

## Chapter 6

# MANAGEMENT OF AUTOTROPHIC MASS CULTURES OF MICRO-ALGAE

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## INTRODUCTION

Interest in the mass cultivation of micro-algae as feed and foodstuff has existed since the turn of the century (Robinson and Toerien, 1982). Experiments using algae in photosynthetic research (Warburg, 1919) also led to an appreciation of the rapidity of growth of certain micro-algae. Algal mass-culture research was initiated in the 1940s, first in Germany (Harder and Von Witsch, 1942) and later in many other countries including The Netherlands (Wassink et al., 1953), Japan (Tamiya, 1957), Czechoslovakia (Necas and Lhotsky, 1967), Rumania (Vendlova, 1969), the Soviet Union (Gromov and Pinevich, 1972), France (Clement, 1975), Belgium (De Pauw et al., 1980), India (Becker and Venkataraman, 1980), Israel (Shelef et al., 1980), Italy (Balloni et al., 1980), Mexico (Durand-Chastel, 1980), Peru (Castillo et al., 1980), South Africa (Toerien and Grobbelaar, 1980), Taiwan (Soong, 1980) and the United States (Oswald, 1982).

Current interest in algal mass cultivation is not confined to the production of single cell protein but also includes the use of algae in waste-water treatment (Oswald and Golueke, 1968; Shelef et al., 1976), in aquaculture (Persoone and Clause, 1980), in closed life-support systems as in space travel (Oswald et al., 1965), in the production of fine chemicals (Aaronson et al., 1980) and health food (Kawaguchi, 1980), and in the bioconversion of solar energy (Goldman and Ryther, 1977; Ashare et al., 1978).

Algal mass-cultivation technology varies from the extremely unsophisticated to the highly sophisticated (Soeder, 1980). Mass cultivation systems are operated on defined or semi-defined media

called "clean cultures", or on wastes or effluents. In the latter case the product contains not only algae, but also other organisms, notably bacteria, and is referred to as algal-bacterial biomass (Soeder, 1980). The "clean culture" systems may further be sub-divided according to their mode of growth as autotrophic (production by photosynthesis alone), mixotrophic (production by photosynthesis and heterotrophy) and heterotrophic modes (production by heterotrophy alone) (Stengel, 1970). We are limiting this discussion to autotrophic "clean culture" systems.

## TYPES OF MICRO-ALGAE

Certain micro-algal species tend to dominate in mass-culture systems, regardless of which algae are used as inocula (Goldman, 1979a). These include the "weed" species of the genera *Chlorella*, *Micractinium* and *Scenedesmus* in fresh-water systems and *Phaeodactylum* and *Skeletonema* in marine systems (Goldman and Ryther, 1976; Goldman, 1979a, 1980). However, in "clean culture" systems a single algal type can be maintained over extended periods — as, for example, *Scenedesmus obliquus* (Stengel and Soeder, 1975), *Coelastrum probiscideum* (Soeder, 1978; Mohn, 1982) and *Spirulina platensis* (Vonshak et al., 1982). For instance, *Coelastrum probiscideum* was cultured continuously on a semi-technical scale in Germany for at least seven years (Soeder, 1978).

The use of extreme environmental conditions tends to select for specific algae. For example,

*Dunaliella* is produced in highly saline conditions (Aaronson et al., 1980), and *Spirulina* in high-bicarbonate environments (Clement, 1975; Durand-Chastel, 1980). High temperatures could select for thermotolerant algae, although mesophilic algae such as *Scenedesmus acutus*, which have a laboratory temperature limit of 34 to 35°C, can survive short-term heating to 42°C (Stengel and Soeder, 1975).

#### DESIGN OF CULTURE UNITS

Culture units currently in use vary from small experimental units (Goldman and Ryther, 1975; Grobbelaar, 1981a) to large ponds of several or many hectares in area (Shelef et al., 1978; Durand-Chastel, 1980). Whereas early work was concerned with the reliability of cultivation units, the emphasis is now on energy-saving designs (Soeder, 1981).

Designs include horizontal raceways (Shelef et al., 1978), sloping cascade systems (Setlik et al., 1970), sloping raceways (Heusler et al., 1978a), circular ponds (Kanawawa et al., 1958; Goldman and Ryther, 1975), long troughs with flat or zig-zag bottoms (Morimura et al., 1955; Clement et al., 1980), U-shaped ponds (Ben Amotz and Avron, 1980), tubular systems (Burlaw, 1953), and ponds covered with transparent plastic (Walmsley et al., 1981).

Two most important considerations in the design of mass algal cultures are culture depth and agitation. Circulation or turbulence prevents sedimentation, allows contact of cells with nutrients, and improves light utilization (Persoone et al., 1980). For autotrophic production a water depth of 15 to 20 cm and paddle-wheels for agitation are recommended (Soeder, 1981). Pumping is more suitable for deeper cultures (Setlik et al., 1970; Goldman and Ryther, 1976).

Algal mass-culture units are designed and operated to maximize the utilization of sunlight energy (Soeder, 1981). A major disadvantage in algal biomass production is the low-energy flux of sunlight (Myers, 1977). Photosynthesis is limited by: (1) its dependence on visible light (photosynthetically active radiation or PAR) which is about 45% of total solar radiation; (2) reflectance losses (10–20%); (3) a maximum possible quantum efficiency (about 25%); and (4) metabolic losses. A wide variety of pond designs are thus used in mass

cultivation of autotrophic algae. No clear consensus exists at present about an optimal design (Robinson and Toerien, 1982).

#### FACTORS AFFECTING PRODUCTIVITY

A multitude of factors determine the productivity of algae in algal mass-cultivation systems (Goldman, 1979a,b, 1980). The success of mass cultivation is dependent upon yield optimization.

#### Yields

Average yields of 15 to 25 g m<sup>-2</sup> day<sup>-1</sup> a weight in open ponds are now common, compared with yields of about 2 to 15 g m<sup>-2</sup> day<sup>-1</sup> attained in the 1950s and early 1960s (Goldman, 1979, 1980; Soeder, 1980). Maximum yields attained over short periods are in the range of 30 to 50 g m<sup>-2</sup> day<sup>-1</sup> (Goldman, 1979b, 1980) but yields up to 54 g m<sup>-2</sup> day<sup>-1</sup> have also been reported (Heusler et al., 1978a; Grobbelaar, 1981a). Bassham (1977) suggested that higher maximum yields are still possible.

A distinction must be made between gross yield which is the increase in algal biomass per surface area per unit time, and net yield, which represents the actual yield in terms of final product per unit surface area per unit time (Soeder, 1981). The difference may be from 25 to 30% of the gross yield for pond areas of 0.1 to 0.2 ha and 10% for pond surfaces of 6 ha (Soeder, 1980). Operational losses during harvesting and processing, plant infections, and unavoidable standstill periods needed for maintenance and/or cleaning are a part of this difference (Soeder, 1980).

The yields of "closed" ponds, covered with transparent plastic (Fig. 6.1), were investigated by Walmsley and Shillinglaw (1984). A pond 100 m<sup>2</sup> operated for a year with a fixed retention period of four days gave a mean yield of 10 g m<sup>-2</sup> day<sup>-1</sup>, whilst the average of peak values was approximately 15 g m<sup>-2</sup> day<sup>-1</sup>. In miniature ponds they found that yields ranged from 7.9 g m<sup>-2</sup> day<sup>-1</sup> (retention period = 12 days) to 16.6 g m<sup>-2</sup> day<sup>-1</sup> (retention period = 2 days) for systems covered with a single layer of plastic.

The net yields that may therefore be expected



Fig. 6.1. "Closed" algal ponds at the University of the Orange Free State, Bloemfontein, South Africa. Left: 100 m<sup>2</sup> pond; right: some of the miniponds.

from "open" autotrophic algal ponds range from 25 to 80 t ha<sup>-1</sup> yr<sup>-1</sup> (Soeder, 1980). Enclosing ponds with plastic will apparently not adversely affect yields because production is stimulated by enclosure which elevates the water temperature.

### Light

The productivity of algae in autotrophic mass-cultivation systems should be a function of irradiation. Castillo et al. (1980) found a linear relationship between yield of *Scenedesmus* and irradiation. Significant linear relationships were also found for "open" (Mostert, 1982) and "closed" systems. Goldman (1979b, 1980) developed a model of productivity and irradiation (see also Tamiya et al., 1953) which suggests a non-linear relationship between yield and irradiation, with yields levelling off at high irradiances. Application of the Goldman model to the results with "closed" ponds resulted in an excellent agreement between predicted ( $X$ ) and observed ( $Y$ ) production values ( $Y = 0.91X + 1.29$ ,  $n = 11$ ,  $r^2 = 0.97$ ) (Walmsley and Shillinglaw, 1984). More data are needed to resolve the linearity or non-linearity of the relationship.

Goldman (1980) pointed out that his model is sensitive to the magnitude of  $I_s$ , the light saturation constant used in the model<sup>1</sup>. Goldman (1979b, 1980) used values ranging from 0.02 to 0.06 cal cm<sup>-2</sup> min<sup>-1</sup> for  $I_s$ , and he suggested that too little is at present known about this constant; it should be estimated from data on the relationship between growth rate and light intensity, or productivity and

light intensity. Productivities of "closed" ponds were consistent with an  $I_s$  value of 0.02 cal cm<sup>-2</sup> min<sup>-1</sup>. Van Vuren and Grobbelaar (1982) reported  $I_s$  values of about 7% of full sunlight for green algae selected for use in "open" algal ponds.

Similarly, the roles of photorespiration, light inhibition and respiration as factors controlling productivities should be clarified (Goldman, 1980). Shelef et al. (1978) postulated that respiration losses are probably negligible, while Grobbelaar (1981b) suggested that they may exceed 10% in some cases.

Photoinhibition and photorespiration might not be overly important in algal mass cultures (Goldman, 1980), but structural changes were detected in the thylakoids (lamellar photosynthetic structures within the algal chloroplasts — see Fig. 6.2) of green algae in "closed" ponds after a few hours of exposure to sunlight (S.N. Shillinglaw, unpubl. data). Ryther (1956) estimated that light inhibition was detected in marine algae at approximately 10% of full sunlight intensity. Studies of primary production by <sup>14</sup>C assimilation assays suggested that photoinhibition can occur at the surface of algal mass cultures when static assays<sup>2</sup> are used. However, the highly turbulent conditions in such systems could prevent photoinhibition from occurring, and some dynamic <sup>14</sup>C assays should be performed to test this hypothesis.

The use of a number of "closed" miniponds, which could be covered with varying layers of plastic material, enabled interesting studies of light-production relationships (R.D. Walmsley, unpubl. results). Enclosure of a pond with one layer of "Uvidek" plastic reduced production only fractionally (less than 3.5%), despite a 20% reduction in light supply. The light saturation characteristics of the algal photosynthetic system could have been the cause of this phenomenon.

<sup>1</sup> Saturation light intensity ( $I_s$ ) is the maximum light intensity that can be accommodated by the photosynthetic apparatus. When the prevailing intensities are higher than saturation, the excess light is not used, and the photosynthetic activity proceeds at a constant level corresponding to that of  $I_s$  (Shelef et al., 1968).

<sup>2</sup> Static assays refer to incubations where a bottle is maintained at a fixed depth, whereas in dynamic assays all bottles are rotated throughout the water column for the full period of the assay.

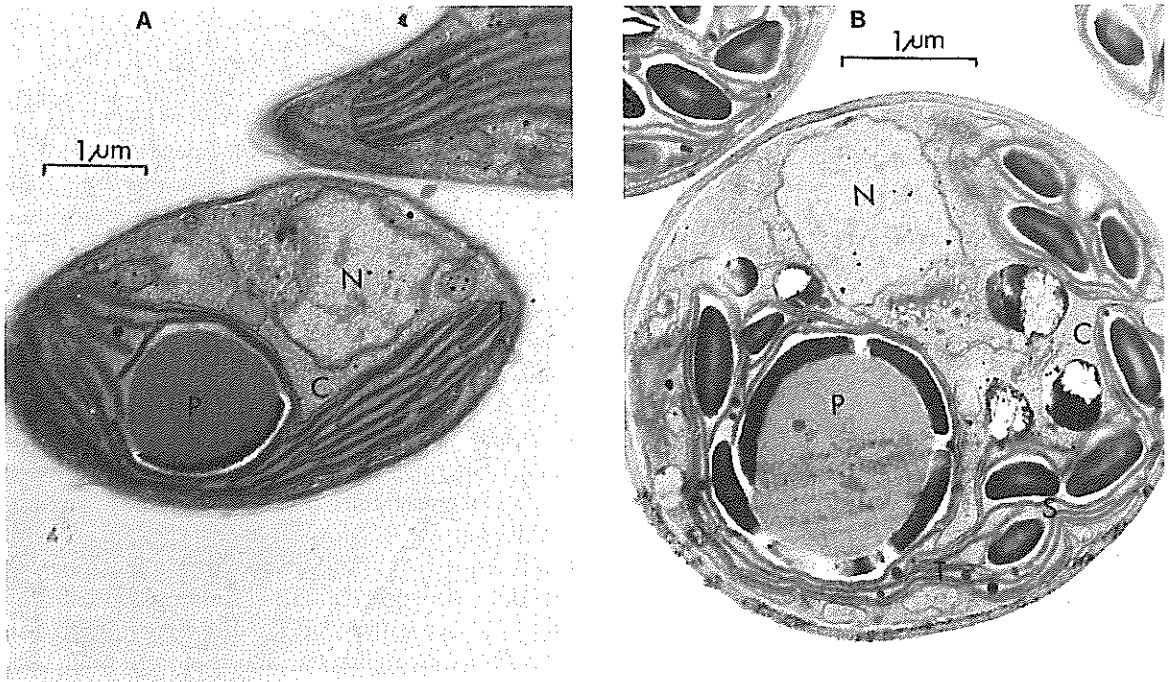


Fig. 6.2. Internal structures of algae from outdoor cultures. A. Overnight in dark. B. After 4 h in light ( $c. 2000 \mu\text{Einsteins m}^{-2}$ ).  $P$  = pyrenoid;  $N$  = nucleus;  $C$  = chloroplast with  $T$  = thylakoids or lamellar photosynthetic structures,  $S$  = starch granules. (Pho S.N. Shillinglaw.)

Estimates of the efficiency of conversion of solar energy into algal biomass vary considerably. Pirt et al. (1980) reported some very high efficiencies for laboratory cultures, but Benemann et al. (1977) and Bassham (1977) postulated maximum efficiencies of between 5 and 6.6% for the use of photosynthetically active radiation (PAR), with a concomitant average annual productivity of  $61 \text{ g m}^{-2} \text{ day}^{-1}$  in the United States. The latter estimate is equal to the maximum possible yields postulated by Goldman (1979b, 1980). Radmer and Kok (1977) reported maximum efficiencies in the use of PAR of 10 to 12% under the most ideal conditions, but in most aquatic systems efficiency rarely exceeds 1 to 2%, primarily because other factors such as nutrients are limiting. Maximum yields observed by Grobbelaar (1982) were consistent with PAR utilization efficiencies of about 8%. However, the average yield values observed for the "open" systems used by Grobbelaar corresponded with efficiencies of 2.0 to 3.1% (Mostert, 1982). The light utilization efficiency increased in covered ponds to a maximum value of 5.8% as the number of plastic layers was increased,

thereby decreasing light availability (Walr and Shillinglaw, 1984).

### Temperature

Temperature and light are both growth-limiting factors to algal mass cultures in winter, as shown by geographic locations such as Massachusetts (U.S.A.), Belgium, Israel and Germany, because of cloud cover and low temperatures (De Pauw et al. 1978; Shelef et al., 1978). Goldman and Ry (1976), Goldman (1977) and Payer et al. (1977) postulated that temperature is probably not as important as light in controlling maximum potential productivity. However, Goldman (1980) derived an equation:

$$P' = 0.087 db (1.066)^{2t}$$

where  $P'$  is maximum productivity per unit surface area,  $d$  is depth of the culture,  $b$  is an empirical constant and  $t$  is temperature in  $^{\circ}\text{C}$ , to describe the influence of temperature. Castillo et al. (1980) and Mostert (1982) reported linear relationships

tween yields of "open" algal mass-cultivation systems and culture temperatures. Open, covered, and open heated miniponds were used by Walmsley and colleagues to elucidate the role of temperature in algal mass cultures. The covered minipond had temperatures higher than those of the open minipond, but the temperatures of the open heated pond were electronically regulated to be the same as those of the covered pond. The difference in productivities between the covered and open heated miniponds were very small, but the productivity of the open pond was 10 to 48% lower than those of the other ponds. Temperature was evidently an important controlling factor in these miniponds, during both the summer and winter.

Castillo et al. (1980), Mostert (1982) and Geldenhuys (1983) presented evidence of interrelationships of algal productivities with both light and temperature. Although such interrelationships may be spurious (e.g. the highest productivities are encountered in summer, when both temperature and light have high values), nevertheless true interrelationships may exist. For instance, the light-saturated productivity of algae is probably a function of temperature only (Harris, 1978, 1980; Stegmann, 1982). Consequently,  $I_s$  must also be a function of temperature (Harris, 1978; Stegmann, 1982). The net result of the above would be an interdependence of productivity on both light and temperature. The mathematical model developed by Grobbelaar (1981b, 1982) predicts such an interdependence, and the results of Vonshak et al. (1982) for *Spirulina platensis* in mass culture confirmed this trend. Their well-stirred cultures never reached light or temperature saturation, even in summer in the Negev desert of Israel. In "closed" ponds the "heat trap" effect and resultant maximum temperatures are of concern. In such ponds, summer water temperatures exceeded 45°C at times, but the cultured algae (mainly *Ankistrodesmus*, *Chlorella* and *Scenedesmus* species) were capable of maintaining reasonable production rates ( $>10 \text{ g m}^{-2} \text{ day}^{-1}$ ) (Walmsley et al., 1983). Temperature is evidently more important in algal mass culturing than envisaged by Goldman (1980). However, he considered temperature to be of critical importance in controlling species competition in mass cultures.

## Nutrients

When nutrients are provided in excess and light is the growth-limiting factor, most algal species display a remarkable consistency in their chemical composition: 45 to 50% carbon, 8 to 10% nitrogen and 1% phosphorus (Goldman, 1980). The high carbon content of algae necessitates a carbon dioxide supply for the mass cultivation of autotrophic algae. The average demand is about  $45 \text{ g CO}_2 \text{ m}^{-2} \text{ day}^{-1}$  (Soeder, 1980), but is dependent on the yield achieved. Carbon dioxide supply for mass cultures is usually expensive, and the most suitable sources are combustion gases and the waste gas from fermentation processes (Soeder, 1978). Carbon dioxide utilization in "closed" cultures appears to be more efficient owing to prevention of atmospheric losses (Robinson and Toerien, 1982), which can also be reduced by adding the gas through special apparatus (Heusler et al., 1978b). In experimental systems carbon dioxide supply on demand can be used to maintain preselected pH values (Grobbelaar, 1981a).

The supply of nitrogen and phosphorus compounds to mass cultures of autotrophic algae must be in the range of  $1.6$  to  $6.5 \text{ g N m}^{-2} \text{ day}^{-1}$  and  $0.2$  to  $0.83 \text{ g P m}^{-2} \text{ day}^{-1}$  (Robinson and Toerien, 1982). In terms of concentration, nitrogen supply has varied from  $25 \text{ mg l}^{-1}$  (Shelef et al., 1980) to  $5000 \text{ mg l}^{-1}$  (De Pauw et al., 1980). Mostert (1982) reported optimal nitrogen concentrations in the input water to "open" mass cultures of 25 to  $50 \text{ mg l}^{-1}$ , but Eyster (1976) reported a value of  $350 \text{ mg l}^{-1}$  for *Chlorella sorokiniana* cultures. Mostert (1982) found that a ratio of nitrate to ammonium nitrogen of 3:1 yielded higher productivities than when either form of nitrogen was supplied alone.

Geldenhuys (1983) evaluated the optimal supply of nitrogen and phosphorus to "closed" cultures and recommended  $30 \text{ mg N l}^{-1}$  (or  $1.89 \text{ g N m}^{-2} \text{ day}^{-1}$ ) and  $4.5 \text{ mg P l}^{-1}$  (or  $0.28 \text{ g P m}^{-2} \text{ day}^{-1}$ ). He also found that all the phosphorus in dissolved superphosphate and the nitrogen in dissolved urea fertilizers are available to algae. Superphosphate was an adequate source of microelements for algal mass cultures, but urea and tap water were not (Geldenhuys, 1983). Tap water together with fertilizers also supplied sufficient of all other major nutrients needed by the algae (Grobbelaar, 1979; Geldenhuys, 1983).

### Algal population density

Optimal productivities in mass cultures are dependent upon optimal concentrations of algal biomass (i.e., optimal areal densities) (e.g. Grobbelaar, 1981b, 1982). Optimum areal density is a function of culture depth, and Grobbelaar (1981b) determined from a model that an areal density range of 16 to 22 g C m<sup>-2</sup> was required for maximum productivity. This optimal areal density range has been confirmed in outdoor experiments where the cultures were maintained at a preselected areal density by daily dilutions with fresh nutrient medium (Grobbelaar, unpubl. results). Vonshak et al. (1982) found that population density was a major factor in the production of *Spirulina platensis*. The photosynthetic capacity, the specific respiration rate and the specific growth rate decreased with increased population densities, whilst the productivity peaked at a population density between 300 and 400 mg l<sup>-1</sup> (Vonshak et al., 1982). Optimal population densities also coincided with the highest concentrations of oxygen in ponds, and Vonshak et al. (1982) indicated that oxygen concentration is a sensitive indicator for detecting divergence from steady state (culture deterioration).

### Residence time (algal growth rates)

Under light-limiting conditions, productivity will be maximal at some specific growth rate (or residence time) of a mass culture. At lower or higher residence times the productivity will be less. Thus, productivity has a bell-shaped curve with respect to growth rate (Goldman, 1980). Goldman (1979b) postulated that the optimum growth rate (or maximum yield) defines the condition of practically complete light absorption over the entire culture depth. Eppley and Dyer (1965) and Oswald (1970) postulated that the optimum growth rate for maximum productivity was 50% of the maximum algal growth rate. Goldman and Ryther (1975) and D'Elia et al. (1977) offered support for these concepts.

In "closed" cultures, evidence was obtained for a skewed bell-shaped curve as the relationship between productivity and growth rate, but the increase of biomass concentration with growth rate was non-linear (Walmsley et al., 1983). Goldman

and Ryther (1975) and D'Elia et al. (1977) reported an inverse linear relationship between cell concentration and growth rate.

Optimal growth rates (Goldman, 1980) and optimal population areal densities (e.g. Grobbelaar 1982) probably reflect the same underlying phenomenon, namely optimal light absorption in mass cultures.

### Turbulence

Turbulence is important in mass cultivation for a number of reasons: (a) prevention of sedimentation; (b) the recurrent exposure of algal cells to light; and (c) improving nutrient exchange between algal cells and the medium by eliminating nutrient gradients. Water movement in mass cultures varies from a few centimetres per second to 50 cm s<sup>-1</sup> (Soeder, 1981; Walmsley et al., 1983).

Grobbelaar (unpubl. data) investigated the effect of turbulence on productivity of three rates of water movement — 25, 37 and 57 cm s<sup>-1</sup>. Overall, there was no evidence of differences in productivity with different rates of stirring (Fig. 6.3). Goldman (1980) indicated that knowledge of the effects of turbulence on parameters such as  $I_0$  might be important in the optimization of algal mass cultivation.

### Infections

Outdoor mass cultures of algae are susceptible to infections by bacteria, fungi, foreign algae or invertebrates (Grobbelaar, 1981b). Not much has been published on this topic (Gummert et al., 1981; Heusler et al., 1978b), which was reviewed by Soeder and Chutter (1981).

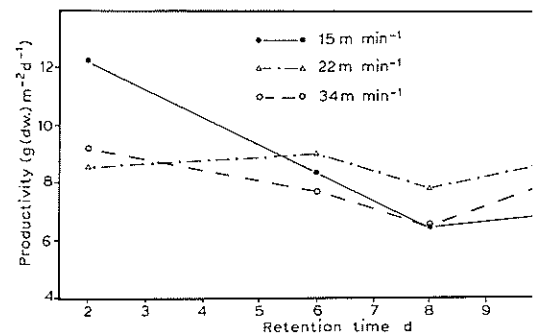


Fig. 6.3. Productivity as a function of turbulence and retention period in outdoor algal cultures.

Bacteria (Heusler et al., 1978b; Grobbelaar, 1981b) and microscopic fungi occur in algal mass cultures. Protozoa and rotifers are almost always present in outdoor mass cultures. Rotifers were not a problem in "open" cultures in Germany (Soeder, 1980) but caused difficulties in South Africa (Grobbelaar, 1981b). Serious infections were not expected in "closed" algal cultures, but rotifers and protozoa were commonly present. Chironomid larvae were also a pest in "closed" as well as "open" systems. Grobbelaar (1981b) showed that algae were digested during passage through the guts of chironomid larvae (Fig. 6.4). He also showed that invertebrates such as the protozoan *Stylonychia* can cause shifts in algal species by grazing upon one type of algae but not on another (Fig. 6.5).

Infections may be controlled by pesticides or pH changes (Payer et al., 1976, 1980). Grobbelaar (1981b) suggested that infections can be controlled in four ways: (1) by maintaining optimal conditions for algal growth; (2) by the use of chemical agents such as "Mercaptothion", though the best drug will have to be established by trial and error; (3) by physical methods for removing unwanted organisms, such as the use of 50  $\mu\text{m}$  screens; and (4) by shock treatments which do not affect algae, such as cooling to below 5°C during winter, acidification to a pH of 3.5 with an organic acid followed by neutralization after a few hours, or allowing the

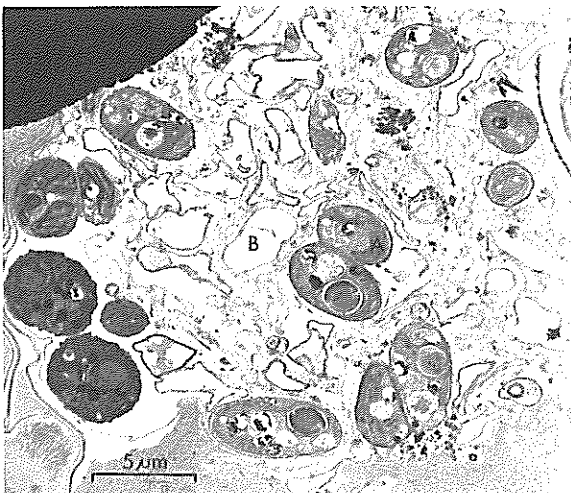


Fig. 6.4. Ingested algal cells in the gut of a *Chironomus* larva. Some cells (B) are being digested. (After Grobbelaar, 1981b.)

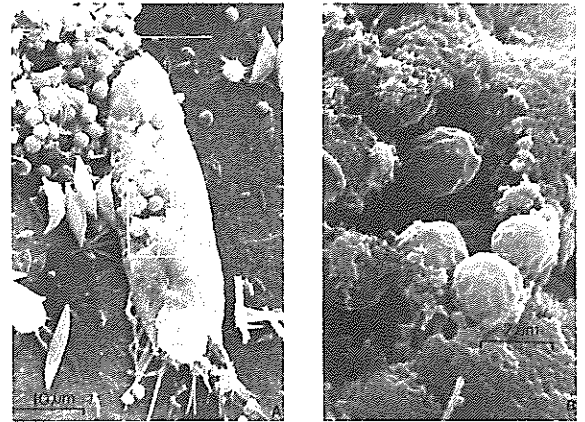


Fig. 6.5. Selective feeding of *Stylonychia* sp. on algae. *Scenedesmus* cells are present (A), but not ingested by the animal (B). Only *Chlorella* cells are ingested. (After Grobbelaar, 1981b.)

cultures to become anaerobic. Through these techniques, clean cultures may be maintained for months on end.

The invasion of "clean" cultures by foreign algae is also a problem since the maintenance in mass cultures of unispecific populations, except under extreme conditions such as salinity (e.g. *Dunaliella*), has largely been a failure (Goldman, 1980). Precise understanding of the mechanisms controlling population shifts is still lacking, but factors such as temperature (Goldman, 1980) and predation (Grobbelaar, 1981b) are important.

### Geographical location

The geographical location partly determines the light and temperature regimes which will be experienced (Robinson and Toerien, 1982). In general, semiarid and/or arid regions with high solar radiation and hot climates are well suited to algal mass cultivation, though evaporation rates may be high in these areas. "Closed" cultures reduce evaporation to almost nil whereas open cultures may lose as much as 9.8 mm day<sup>-1</sup>.

### MODELLING PRODUCTIVITY

Goldman (1979b) proposed a model based on equations developed by Van Oorschot (1955), Ryther (1959) and Shelef et al. (1968) to predict gross yields in algal mass cultures. The model

predicts an upper limit of about  $60 \text{ g m}^{-2} \text{ day}^{-1}$  for algal mass cultures. This model does not take processes such as respiration, photorespiration and photoinhibition, or temperature influences, into account.

Grobbelaar (1981b, 1982) developed a deterministic model in which a number of factors, such as efficiency of light utilization, respiration, excretion, and interaction between temperature and light were reflected. His model was verified during experiments lasting over 56 h. He stressed that respiration or other loss factors could be much more important than generally believed.

The large number of assumptions (Robinson and Toerien, 1982) in the model of Goldman (1979b), and the fact that factors such as temperature are ignored in the model, suggest that the approach followed by Grobbelaar (1981b, 1982) should be developed further.

#### PRODUCT QUALITY

When nutrients are provided in excess and light is the growth-limiting factor, most algal species display a remarkable consistency in their chemical composition: 50% protein, 30% carbohydrates and 15% lipids (Myers, 1957; Goldman, 1980). However, in some algae and under other limitations, the composition of algal cells can vary much more (Spoehr and Milner, 1949; Miyachi et al., 1977; Dubinsky et al., 1978; Shifrin and Chrisholm, 1980; Robinson and Toerien, 1982).

Crude protein in mass-cultured algae varies from 36 to 74% when nitrogen is not limiting (Robinson and Toerien, 1982). However, when nitrogen is growth-limiting, the crude protein content of "open" cultures may be as low as 5% (Mostert and Grobbelaar, 1981). The protein content of micro-algae can be manipulated by varying the nitrogen concentration in the feed water (Mostert and Grobbelaar, 1981). Mostert (1982) found an inverse relationship between protein and carbohydrate content of micro-algae when she manipulated the nitrogen input into mass cultures. Climate and temperature (Soeder, 1980), light intensity and quality (Miyachi et al., 1977), and potassium concentration (Van Vuren and Grobbelaar, 1982) also influence the protein content of micro-algae. Algal cells produced in "closed" mass

cultures did not differ materially in composition from those produced in "open" cultures (Geldenhuis, 1983).

Micro-algal biomass is usually low in sulphur containing aminoacids (Soeder, 1979), but this problem might be overcome by managing culture conditions, strain selection or genetic manipulation (Soeder, 1981). The fact that a high-methionine strain of *Chlorella* was isolated (Cacco and Ferraro, 1975) is a positive indication in this direction (Soeder, 1981).

Cell walls contain complex polysaccharides (Pabst, 1978) and represent about 10% of micro-algal biomass (Soeder, 1979). These carbohydrates could be cellulose, mannans and xylans (Soeder, 1979; Aaronson et al., 1980). Starch and other carbohydrates act as storage products in micro-algae (Soeder, 1979; Aaronson et al., 1980), but the low carbohydrate digestibility of algal biomass may be due to both physical and chemical barriers to digestion (Moraine et al., 1979; Soeder, 1981).

The lipid content of micro-algal biomass is usually not high (<15%), but in some cases can be as high as 53% of dry weight, and indicates a potential for production of lipids and oils from micro-algae (Aaronson et al., 1980). Some long chain fatty acids can be present in algae (Soeder, 1979, 1981). The lipid content (14.6% average) of micro-algae cultured in closed ponds (Geldenhuis, 1983) was very similar to that of "normal" micro-algae cultured in open ponds. Pigments such as chlorophylls and carotenoids constitute 3 to 5% of micro-algal biomass (Soeder, 1981). These pigments could cause colouring problems in animal diets (Walz et al., 1975). At low dosages in fish feed, micro-algae improve the colour of the fish, but high levels can lead to overpigmentation (Soeder, 1981). Micro-algae grown in "closed" ponds usually had chlorophyll *a* concentrations between 1.0 and 3.0%, values which differ from those grown in "open" cultures (Geldenhuis, 1983).

Micro-algae contain appreciable quantities of nucleic acids which could pose health hazards to humans (Kofranyi, 1978; Soeder, 1979). Nor do the nitrogen contents of micro-algae grown in "closed" algal ponds appear to differ appreciably from those grown in "open" ponds (Geldenhuis, 1983).



### NUTRITIONAL ASPECTS

Systematic studies in Germany indicated that the nutritive value of green micro-algae depends largely on processing technology (Kraut et al., 1966) and that *Scenedesmus*, as an example, is an excellent protein source for humans (Kofranyi and Jekat, 1967; Muller-Wecker and Kofranyi, 1973). Similarly *Spirulina* was shown to be an excellent source of protein in Mexico (Durand-Chastel, 1980).

Certain *Chlorella*, *Scenedesmus* and *Spirulina* species are free from specific toxins (Soeder, 1981), and long-term feeding studies suggest that algae produced in "clean" cultures are toxicologically safe (Pabst et al., 1978). However, humans cannot use unlimited quantities of algae because of a build-up of uric acid with concomitant adverse health effects (Becker, 1978). A further problem is the accumulation of environmental pollutants such as toxic minerals and polycyclic hydrocarbons in or on mass-cultured algal material. These pollutants originate from fertilizers, air or water pollution (Soeder, 1980). Enclosing cultures under plastic prevents pollution with dust and probably other environmental pollutants.

### MANAGEMENT OF AUTOTROPHIC SYSTEMS

Technology for mass cultivation of algae may be extremely unsophisticated, like a simple ditch in India where a dense growth of *Spirulina* grows on fermented cattle manure, is stirred occasionally with a broom handle, harvested by cloth filtration and dried in the sun (Venkataraman, 1978; Venkataraman et al., 1980). On the other hand, in Japanese *Chlorella* factories, mass culture vessels with automatic controls are seeded from sterile stocks, cells are harvested by centrifugation, and the product is washed and finally freeze-dried (Kawaguchi, 1980). The technologies employed range from a state close to subsistence agriculture to elaborate biotechnology (Soeder, 1980).

The prime operational purpose of any mass culture is to optimize the rate of production rates of the alga as biomass or as a source of fine chemicals. This presupposes a predictive capability about such mass cultures.

Quantified predictive capabilities (mathematical

models) have been developed for certain aspects (Goldman, 1979b, 1980; Grobbelaar, 1981b, 1982), but are lacking for others. For instance, it seems to be extremely important that the correct areal densities of micro-algae are used (Grobbelaar, 1982; Vonshak et al., 1982). Grobbelaar (1982) postulated through his mathematical model that the optimal areal density for maximum productivity is from 16 to 22 g C m<sup>-2</sup>. Below this level, productivity is extremely sensitive to small changes in the areal density, whereas at higher values the same is not true. To deal with the occasional adverse conditions that are encountered in any mass-culture situation, it would be better to operate at or just above the optimal areal density, rather than just below it. Mathematical models are thus extremely helpful in delineating operational strategies and optimizing the design of systems. Unfortunately no mathematical model can predict the invasion of a culture by other organisms. Consequently frustration may result if a specific alga is to be mass-cultured, unless it has some extreme tolerance to one or more environmental variables. In such cases, trial-and-error solutions based also on the experiences of other workers will have to be developed.

Management of autotrophic micro-algal mass cultures is not yet a fully developed science. However, we hope that this account has highlighted, not only the advances which have been made, but also the lack of knowledge in many important areas. The high productivity and other desirable characteristics of micro-algae still make them a fascinating subject for research and a potential source of proteins or other chemicals to man.

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