しい藻類学の教科書が出版された。それらは TRAINER, F.R. (1978) の "Introductory phycology" (John wiley & Sons, New York), BOLD, H.C. and M.J. WYNNE (1978) の Introduction to the algae" (Prentice Hall, New Jersey), VAN den HOEK (1978) の "Algen" (Einführung in the Phykologie, Georg Thieme Verlag, Stuttgart) である。そして, 昨年, 今回の新刊紹介として取り上げる上記の本が出版された。なぜ, ここ数年で 4 冊もの教科書が出版されたのであろうか, その原因を考えてみると著者も書いているが, 1950 年代から電子顕微鏡を使った, 藻類の鞭毛構造, 細胞構造の観察結果が報告されるようになり, 特にここ 20 年来, 固定・包埋・切片作成技術等の急速な進歩から, 藻類細胞の微細構造の新しい知見がふえてきた。それにもとずいて, 従来の分類体系が見なおされ, 新しい門や綱が設立され, また新しい系統樹が提唱されるようになった。また, 特に最近は緑藻類を中心にした細胞分裂機構や鞭毛装置構造の研究は目をみはるほどの進歩をとげた。その他に, 生化学・生理学的分野の進歩を見のがせないし, 培養技術の発達とともに従来から進んでいた生活史の研究もさらに進歩した。このようなことから, 新しい知見にもとずいた藻類の教科書を出版する必要にせまられたからであろう。まず Phycology の内容を紹介すると 16 章からなっており, 1. Basic characteristics of the algae（藻類の基本的特徴）. 2. Cyanophyceae（藍藻綱）. 3. Glaucophyta（グラウコキスチス門）. 4. Euglenophyceae（ミドリムシ綱）. 5. Dinophyceae（渦鞭毛藻綱）. 5. Cryptophyceae（クリプト藻綱）. 7. Chrysophyceae（黄金色藻綱）. 8. Prymnesiophyceae（プリムネシオ藻綱）. 9. Bacillariophyceae（珪藻綱）. 10. Rhaphidophyceae（ラフィド藻綱）. 11. Xanthophyceae（黄緑藻綱）. 12. Eustigmatophyceae（真正眼点藻綱）. 13. Phaeophyceae（褐藻綱）. 14. Rhodophyceae（紅藻綱）. 15. Chlorophyceae（緑藻綱）. 16. Charophyta（車軸植物門）, それに付録として glossary（用語説明）がふくまれている。どの章もほぼ同じような形式でかかれ, まず始めに細胞の構造の特徴が記述されている。これには細胞壁の構成物質, 鞭毛構造, 眼点, 葉緑体や色素体, その他細胞内小器官についてその形態, 構成物質などの特徴が書かれている。また, ほとんどの章で, 電顕で見たそれぞれの分類群の細胞の微細構造の図がのっている。その他に渦鞭毛藻, 珪藻, 緑藻などでは細胞分裂の過程について, その電顕での観察結果が記述されている。その後にその門や綱の特徴となる現象や生活史の記述がある。最後にそれぞれの門や綱に所属する目や科についてその特徴, 形態, 生活史などが代表的な種を取り上げて説明されている。また, この本では海藻として知られる褐藻, 紅藻, 緑藻が最後の方の章にまとまってのっており, かつかなりの頁数をさいて記述されている。全体を一読したところ, 非常に簡潔明瞭な文章で書かれており, 藻類全体を知るうえで重要な項目はほとんど網羅されている。上記に書いたように, 特にそれぞれの門や綱の特徴となる細胞構造や鞭毛構造などの新しい知見をもとにした門や綱の特徴が良くわかるようになっている。また, 現在まで分っている細胞分裂過程やその他電顕観察から得た新しい知見もかなり掲載されているし, 形態や生活史の図もかなり最近に報告されたものから転載されているので, これから藻類学を学ぶ学生には非常に便利である。しかし, それぞれの章で取り上げている属や種はかなり限定されており, それぞれの門や綱に所属する多種多様の種について知るには若干不便である。

この本で取り扱われた門や綱はほぼ CHRISTENSEN (1966) の提唱した分類体系に基づいているが, 若干の相違点がある。まず Glaucophyta を加え, Euglenophyceae を Chlorophyta から Chromophyta に移している。また, Craspedophyceae を藻類起原ではないとの意見から (PARK and DIXON, 1976) この本では削除されている。Prasirophyceae は綱として存続すべきでないとの STEWART *et al.* (1974) の主張に従って, 従来所属していた Chlorophyta の Volvocales に入れられている。また Chlorophyta は PICKETT-HEAPS (1971) の提唱した系統樹に従って書かれている。このように最近の知見を入れた形で編集されているが, まだ充分にその取り扱いについて論議がつくされていないことからも, 特にこの本のような教科書では, 門や綱の分類にはもうすこし慎重にあつかう必要があるのではないかと思われる。最後にこの本ではほとんど写真がないことと, 電顕写真から転写した図が少々雑であるのは残念である。

（東邦大・理・生　宮地和幸）