1. Product Code: _____

2. Finished Product Specifications:

	Limits	Predicted
a. SFI (°F)		
(1) 50		
(2) 70		
(3) 80		
(4) 92		
(5) 104		
b. Iodine Value (IV)		
c. Melting Point (°F)		

3. Formulation:

Base Stock	Quantity (Lbs.)	Storage Tank No.
Total		

4. Finished Product Cost ($/Lb.): _____

FIG. 6. Computer output format for product blending.

ties), improved product quality consistency, and reduced quality control laboratory workload.

- Reduced workload for higher-skilled personnel involved with blending (opportunity to reduce staffing requirements).
- Availability of actual product cost by lot.
- Increased profit margin from most favorable cost component blending within restraints imposed.
- Achievement of a significant step in developing computer controlled and integrated manufacturing.

Summary

Production of multi-component edible oils is both technically demanding and financially significant. Conventional methods used in product blending have serious shortcomings in achieving optimum product quality and financial results. Computer application to product blending provides the opportunity to consistently attain "best case" product quality and financial objectives. The use of computers for product blending can be as simple as a single personal computer for performing "best fit" blend calculations or as complex as networking departmental functions for relevant data access (financial, quality control and production) and for control of the production processes and operations involved.

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Market applications for microalgae

Alternate sources of fats and oils are being sought on an ongoing basis, with particular attention being paid to novel specialty products of possible high commercial value. At the request of J.B.M. Rattray, Associate Editor for JAOCS *News for Biotechnology, David Kyle of the Martek Corp. in Columbia, Maryland, has prepared the following article on potential applications of microalgae and their relevance to the fats and oils industry.*

Microalgae represent a subset of single-cell microorganisms that generally grow autotrophically using CO_2 as the sole carbon source and light as energy. Some species of microalgae, however, are heterotrophic and can use different forms of organic carbon as nutrient sources.

Although microalgae most commonly inhabit an aquatic environment, they are ubiquitous in nature and have been identified in almost every ecological niche on this planet. Aquatic microalgae have been isolated in areas ranging from hot springs to glacial ice flows; terrestrial species have been identified from the desert to the arctic tundra. Some species are intercellular symbionts with fungi (i.e., lichens) or marine sponges and hydrazoans. In total, there are thought to be over 50,000 different species of microalgae, of which only a few have been characterized in any detail.

This lack of detailed information on microalgae compared with other microorganisms (i.e., bacteria, yeast and fungi) is primarily a consequence of their unconventional cultivation. As a result, they generally have not been included in large-scale industrial screening programs until very recently. Thus, microalgae represent a major untapped resource of genetic potential for valuable bioactive agents or special purpose biochemicals. I will address the question of how this potential resource can be exploited and identify some of the

(Continued on next page)

limitations for industrial uses of microalgae.

Commercial manufacturing

There are several possibilities for commercial production of microalgae and/or their products. The choice of an open pond or "farming" strategy versus the photobioreactor or fermentation-like strategy depends entirely on the product and its ultimate use.

For example, *Spirulina* has been harvested for hundreds of years from Lake Chad and used as a food source in Central Africa. Because of its high protein content (>60%) and digestibility, it has recently been cultivated in large ponds by private industry in Israel, Mexico and the U.S. In the U.S., it has been targeted for the health food market. However, problems with the quality of the product produced in these open systems (e.g., excessive contamination with insects) led to the U.S. Food and Drug Administration (FDA) banning *Spirulina* from the Mexican source. *Chlorella,* another microalgal food supplement, also has been grown in open ponds on a commercial scale in Japan and Southeast Asia. *Dunaliella,* a natural overproducer of carotenes, has been grown in open ponds in Israel, Australia and the U.S. for the production of foodgrade β-carotene. In this case, however, the carotenoid is processed from the microalgae and the importance of quality control is at the processing plant stage, not at the pond level.

The principal advantages of open-pond cultivation systems include a relatively small capital investment for production of the biomass and a free energy source for the growth of the organisms (i.e., sunlight). Because culture ponds are dependent on the environment for temperature control and light, their productivity is affected by environmental fluctuations. For this reason, the ponds are usually situated in areas with stable climatic conditions. The open nature of the ponds not only allows the possibility of chemical and biological contamination from nonalgal sources, but prevents the growth of many microalgae in such systems because they are not competitive with microalgal "weed" species borne by aerial contamination.

The reason for the successes with *Spirulina* and *Dunaliella* is that these species are vigorous under conditions of high alkalinity (pH>10) and high salt (>2M NaCl), respectively. *Chlorella,* on the other hand, is so vigorous in fresh water under neutral pH that it is unlikely any weed species could compete under these conditions. In fact, no matter what species is inoculated in such a pond, the arrival of airborne *Chlorella* usually results in a full-scale *Chlorella* bloom. However, since other algal species are known to bloom under certain conditions in the wild (e.g., the red tide of *Gonyaulax,* or diatom, blooms in waste treatment plants), we can be encouraged; if the exact conditions leading to the bloom are understood, they can be used to establish a successful open-pond cultivation system.

Such studies are currently under way, sponsored by both the public and private sectors. A biotechnological alternative to increasing species' competitiveness involves the genetic modification of a microalgal strain establishing a resistance to some compounds (i.e., certain metals, salts or herbicides) which is toxic to contaminating species. Although this line of research is being investigated, no such bioengineered strains are presently in commercial use.

The antithesis of the open pond is the electric light-driven photobioreactor. Such a system is capital intensive, bears high operating costs in terms of electricity and is more appropriately used for the production of high-value products. This strategy for microalgal cultivation offers several unique advantages that make it preferable to open ponds for commercial-scale production in many cases.

Such systems have regulatable illumination for up to 24 hours a day rather than a variable 8 to 12 hours of sunlight impinging on the ponds. Culture depth or optical pathlength in a photobioreactor can be kept to a minimum (1–3 cm) without fear of evaporation. (Ponds depend on evaporation for cooling and a certain depth is required to not endanger the pond's conditional status.) The shorter optical path allows a ten- to hundred-fold improvement in biomass density over the open ponds before light compensation is reached (i.e., the point at which photosynthesis equals respiration and there is no net increase in biomass). The higher cell density results in reduced harvesting volumes and reduced processing costs. Furthermore, the increased duration of illumination and a much tighter control of growth conditions result in much faster growth rates. Thus, in many cases, the greater productivity of a photobioreactor and the simplicity of downstream processing may more than justify the additional capital and operating costs for the commercial production of a particular algal product.

The antithesis of the open pond is the electric light-driven photobioreactor.

Between the two extremes (the low-technology open pond and the high-technology electrically driven photobioreactor) is a middle ground—enclosed photobioreactors that use sunlight as an energy source. These generally are tubular in design. Pilot units up to 5000 liters are currently being tested in Europe. A large-scale tubular unit in the United Kingdom is housed indoors for temperature control. It is angled toward the sun for maxi-

mum light utilization and can be supplemented with artificial lighting at night. A horizontal outdoor tubular unit in France is raised or lowered into an outdoor pond for temperature control.

Such systems have both the operational advantage of the controlled photobioreactor (as well as a high capital investment) and the reduced energy costs of the ponds (because they use sunlight for energy). Consequently, the choice of microalgal culture geometry for commercial-scale production will depend on the value of the product and the culture requirements of the organism itself.

Market niche for microalgae

There are only a few examples of microalgal cultivation that have led to commercial application. As noted before, *Spirulina* has been—and continues to be—produced in quantity for sale to the health food industry. This is a relatively small and variable market. On the other hand, β-carotene represents a very large market, but currently its production in large open ponds using *Dunaliella* is not competitive with the chemical synthesis. Given the perception of being a value-added, "natural" source (i.e., at least two times the value of the synthetic β-carotene), an efficiently run pond with no major losses should be able to operate profitably. However, it should be remembered that the market for the "natural" β-carotene is only a small fraction of the total market in the U.S.

Other pigments such as phycocyanin and phycoerythrin are produced by cyanobacteria (i.e., *Spirulina*) and have recently been used as fluorescent labeling agents. They are proteinaceous in structure and exhibit a high extinction coefficient and fluorescence. Consequently, they can be used as cosmetic pigments. Overall, this represents a small specialty market; although these pigments can be obtained from *Spirulina* cultivated in open ponds, there may be interest in genetically engineered organisms with altered pigments. Using patentable strains, a developer can easily protect his invention.

One future market application for microalgae might be in the production of specialty lipids.

One future market application for microalgae might be in the production of specialty lipids. For example, the omega-3 fatty acids, found in the oils of certain cold-water marine fish and thought to be responsible for the reduced incidence of coronary heart disease in populations which consume large quantities of fish, likely originate from the phytoplankton in the food chain. Indeed, many of these phytoplankton species are found to be rich in reserves of oil containing various amounts of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA).

The commercialization of this concept will depend on its competitiveness with fish oil as a source of these fatty acids. Commercial success of this concept will require improving the fat and EPA contents of the microalgae themselves (i.e., selecting overproducing clones), reducing the cultivation costs, exploiting the added value of a controlled production process (compared to harvesting fish), and developing a higher quality oil containing large amounts of a single bioactive fatty acid (as compared with fish oils which contain a mixture of many metabolically active fatty acids). Without genetic improvement, it is unlikely that any EPA-containing product will be produced from microalgae that is competitive with refined, deodorized fish oils for food or pharmaceutical use. This fact itself may rule out the possibility of using open ponds for cultivation unless the competitiveness of the genetically modified clone can be enhanced by inserting a resistance factor as described previously.

As an alternative, we might consider a fermentation-like approach using algal strains capable of heterotrophic growth. Such systems have been commercialized in the United Kingdom for the production of specialty oils (containing gamma linolenic acid) using fungi. If using very low-cost carbon sources, conventional fermentation technology and heterotrophic algal strains results in production costs approaching those of refined fish oils, then the production of "designer" EPA- and/or DHA-containing oils by microalgae could be commercially feasible.

Exploiting autotrophy in microalgae as a method of producing labeled biochemicals has been used for many years. This involves the substitution of tritiated water(^3H_2O) for ^1H_2O or $^{14}CO_2$ for $^{12}CO_2$ and results in the production of radioactively labeled cells which can then be used for the isolation and purification of various labeled biochemicals. The recent growing interest in stable isotopically labeled biochemicals as replacements for their radioactively labeled counterparts has resulted in the use of the same strategy with the substitution of heavy water (D_2O) or $^{13}CO_2$. In these latter cases, one can produce enrichment levels up to 100% depending on the isotope enrichment of the culture medium. These specialty biochemicals have a very high value (>$200/g) and are generally manufactured in small quantities. Consequently, the electric light-driven photobioreactor is the culture method of choice. Uses of these stable isotopically labeled compounds include production of very high stability deuterated lubricants, a labeled nutrient medium in which to grow cells for metabolite or recombinant protein structure determination using NMR, new NMR imaging applications, and diagnos-

tic breath tests in the clinical setting.

The future

Where is this new technology headed? It is clear that the market for stable isotopically labeled biochemicals is increasing, and the most economically attractive sources of the ^{13}C- and ^{2}H-labeled compounds are autotrophic microalgae. It appears unlikely that production concepts other than using microalgae will be used in the near future for this market. If diagnostic tests are developed using these compounds, the market will increase dramatically.

On the other hand, markets for "natural" β-carotene and *Spirulina* are dependent on consumer perception of value in a "natural" product; demand is unlikely to increase significantly in the near term unless production costs can be reduced.

The importance of microalgae as an alternative source of omega-3 fatty acids will become more significant as these products find their way into the food industry. Microalgae can provide a "designer oil" specially tailored to the food industry. Also, based on the assumption that at least 1% of our daily fat intake should be in the form of EPA (the consensus reached at the AOCS Short Course on Polyunsaturated Fatty Acids and Eicosanoids, held in Biloxi in 1987), there simply is not enough fish oil available to meet future demand. In this case, commercial-scale production of EPA from microalgae may become more attractive economically.

Finally, let's return to my original statement about the "untapped" resource of bioactive compounds. Screening of bioactive agents from any microorganism is always risky, but the payback is very large. As we begin to see more of the microalgal species screened as potential antiviral, antiAIDS, antibiotic and other bioactive agents, we may see the appearance of more uses for the microalgal industry. This may lead to the realization that microalgae are indeed only a somewhat unconventional type of microorganism.

Biotech update

The following items of relevance to biotechnology and the fats and oils industry are provided for informational purposes. J.B.M. Rattray, Associate Editor for JAOCS *News for Biotechnology, compiled this listing.*

Conferences, meetings

3rd Annual Seminar on Analytical Biotechnology, May 22-25, 1989, Baltimore, Maryland. Information: Janet Cunningham, Barr Enterprises, PO Box 279, Walkersville, MD 21793.

7th Annual DECHEMA Meeting of Biotechnologists, May 30-31, 1989, Frankfurt am Main, West Germany. Information: DECHEMA, Abt. Tagungen, Postfach 97 01 46, D-6000 Frankfurt am Main 97, West Germany.

First Eurolipid Congress, June 7-9, 1989, Angers, France. Information: French Association for the Study of Fats (AFECG), 10A rue de la Paix, 75002, Paris, France.

4th European Conference on Industrial Biotechnology, June 12-14, 1989, Varese, Italy. Information: Sergio Merli, Farmitalia Carlo Erba 35, Via Dei Gracchi 35, 20146, Milan, Italy.

The 1989 ASM Conference on Biotechnology, June 22-25, 1989, Orlando, Florida. Contact: ASM Meetings, 1913 Eye St., NW, Washington, DC 20006.

Progress in Recombinant DNA Technology and Applications, June 25-30, 1989, St. Charles, Missouri. Information: Washington University Biotechnology Course, Campus Box 1198, St. Louis, MO 63130.

International Conference on Biotechnology, July 5-7, 1989, Beijing, People's Republic of China. Information: Secretariat, Chinese Society for Microbiology, Zhongguancun, Haidian 10080, Beijing, People's Republic of China.

Annual Meeting of Society for Industrial Microbiology, Aug. 13-18, 1989, Seattle, Washington. Information: Ann Kulback, Society for Industrial Microbiology, PO Box 12534, Arlington, VA 22209-8534.

Biotechnology: Principles and Processes, Aug. 14-18, 1989, Cambridge, Massachusetts. Information: Director of Summer Session, MIT, Room E-19-356, Cambridge, MA 02139.

Biochemical Engineering Summer Course, Sept. 4-8, 1989, London, United Kingdom. Information: Lynne Mason, Department of Chemical and Biochemical Engineering, University College London, Torrington Place, London, WC1E 7JE, UK.

Industrial Bioprocessing Short Course, Sept. 24-28, 1989, Colorado. Contact: Bruce Dale, Department of Chemical Engineering, Texas A&M University, College Station, TX 77843.

AchemAsia '89, Oct. 11-17, 1989, Beijing, People's Republic of China. Contact: DECHEMA, Organization AchemAsia, Postfach 97 01 46, D-6000, Frankfurt am Main 97, West Germany.

International Biotechnology Exposition, Oct. 24-26, 1989, San Mateo, California. Information: Linda H. Cartlidge, Cartlidge & Associates Inc., 3097 Moorpark Ave., Suite 202, San Jose, CA 95128.

Cell Culture Engineering II, Dec. 10-15, 1989, Santa Barbara, California. Contact: Engineering Foundation, 345 E. 47th St., New York, NY 10017.

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