THE ROLE OF ALGAE AND PLANKTON IN MEDICINE

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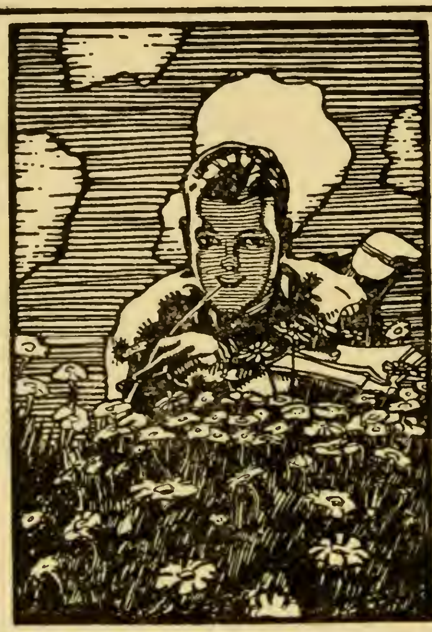

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To

GERTRUDE

For her patience and understanding

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—*The Authors*

FOREWORD

THIS MONOGRAPH on the role of algae and plankton in medicine is a timely one. It is, so far as the present writer knows, the only medical survey of the field, and it appears at a time when specialists in various non-medical branches of biology are exhibiting a keen interest in the subject. The literature is expanding so rapidly that it may soon be difficult to keep abreast of the advances.

Although algae and plankton have been empirically utilized as food since ancient times, scientific observations were at first limited mainly to academic botanical aspects In recent years, world food shortages have focused attention on the nutritional utility of these substances for man and the animals upon which he is dependent. An important corollary—the relationship of these forms of life to clinical medicine—appears to have been neglected until the present publication.

It should be stressed at the outset that the authors have been content merely to point out the great medical potential of algae and plankton, rather than to promote panaceas. Nor have they sought, with their historical references, to justify various dietary regimens and practices of primitive medicine. The fact that additional investigations might rationally explain ancient or aboriginal medical practices is presented merely as a matter of interest rather than importance.

However, in the opinion of this writer, no apology would be necessary even if the authors had utilized the historical approach and examined modern medicine in the light of primitive conceptions. Even Hippocrates, in his "On Ancient Medicine," stated that the greatest medical discovery up to his time (400 B.C.) had involved countless experiments ranging from field to kitchen and finally resulted in the baking of bread. Perhaps the discovery of sugar was almost as complex. The historical approach would also have revealed that cinchona for malaria, ipecac for amoebic dysentery, chaulmoogra for leprosy, iron filings for anemia, and digitalis for the heart all represent refinements of the dross of folk medicine and likewise require no apology. Pertinent examples can be cited from medicine of the present century. About 25 years ago, Indian physicians brought reports about the action of a drug and specimens of the preparation to the writer for evaluation and criticism. Preoccupation with seemingly more pressing problems led to filing the reports and discarding the specimens. Had time permitted an examination, Rauwolfia might have entered medicine at that time.

The authors are less concerned with past neglect than they are with emphasizing possible immediate medical applications in a promising field. Much important data has been presented on nutritional, physiological, toxicological, and public health problems. Also, some rather interesting clinical hypotheses have been propounded, particularly in the neurologic realm. Since a vast majority of the references included in their excellent bibliography are derived from non-medical sources, the implication of neglect by physicians does not seem unwarranted.

Although countless other subjects occur to the writer (and, it is hoped, to the reader), an unwritten rule must be obeyed: the introduction should not be longer than the actual text. In summary, the authors have surveyed the world literature on algae and plankton in relation to medicine. They have appended an excellent bibliography of widely scattered, but generally available, references, most of which will not be found in the usual medical index. They have accented the recent literature dealing with promising fields of investigation and possible application to current medicine. There has been made available a concise introduction to a subject as old as recorded medical history, yet so new that many medical investigators do not realize its existence.

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I. INTRODUCTION

DURING THE PAST SEVERAL DECADES, there have appeared increasingly insistent reports on algae and plankton—reports concerned chiefly with developing a new source of food for a world population rapidly outgrowing its arable land areas. These articles, appearing in technical journals[1-18] and in the public press,[19-26] have been authored primarily by botanists, nutritionists, economic philosophers, and lay scientific writers.

The concern of these individuals seems amply justified, not merely by future census potentialities, but also by actual present shortages. These include the frequent caloric famines affecting many nations, as well as the qualitative nutritional deficiencies. The latter are pointedly exemplified by the dietary fat shortages in Europe during World War II, the notorious hypovitaminoses of many Asians, and the recently highlighted protein deficiency, kwashiorkor,[27,28] of the Africans.

With the growing recognition of the importance of nutritive and biochemical substances in the prevention and therapeusis of disease, it is remarkable how singularly little attention has been paid by the medical press to algae and plankton. As Tilden[29] has so aptly put it: "Here in America only a small group of college professors of botany are familiar with marine plants. Phycology has been a science far removed from all other subjects. It is too difficult for the biochemist, the zoologist, or the physicist to attempt to work with forms of which he knows nothing. *As for physicians, the depth of their ignorance in the subject is abysmal.*"

The present monograph attempts to assemble in tangible form such knowledge of algae and plankton as may be pertinent to medicine—knowledge which presently is diffused in a wide variety of non-medical publications. Such a compilation of background data, of current concepts, and of future potential might conceivably serve as a stimulus for wider activity in this field.

II. DEFINITIONS AND CLASSIFICATIONS

A. ALGAE

ALGAE ARE THE SIMPLEST FORM of plant life.[30] The term "alga,-ae" derives directly from the Latin word meaning "seaweed,"[31] but as used today encompasses many additional forms. While the vast majority of algae are characteristically aquatic, some few species have become adapted to growth in soil and even in such exotic habitats as floating icebergs. Some are unicellular, others multicellular, with great variations in cellular structure. They may vary in length from less than one micron for the slimy growths, to hundreds of feet for the giant kelps.[32]

Unlike the higher plants, algae possess no true roots, stems, leaves, or seeds. The basic metabolic processes of nutrition and reproduction are all localized in the individual cell, without the transport system of land plants. The one feature common to all algae is the property of photosynthesis—the utilization of light energy to convert inorganic to organic substances. By this method, algae can convert carbon dioxide, water, nitrogen, and mineral substances into diverse types of proteins, carbohydrates, lipids and sterols, vitamins, and other metabolic complexes.[33,34]

Algae have been extensively studied during the past sixty years, and over 17,000 species have been described.[35,36] Their classification has been varied, unstandardized, and in many instances rather badly confused. For many years, they were simply lumped into four principal groups based on gross coloration: green, brown, red, blue-green.[37] With increased understanding of pigments, of metabolic reactions, and of reproductive mechanisms, algae are today classified into seven main Divisions, each subdivided into Classes, Orders, Families, Genera, and Species[38-41]:

I. CHLOROPHYTA: grass green; unicellular, multicellular, a few macroscopic; cell wall of cellulose and pectins, mostly freshwater, some marine.

II. EUGLENOPHYTA: grass green; unicellular; motile; lacking a cell wall; largely freshwater.

III. CHRYSOPHYTA: yellow green to golden brown (xanthophylls and carotenes may mask chlorophylls); microscopic, mostly unicellular; cell wall containing silica (e.g. diatoms); largely freshwater, but many marine forms.

IV. PYRROPHYTA: yellow green to dark brown, xanthophylls predominant; unicellular, motile; cellulose cell wall; many marine forms.

V. PHAEOPHYTA: olive green to dark brown; mostly macroscopic; cellulose and pectin cell wall; chiefly marine.

VI. CYANOPHYTA: blue green, phycocyanin; multicellular, but usually microscopic; usually with gelatinous sheath; fresh water, marine, some terrestrial.

VII. RHODOPHYTA: red, phycoerythrin; usually macroscopic; pectin and cellulose cell wall; mostly marine.

These seven Divisions include over a dozen Classes, about sixty Orders, hundreds of Families, and thousands of Genera. However, only a few score Genera are of sufficient historical or current medical importance to merit individual attention. Among these are the following:

CHLOROPHYTA: *Chlorella, Scenedesmus, Ulva, Nitella, Chlamydomonas, Polytomella, Prototheca, Stichococcus, Cladophora, Oedogonium*

EUGLENOPHYTA: *Euglena*

CHRYSOPHYTA: *Nitzschia* (diatoms)

PYRROPHYTA: *Gymnodinium, Prorocentrum, Gonyaulax*

PHAEOPHYTA: *Macrocystis, Nereocystis, Laminaria, Fucus*

CYANOPHYTA: *Chroococcus, Microcystis*

RHODOPHYTA: *Porphyra, Gelidium, Corralina, Chondrus, Rhodymenia, Gigartina*

B. *PLANKTON*

The interrelationships of plankton and algae can be simply illustrated by the following schematic diagram:

```
      PLANKTON                               ALGAE
     /        \                             /     \
Zooplankton   Phytoplankton = Microscopic Algae   Macroscopic Algae
                                                     (Seaweed)
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"Plankton," directly from the Greek, is defined by Webster[31] as "the passively floating or weakly swimming animal and plant life of a body of water." The term appears to have been first used by Hensen in Germany about 1889, although Johannes Mueller collected microscopic organisms as early as 1845 by dragging a very fine net through the sea.[30]

As used today, the term "plankton" generally encompasses the countless billions of minute animals and plants (excepting bacteria) which reside in water. This includes the algal growths seen floating in fresh water ponds or lakes, and also the minute organisms in the sea. The latter provide, directly or indirectly, the basic food for all forms of aquatic life, from the smallest polyps to the largest whales, with each devouring some form smaller or weaker than itself. Whipple[42] describes it thus: "Fish feed upon crustacea and insect larvae, the crustacea prey upon the rotifera and the protozoa, the rotifera and protozoa consume algae and bacteria, and finally algae nourish themselves by the absorption of soluble substances and gases provided in part by the decomposition of animal and vegetable matter brought about by bacteria." This biological sequence is perhaps more graphically characterized in the old Chinese proverb[43]:

Big fish eat little fish
Little fish eat shrimp
Shrimp eat mud.

And the "mud" comprises the multitudinous microscopic organisms at the base of this amazing nutritional pyramid!

It is apparent, then, that plankton exists in two basic forms, *zooplankton* (animal) and *phytoplankton* (plant)[44]:

1. *Zooplankton,* consisting of shrimplike crustaceans; floating fish eggs and larvae; and larvae of crabs, molluscs, and a number of other small animals. Most important, in volume and edibility, are the copepods and krills.
2. *Phytoplankton,* consisting almost exclusively of microscopic algae. Main types are the diatoms and unicellular flagellates.

The term "ultraplankton" refers generally to the most minute forms (nannoplankton) of marine plankton, as differentiated from the bulk of phytoplanktonic organisms too large to be eaten by young larvae. Cole[45] suggests substituting the term "hekistoplankton" for greater accuracy; he defines it as including all those elements in plankton ten micra or less in diameter of cell body. Other descriptive terms sometimes used are "littoral" and "benthal," indicating growths along the shore or in deep waters; and "limnetic," designating growths occuring in moving water.

Of great practical importance is the fact that the quantities and composition of plankton vary tremendously with geographical area, season, temperature, light, water depth and currents, and nutrient concentrations.

III. MACROSCOPIC ALGAE (SEAWEEDS)

A. NUTRITIONAL ASPECTS

THE USE OF MACROSCOPIC ALGAE (seaweeds and occasional fresh water forms) as food extends far back into antiquity, most notably in the Oriental community.[46] One of the earliest records of algae is found in the Chinese Book of Poetry, *circa* 800 B.C., wherein "pondweed" and "duckweed" appear as edible delicacies worthy even of sacrifice to revered ancestors. References to the emblematic beauty of algae are made in the Book of History and in the Analects of Confucius.[43]

In the Far East, seaweed generically goes by various names[47]: in China, it is called *hai t'sai;* in Japan, *nori;* in Guam, *lumut;* in Tahiti, *rimu;* and in Hawaii, *limu.* Hawaii has the largest variety of edible seaweeds in the world—75 types—but these are not all of choice quality. The considerable regard the Japanese manifest for algal foods has been related to the fact that, apart from rice, they have very few vegetables. Yet, in spite of the numerous types of seaweed available, it is striking that seaweeds provide less than one per cent of the total vegetable food of the Nipponese.

Although scores of different seaweeds have been eaten in various parts of the world, only a few have enjoyed truly widespread consumption.[48,49] Probably the most cosmopolitan seaweed, in geographical distribution and usage, has been the membranaceous *Porphyra,* commonly known as purple laver. The Chinese used it mainly as a base for soups. Called *Amanori* in Japan, it was at one time considered one of the most important food plants in that land, and was consumed chiefly as a salad. It grew both naturally and under cultivation in surrounding waters; in fact, the most famous grounds for cultivation of *Porphyra tenera* were in Tokyo Bay itself. The purple laver was also well known in the British Isles and in France; it went by such diversified names as *sloke, slouk, slack, sloucawn,* and *marine sauce.* Its preparation was also more varied here. The Europeans not only used it as a salad, but they also fried it in fat, or boiled it into a dark brown semi-fluid breakfast dish, or even baked it into a laver bread. The British sometimes sealed the fresh plant hermetically for use as a food by the crews of whaling vessels. In Alaska, the use of *Porphyra* as "sea lettuce" was common with the Indians. On the Pacific coast, from Canada to Mexico, *Porphyra perforata* was the only local seaweed in common use.

Green laver, or *Ulva lactuca,* was known to the Orientals as *Wakone,* but was chiefly eaten in Scotland, either as a salad or as a soup. The kelp *Laminaria japonica* was utilized primarily as a soup ingredient by the Japanese (who called it *Kombu*) and by the Alaskan Indians. Dulse (*Rhodymenia palmata*), also variously designated as *water leaf, sea kale, dillisk, sol,* and *crannogh,* was one of the favorite seaweed nutrients in Scotland, Ireland, and the northeastern United States in the nineteenth century. It was chewed fresh or after drying, or used as a flavoring, salad, or relish.

Outstanding among seaweeds finding a place in the cuisine were those of the Gelidiaceae family. *Gracilaria lichenoides, Gelidium corneum,* and other closely related species were the prime source of agar, familiarly referred to as *seaweed isenglass* and *kan ten* ("cold weather"). The latter, a pearly white, shiny, transparent, tasteless and odorless extract, was introduced by the Chinese to Japan in 1662 A.D. It was consumed mainly as a soup stock, summer jelly, dessert, or candy. *Chondrus crispus* (Rhodophyta) became highly popular in the production of jellies. This alga was the source of Irish moss, also designated as *carragheen, Dorset weed,* *pearl moss, jelly moss, rock moss, gristle moss,* and *Tsunomata* (among the Japanese). This was the seaweed so popular in Ireland and in the United States in the making of *blanc mange. Chondrus crispus* and *Laminaria saccharina* were at one time combined on the coast of Armorica to make a jelly-like "pain des algues" or seaweed bread.

The general utilization of seaweeds as food in Europe—particularly in Scotland, Ireland, and Scandinavia—diminished notably in the latter half of the nineteenth century. With the food shortages incident to World War II, there was some revival of interest in macroscopic algae. It has been reported that German occupation troops in Norway built two bakeries to make bread from dried algae which had been desalinated and ground.[47] German technicians are also credited with having fabricated from Norwegian seaweed an edible casing for sausages weighing one-twenty-ninth as much as cellophane.[7] Prior to the war, the English made a custard powder from brown seaweed; the New Zealanders were marketing a similar product after the war. In 1944, Yarham[47] told of a botanical banquet some years earlier in Wisconsin that featured a dehydrated algal menu: fried seaweed, puree of seaweed, roast seaweed, and devilled seaweed!

It is clear that the employment of macroscopic algae as food was on a purely empirical basis—some of it founded on local custom, some on superstition, some simply on the belly-filling qualities of seaweed and its freshwater relatives. As with many other traditional practices, modern science has tried to explain, *post facto,* why the ancients were actually right. That many macroscopic algae are edible and not poisonous is amply evidenced by the survival of both the custom and the customer. That these algae are also endowed with some desirable nutritional qualities is being gradually demonstrated. The exact composition has been determined for only a relatively few species. Even in these, marked variations are found from crop to crop and season to season.[50,51] The main components appear to be a variety of carbohydrates, some proteins and fats, mineral salts, vitamins, and water.

Carbohydrates are present in large amounts, chiefly as cell wall components and as intracellular storage matter.[40,52-54] Most of them are of a heterocyclic six-membered pyranose structure. Glucose and other reducing sugars are either absent, or present in only trace quantities. Free pentoses are likewise of no quantitative significance. Mannitol, a hexahydric alcohol derived by reduction from mannose or fructose, is found in amounts up to 30 per cent of dry weight in species of Phaeophyceae. Mannitan (as mannitol anhydride), dulcitol and sorbitol (mannitol isomers), and floridoside (a glycerol-*D*-galactoside) all occur in various species of Rhodophyceae. Trehalose, a glucopyranoside, is present only in fresh water species of Rhodophyceae. The chief intracellular reserve products in many species are glucose polymers. Thus floridean starch is characteristic of the Rhodophyceae; it is composed exclusively of glucose residues, but differs from ordinary starch in containing a large proportion of 1:3 linkages and in being able to resist the effect of β-amylase. The Phaeophyceae lack starch and allied substances. Laminarin, another glucose polymer found in some Phaeophyceae, comprises up to 25 per cent of the plant on a dry weight basis.

Cellulose is the main cell wall component in the Phaeophyceae and Rhodophyceae, and it makes up from 2 to 15 per cent of their dry weight. By chemical and radiological examination, it appears to be similar to the cellulose of higher plants. Other related carbohydrate substances in algae include chitin, pectin, and hemicellulose. Algin, a characteristic intracellular substance of the Phaeophyceae, forms about 25 per cent of the seaweed on a dried basis. It is a calcium-magnesium salt of alginic acid, a polymer of β-*D*-mannuronic acid. Xylan, a polysaccharide from which xylose is derived by hydrolysis, has been found in *Rhodymenia palmata*. Other intracellular polysaccharides, composed of residues esterified by sulfuric acid, include agar-agar, carragheenin, and fucoidin.

Relatively little is known about the protein components of macroscopic algae, except that they are present in only small quantity and that they are minimally assimilable by human beings. According to Cameron,[55] analyses indicated 6.85 per cent protein in *Gelidium corneum*, and 5.49 to 5.82 per cent in various species of *Laminaria*. Analyses of various kelp meals and feeds for animals (who may utilize contained proteids better than humans) indicated levels of 7.04, 13.12, and 5.60 per cent, respectively. At the 1952 International Seaweed Symposium,[56] the Youngs indicated that the proteins and peptides were difficult to extract from algae, with only 50 per cent being obtained even by strong alkali. The nitrogen of various common brown seaweeds consisted of volatile base nitrogen, nitrate nitrogen, and residual Kjeldahl nitrogen. E. T. Young also stated that the α-amino-nitrogen after hydrolysis was always lower than the residual Kjeldahl nitrogen because of amino-acid decomposition. The amino-acids found on acid hydrolysis of *Laminaria saccharina* and *Alaria nodosum* included glycine, alanine, valine, leucine, aspartic acid, serine, glutamic acid, threonine, proline, phenylalanine, lysine, and arginine. Some algae have also been shown to contain nucleic acids, and some various water-soluble peptides.[57-60]

The lipid content of macroscopic algae may vary from zero to 3 or 4 per cent. Of the fatty acids identified, the commonest are the unsaturated ones with 16, 18, and 20 carbons. These are present in most algae, with the exception of *Alaria crassifolia,* which has over half its fatty acids in saturated forms. The predominant algal fatty acid is the 16-carbon palmitic acid. Besides these fats, there are varying small quantities of such lipoid substances as photosynthetic pigments, sterols, and hydrocarbons.[61-63]

The mineral ash content varied from 5.0 to 38.9 per cent in a number of reported studies. The water-soluble ash exceeded the insoluble fraction by as much as four or five times. One investigation of kelps[64] listed identified substances and their percentage of the total ash as shown in Table I.

TABLE I

Sulfates	6.14—11.24%	Potassium	9.40—10.43%
Calcium oxide	3.35— 4.46	Sodium	10.51—13.57
Magnesium oxide	1.99— 4.34	Chlorine	27.62—32.59
Phosphates (as P_2O_5)	2.52— 5.60	Iodine	0.06— 0.09
		Bromine	Trace

The seaweeds containing the most iodine are *Laminaria digitata, Laminaria saccharina, Fucus vesiculosus,* and *Saccharina bulbosa.* The richest in bromine is *Fucus serratus.*

Vitamins in good quantity have been found in some varieties of seaweed investigated.[47] Vitamin A has been demonstrated in the sea lettuce *Ulva lactuca, Laminaria digitata,* and *Codium tomentosum,* as well as in ground meal prepared from Pacific coast kelps. This vitamin is even more abundant in phytoplankton, which will be discussed later.

Freudenthal[65] in 1949 conducted biological investigations on three species of algae from the coast of Denmark—*Furcellaria fastigiata, Fucus serratus,* and *Fucus vesiculosus.* He reported that they possessed a growth-promoting and anti-xerophthalmic vitamin activity (in Wistar rats) exceeding that of the best Danish summer butter. He also claimed that feeding these algae prevented the epithelial dysplasia and rickets that developed in animals maintained on vitamin-deficient diets. Freudenthal, incidentally, takes issue with the common belief that rickets is related to Vitamin D deficiency; he feels that the disease is "the retarded, inhibited growth dynamics of cartilage and bone caused by a suboptimal intake of Vitamin A."

The American brown kelps, Phaeophyta, studied extensively for their vitamin content, were found rich in thiamin, niacin, and riboflavin. Norris and others[66] found a thiamin concentration comparable to that of many fruits and vegetables in *Alaria valida,* various *Laminaria* species, *Porphyra nereocystis, Rhodymenia pertusa, Ulva lactuca,* and especially *Porphyra perforata.* The Vitamin B values of these algae were determined in rats first fed on deficient basal diets for about two weeks, when weight loss and cessation of growth took place. There appeared to be no loss in Vitamin B activity from drying the algae, nor was there any noted relationship to the depth of the water from which the algae were harvested.

Limu eleele and *Limu lipoa,* edible Hawaiian seaweeds, were found by Miller[67] to be poor sources of both Vitamins B and C. Only small amounts of Vitamin C were found in dulse (*Rhodymenia palmata*). However, concentrations comparable to those in lemons have been reported in both purple and green laver. Generally the spring and autumn harvests of seaweed are highest in ascorbic acid content. Unlike the case with Vitamin B, seaweed collected from littoral zones is richer in Vitamin C than that obtained from deeper waters. Although ascorbic acid is usually destroyed by the drying preparatory to shipment of seaweed to market, coastal populations still derive significant quantities of the vitamin from eating the fresh algae.[68,69]

The occurrence in seaweed of the Vitamin D precursor, ergosterol, has also been recognized.[70] Sauvageau reported marked anti-rachitic benefits in 25 human subjects fed various macroscopic algae.[71] In 1952, Ericson[56] reported the presence of Vitamin B_{12}, folic acid, and folinic acid in various brown and red seaweeds from the Baltic and North Seas. The B_{12} content of some of these was comparable to that in liver. The presence of a-tocopherol in seaweed was noted by Brown.[72]

Norris[66] would appear to have been correct when he wrote in 1937: "Seaweeds contain no starch, and the complex carbohydrates which are present are not readily hydrolyzed by the human digestive enzymes. The value of [macroscopic] marine algae as food is due mainly to the inorganic salts present and the vitamins." It is possible, however, that broader chemical studies in the future may disclose more assimilable substances; and it is also conceivable that materials which *in vitro* presently appear inutile may be shown to be otherwise *in vivo.* At any rate, the nutritional compositions and values of the macroscopic algae are in striking contrast with those of certain microscopic algae to be discussed in another section.

While the direct nutritional utility in humans has been limited, the readily available hundreds of thousands of tons of seaweed might well be more effectively exploited in agriculture and industry. Seaweed has been used as a fertilizer in the coastal portions of southeast Asia for many centuries. Even in the fourth century, Pallidus mentioned the use of seaweeds in western lands as a partial substitute for manure.[73] To this day, particularly in Brittany, Normandy, and the Channel islands, algae are used in soil conservation. Marrett[74] has attributed the small size and small bones of Jersey cows to a calcium deficiency associated with the high iodine content of the grass, a state resulting from the application of seaweed fertilizer to the ground. The iodine content of algae is highest in the fall and lowest in the spring, with marine specimens containing much more than freshwater ones. According to Lunde and Closs,[75] iodine—whether derived from seaweed fertilizer or meal—causes an increase in the iodine content of the cow's milk, but this changes neither the quantity nor the quality of the milk. That the cattle on the Isle of Jersey are free from bovine tuberculosis, hoof and mouth disease, and brucellosis is attributed by some to the use of algal fertilizers; it seems more reasonable, however, to explain it on the simpler basis of relative isolation from contagion.

On the Pacific coast of the United States, *Desmarestia,* a brown seaweed, is believed to be poisonous to cattle, possibly because of the free sulfuric acid present in the plant.[76] This has doubtless acted as a deterrent to the wider employment of other algae in that area.

Many forms of algae have been used as dietary supplements for livestock. Caesar, in his "Commentaries," tells of saving a crucial battle by feeding dried seaweed to his hungry horses when the supply of grain became exhausted. A series of reports from 1745 to 1884 indicates that livestock in the northern British Isles and in the North Sea areas were encouraged to feed on algae washed up on the shore; in addition, the natives collected and mixed the seaweed with conventional fodder.[65] Large aquatic plants reportedly have also been used as forage for cattle in Yugoslavia for many years.[4] It was not until World War I, however, when the French were confronted with a shortage of oats and other grains, that any strong interest was manifested in the provender value of seaweed. The military service then began studies on the nutrient effect of seaweeds on horses and cows.

Sauvageau and Moreau[77] reported that *Fucus serratus* and *Laminaria flexicaulis* formed excellent animal aliment. Adrian[78] found in animal experiments that seaweed caused an increase in weight proportional to the weight of the material ingested; seaweed was also credited with stimulating the assimilation of conventional nutrients. In other studies at the Sorbonne, cats and chickens were well maintained on seaweed feedings. Gloess[79] found that macroscopic algae, when cleared of their potassium, bromide, and iodide salts, made a satisfactory replacement for oats in horses' diets; they could also be used as a supplement to standard rations for swine and poultry. Beckman[80] successfully maintained dogs and hens on a baked loaf containing a mixture of ground seaweed, rye, and potato flour. Irish moss was also used to feed cows, calves, and pigs. Freudenthal[65] in 1920 reported the use of seaweed as cattle fodder in Iceland. During World War II, a dried seaweed meal was prepared in a factory in County Clare, Ireland, for use as a stock feed.[81] Following a series of studies with pigs, it was concluded that the main value of the meal lay in improving the total amount of digestible ration. Use of seaweed for fodder has also increased in Sweden as a result of scarcities occurring during the Second World War.

To guard against adverse effects from seaweeds used in their entirety, it has been found advantageous to rinse the algae first in fresh water to leach out excess salts.[47] In some of the feeding experiments reported, the animals required from several days to a week or more to accommodate their dietary habits to a predominantly seaweed ration. Undernourished animals often responded better to algal diets than did well nourished beasts. From this it might be surmised that seaweed supplements had overcome some existing deficiencies.

Pälsson and Grimsson[82] have described in seaweed-fed lambs a syndrome demonstrated to be associated with a generalized spotty neural demyelinization. The clinical picture resembles that in the previously reported "swayback," a disease characterized by muscular weakness, incoordination, paralysis, and death.[83] Pälsson and Grimsson state that supplementing the diet of the ewes with copper during the latter part of pregnancy markedly decreased the incidence, morbidity, and mortality of this malady. They do not make clear why supplemental copper should be either necessary or helpful, since the offending seaweeds showed a relatively high copper content. It is possible that something in the seaweed (or some contaminant on it) may have interfered with the proper utilization of copper, or perhaps markedly increased the need for it. In an interesting comparable situation, Dunlop in western Scotland administered single doses of copper sulfate to lactating cows fed on seaweed and obtained increased butterfat production within a few days.[84]

B. MEDICINAL USES

The medicinal purposes to which algae have been put are indeed diverse.[43,47,71,85-94] Their origins are often clouded in misty legend and hearsay, their rationale generally empirical. The Chinese credit the therapeutic qualities of algae to Shen Nung, the mythical First Farmer or Father of Husbandry and Medicine, who supposedly lived about 3000 B.C. The Materia Medica of the Chinese in the eighth century A.D. described one alga thus: "Whole plant is officinal. Taste bitter and salt. Nature cold. Nonpoisonous . . . the *hai tsao* grows in Tung Hai (Shantung) in ponds and marshes. It is gathered on the seventh day of the seventh month and dried in the sun." Of the seaweed *Porphyra coccinea,* the Chinese Materia Medica said: "This algal plant is a sort of laver which is green in the fresh state and purple when dry. It grows on the seashore . . . and the Fukienese . . . press it into cakes. It is not poisonous, but when taken in excess it produces colicky pains, flatulence, and eructations. It is recommended in diseases of the throat, especially goiter."

A related laver of the *Porphyra* grouping was the source of a preparation called *k'unpu,* utilized for the relief of dropsy. A similarly named viscous solution, *kwanpu* or *hai tai,* derived from a *Laminaria,* was used by the Chinese for menstrual difficulties. They also used a seaweed concoction known as *lung-she-tsai* for abscesses and cancer.

For many years Chinese gourmets have used the nests of some passerine birds (such as the sea swift) to make an exotic soup with reported aphrodisiac powers, and millions of these nests have been imported from Borneo. Although they were once thought to be made of a gelatinous seaweed, *Gracilaria spinosa,* it is now known that the main constituent is dried avian salivary juice!

In upper India, an algal prepartion known as *gillur-ka-putta* served to treat bronchocoele. Sugar wrack, *Laminaria saccharina,* was here considered useful against goiter, while in the Himalayas it was more prosaically employed to treat syphilis. The ancient Polynesians used filamentous algae to make poultices for bruises, cuts, and inflammations. The Hawaiians made watery extracts of *Centroceros* and *Hypnea nidifica* for constipation and other gastro-intestinal disturbances. The primitive Maori tribesmen, living in close contact with their domestic animals, frequently acquired their scabies; to treat this, they used *Durvillea* ("rimuroa"). The latter was also fermented with an extract of the poisonous Tutu shrub to make a cathartic.

The Indians of Sitka, Alaska, devised an unique mechanical nostrum for earache. Utilizing the tube-like character of the stipe of the bull kelp, they placed the thin end in the victim's ear and the bulb on a hot wet stone, thereby allowing steam to enter the afflicted external auditory canal! South American Indians are reported to have valued *Sargassum baccifer* to cure goiter and renal disorders, just as many other primitive tribes considered bladderwrack helpful in inflammations, sprains, and rheumatism.

In the West, algae were exploited somewhat as in the Orient, but to a much lesser extent. The Latin poets Virgil and Horace (first century B.C.) held algae in very low esteem. Nevertheless the Romans must have attributed to certain algae the property of healing sores; indeed the name *Confervae* (Class Chlorophyceae) derives directly from the Latin word "confervere," meaning "to solder." In addition, Pliny recommended certain seaweeds for gout, which the Romans knew only too well.

Discorides, a Greek physician of the first century A.D., recognized absorbent properties in white or medicinal corraline, *Muscus corralinus*. The powder from this alga was highly valued. It was prescribed as an external absorbent for burns, skin abrasions, rashes, and scurvy; and as an internal absorbent for chronic gastritis, heartburn, and diarrheas resulting from hepatic and intestinal disorders.

The American Indians, according to Chase,[43] did not believe in the use of salt on food. It seems that the Iroquois, having occasionally feasted on the white intruder, learned that he tasted saltier than the red man. Since salt was "white man's style," they avoided using it, firm in the belief that salt would make their hair turn gray and their toes turn up before their time. However, by taking "sea lettuce" or laver each spring, they supplied directly from nature the salt needs of their bodies. The saline tastes of the Iroquois prior to Columbus' (or Leif Ericson's) visit are not recorded.

In cases of goiter and scrofula, ancient medical men were known to prescribe calcine sponges, watercress, and similar substances which condensed and fixed marine elements. The Chinese Pentsao recommended all the medicinal algae in goiter therapy. The fact that goiter is practically unknown amongst the maritime Chinese and Japanese is doubtless due to the abundance of iodine in their seaweed dietary supplements and in their fish foods. In striking contrast is the noted prevalence of goiter in Alpine inhabitants, who eat no marine algae, and whose agricultural soil has been leached of iodine by ancient glaciers.[95,96] In the Andean regions of South America, the natives indeed learned to chew "goiter sticks"—the stipes or stems of species of *Phyllogigan*—to counter the high incidence of thyroid enlargement. Likewise because of their iodine content, the therapeutic use of oarweeds and wracks was common. Most popular were *Laminaria digitata* and *saccharina* and *Fucus vesiculosus,* the latter being prepared as an infusion. Another favored way of administering iodine was in the form of kelp pills, ash, or charcoal. Charcoal kelp was used under the name "Aethiops vegetabilis" for goitrous and scrofulous swellings. Coudert, in Geneva, was among the first to utilize iodine by itself as a therapeutic agent. In time it was found more economical to derive iodine from the saltpeter beds of Chile, and soon thereafter the great kelp industries of northern and western Europe began their rapid decline.

One of the earliest therapeutic applications of algae by the Chinese and Japanese was that of the derivative agar in gastrointestinal disorders. In cases of fever attributed to stomach conditions, their monks recommended *Gelidium* boiled into a jelly and sprinkled with ginger and sugar. For relief from disorders caused by extreme heat, an agar milk paste flavored with sugar and vinegar was given. This preparation was particularly favored for children. *Gracilaria lichenoides* was also utilized as a demulcent in intestinal and bladder difficulties.

In more recent times, the U. S. Dispensatory recommended agar as a laxative, eaten in small pieces like cereal with sugar and cream, or else in chocolate-coated form. Its value as a laxative is due to its colloidal property of absorbing and holding water without being digested. This attribute makes it desirable as intestinal bulk or roughage.[97,98] Agar also decreases gastric emptying time. The jelly extract of Irish moss, like agar, was advised in general irritability of the alimentary tract. During the past century it was prescribed for gastric ulcer, as well as for diarrhea and dysentery.

On the eastern Asiastic coast, a red alga, *Digenea simplex,* was dried and sold by Oriental apothecaries; its extract, "helminol," has been employed in the management of ascariasis and oxyuriasis.[99] In China, a combination of algae, including an *Enteromorpha,* a *Chordaria,* and seven

Floridees, was favored for its vermifugal virtues; it is thus not too surprising that the Chinese credited practically all marine algae with such powers. In Europe, until the end of the eighteenth century, vermifuges prepared from the calcareous algae *Laminaria officinal* and *ruben* enjoyed high public approval. In 1775, the Greek physician Stephanopoli discovered the anthelmintic properties of the small red-tufted alga *Alsidium helminthocanton,* found at low tide along the rocky Corsican shores. Additional seaweeds of help in deworming include *Hypnea musciformis* (particularly in Turkey and Greece), two species of *Chondria,* the green fresh-water alga *Rhizoclonum vovulare,* and dulse. Employed by the Maoris of New Zealand was yet another vermifuge, prepared from the bull kelp *Durvillea.*

Irish moss, the red seaweed that flourished in Carragheen, was used —after cleansing, curing, and bleaching—as a demulcent for coughs. Its mucus-forming qualities made it popular in the nineteenth century for pulmonary disorders generally, and for "consumption" particularly. The purest *Carragheen* preparation, *Electum albissimum,* was used for this purpose. It was sometimes prepared as a liver oil emulsion. *Carragheen* was also used occasionally to disguise the taste of bitter drugs. A related extract of *Chondrus crispus* found usage in World War I in easing the throat irritations of soldiers who had been gassed. *Carragheen* has also been denatured with whiskey and used liberally for coughs in some Hibernian bars in New York.[47]

A distinctive French preparation, to serve in place of linseed meal poultices, was cotton-wool soaked in a *Carragheen* concoction, then dried. Other seaweeds considered useful in lung diseases and scrofula included *Gelidium cartilagineum* (in Japan), *Dictyopteris polypodiodes* and *Stilophora rhizoides* (in the Mediterranean area), and *Laminaria saccharina.* Irish moss also served as a demulcent in diarrhea and in irritations of the urinary tract. In the middle of the eighteenth century, Scottish peasants were said to chew dulse to induce sweating in states of pyrexia.

The bladderwrack *Fucus vesiculosus* also enjoyed a role in certain concoctions and "slimming teas" in the management of obesity.[100] Its therapeutic merit might be attributed to its iodine content and its effect upon the thyroid gland. Agar, by virtue of its bulk and relative inabsorbability, also served in the treatment of problems of overweight. In the surgical field, stipes or "stem pieces" of *Laminaria cloustoni,* known as *Stipites laminaria,* were utilized to widen fistulas and wound openings because of their ability to swell markedly following moistening. These stipes were also used in distending the uterine neck in labor.

Despite the many old traditional uses of algae in therapeusis, the early twentieth century German medical lists included only *Carragheen,* Agar-agar, *Laminaria,* and *Fucus.* The *direct* medicinal uses of macroscopic algae have clearly undergone a marked decline. Certain extracts of seaweed are, however, of current significance in medical and para-medical fields. The most important of these are agar, algin, and carragheenin. They are extracted from algae by either water or alkali. Though they are hydrophilic colloids, they have often been erroneously referred to as seaweed "gums." The term "phycocolloid" has been introduced as a more correct designation. Aside from the medical areas, these extracts are also of value in the drug, food, and cosmetic industries because of their properties as gelling, suspending, emulsifying, thickening, and body-producing agents.

Shabrin and Shapiro[101] in Russia have recently reported success in treating trichomonal vaginitis with a *Laminaria* derivative. Monceaux[102] has enthusiastically advocated the use of the cellular plasma of marine algae for a diversity of clinical conditions. He advises oral ingestion for systemic diseases and deficiencies, and local application for dermatological afflictions. Monceaux values seaweeds so highly because of the naturalistic premise that they contain many of the therapeutically efficacious minerals and trace elements present in sea water.

Since its utilization by Koch[48] in 1881 for the culture and isolation of pathogenic microorganisms, agar has become an important adjunct of every hospital and bacteriological research laboratory. This dried and bleached gelatinous extract of the red seaweed Gelidiaceae is produced primarily by the Japanese; the British derive some agar from *Gigartina.* It possesses the desirable property of absorbing water easily and swelling up to a considerable extent. Only when heated to boiling does it dissolve; upon cooling it reverts to a somewhat colorless translucent gel. Agar is good as a solid culture medium particularly because it is relatively inert, not digested by most bacteria. It has also been used as a sealer and coating for pills, as well as a vehicle for lactic acid administered in intestinal infections.

Algin is the common name for derivatives of alginic acid, such as the sodium, potassium, and ammonium salts and the propylene glycol ester.[103] The discovery of alginic acid and the production of algin therefrom is credited to Stanford in 1885.[47] Alginic acid is the hydrophilic colloidal polymer of anhydro-β-*D*-mannuronic acid. It is extracted from the brown algae, especially the giant kelp *Macrocystis pyrifera, Laminaria digitata* and *hyperborea.*[104] The action of therapeutic agents can be modified by combining them with algin, which is free of nitrogen and possesses a low allergenicity. It lends itself well to incorporation into medications to be injected, since alginic solutions are clear and colorless, soluble in hot or cold water, and retentive of their smoothly flowing nature throughout wide temperature ranges. By varying the algin concentration only slightly, it is possible to control not only the viscosity, but also the rate of absorption and assimilation of medication.[105] One example is the delayed-action algin-epinephrine mixture being used in allergic disorders.[106] Another is its use in retarding the absorption of penicillin and various mycin drugs.[107] In this connection, it is interesting to note that Suzuki[108] has been able to produce anaphylaxis in guinea pigs by intraperitoneal injections of saline extracts of seaweeds common in the Japanese diet. Ouer, on the other hand, has reported minimal human sensitivity on intradermal injection of propylene glycol alginate in 50 allergic and 50 non-allergic individuals.[109]

Differing results have also been reported on toxicity and other adverse reactions. Millis and Reed[110] noted that sodium alginate does not interfere with calcium absorption in healthy human adults. Gill and Duncan[111] likewise observed no electrolytic alterations, nor any significant gastrointestinal disturbances. Nilson and Wagner[112] fed sodium alginate and propylene glycol alginate to dogs, mice, and chicks; they considered these substances harmless and "nutritionally wholesome, but bulky." Chenoweth[113] reported a high degree of toxicity in cats. On intraperitoneal or intravenous injection of sodium alginate, he found large antemortem intracardiac clots, as well as frequent indications of injury to the brain, liver and kidney. He asserted, too, that sodium alignate thus given elevates the erythrocyte sedimentation rate just as do the various gums. Chenoweth also found much toxicity with the nitrate derivative, which also greatly prolongs blood clotting time. The dialdehyde derivative of alginic acid was somewhat less toxic. Solandt[114] reported sodium alginate toxic for rabbits, while Frantz found the same with rats.[115] Blaine[116] considered that alginates make a good absorbable surgical substance, capable of being used as a gel, foam, or gauze with a minimum of local tissue reaction. He also used laminarin as a surgical powder in place of talcum.[117] Some of these disagreements in reported results may be due to product impurities or to differences in test animals. Others may be caused by variation in the source of materials; thus, Solandt and Franz both used *Laminaria* as the origin of their alginates, whereas Ouer used *Microcystis.*

Because alginates are not subject to breakdown by gastric juices, they have been used successfully as a coating for medications designed to have their effective activity in the intestine. Alginates have also seen much application as hydroabsorbent agents in the management of constipation. Berger and his colleagues[118] considered the water-absorbing and retaining properties of sodium alginate superior to those of natural gums, methylcellulose, and carboxymethylcellulose. They also found the acid-binding properties of sodium and calcium alginate comparable to those of carboxymethylcellulose. Gill and Duncan[111] thought that alginates were less effective in this respect than the carboxyl resins. Mulinos[119] tested the sodium and calcium salts of alginic acid in 60 humans with colonic constipation. He found them effective, and thought that, even though the alginates swelled minimally in water or gastric juice, they increased their original bulk 25 to 35 times in the alkalinity of intestinal secretions. Ludwig and his associates[120] compared the alginates favorably with the carboxylic type of cation exchange resins in their effect on intestinal sodium and potassium. According to Feldman,[121] human subjects tolerated 45 gm. of alginic acid quite well. It acted as a mild laxative, and it increased the fecal sodium and potassium to 16-20 per cent of that seen with the common cation exchange resins.

Since 1937, when Sears introduced the "reversible hydrocolloid technic" in the preparation of dentures, the use of alginates in dentistry has increased notably.[122] With this method, the dentist is able to secure with one impression reproductions of multiple cavity preparations, to reproduce undercuts without rupture or distortion, and to save chair time for himself and the patient.[123] In a similar fashion, alginates are used for fixed bridges[124] and dental hemostasis,[125-128] as well as for various types of medical moulages.[129]

Alginates have also been put to the following medical uses:

1. As an endaural wound dressing.[130]
2. As a mold for applying skin grafts after exenteration of the orbit.[131]
3. As a hemostatic in brain and thoracic surgery,[132-134] especially to close the bronchial tree in pulmonary tuberculosis, and in extrapleural pneumothorax.
4. As a decontaminant and treatment in mustard gas poisoning and burns.[135]
5. As swabs in the bacteriological examination of eating utensils,

since alginate "wools" adsorb more bacteria on wiping than does cotton.[136]

6. As a replacement for tragacanth and other natural gums in the manufacture of lubricating jellies.[104]

These diverse applications of the alginates utilize only a fraction of the phycocolloid production of the United States. Most of it is serving the dairy industry. By conservative estimate, for example, over half the ice cream made is stabilized with algin to provide smoothness of texture and body. Carragheenin has found some utility as a stabilizer in bakery products.[48]

An antibiotic property has been attributed to some algal substances. The natives of some Pacific islands wrap their fish catches in fronds of certain local seaweeds to guard against rapid decomposition,[47] but no specific antibotic has yet been identified here. The antiviral activity of the algal polysaccharides sodium alginate, laminarin, and fucoidin has been tested in chick embryos.[107] While no toxic effect upon the embryos was noted, neither was there any upon the viruses. Pratt, Mautner, and their associates[137] in 1951 demonstrated an *in vitro* inhibition of the growth of *Staphylococcus aureus, E. coli,* and *P. pyocyaneus* by extracts of several species of marine algae collected in California during the spring and fall. The same investigators[138] in 1953 obtained *in vitro* inhibition of the growth of several Gram positive and Gram negative pathogens with ether extracts of the red alga *Rhodomela larix*. They assert that the effectiveness of these algal substances is not due to their iodine content, but may be on the basis of some brominated phenolic compound.

The anticoagulant properties of algal substances have also received some attention. Following the identification of heparin as a mucoitin polysulfuric acid ester, and the finding that blood coagulation was inhibited by polysaccharides artificially esterified with sulfuric acid, Elsner[139] in 1937 began testing natural polysaccharide sulfuric acid esters for anticoagulant effect. Considerable anticoagulant activity was found in watery extracts of the red algae *Gelidium, Chondrus crispus, Iridea laminaroides,* and *Delesseria sanguinea.* The aqueous extracts of the brown algae *Chordaria flagelliformis* and *Laminaria digitata* also had anticoagulant powers, but of a lesser degree.[140] The strength of these extracts was found to be dependent upon such variables as seasonal fluctuations, length and extent of drying of the algal fronds, the age of the extraction, and the amount of heat to which the substances were subjected. Meunier, Molho, and their co-workers in France reported similar studies[141,142] with alginic derivatives in 1949 and 1951. Burt, at the Institute for Seaweed Research in Edinburgh,[107] in 1952 reported some heparin-like activity *in vitro* for laminarin sulfate; others there reported even greater activity the following year for sodium laminarin sulfate used in rabbits and rats. These extracts contained 0.61 to 2.15 sulfate groups per glucose unit. Even though the evolution of the coumarin compounds has progressed tremendously and overshadowed most others, it is important to know the anticoagulant potentials of algae. Not only might it be possible to perfect better agents in this category, but it could also help assess possible undesirable effects from eating certain algae.

Blaine, working in Scotland and in Jamaica, studied the potentialities of laminarin as a blood plasma substitute. He used it successfully on dogs in shock; small test doses in humans produced no adverse reactions. Wilkerson likewise reported some success in administering laminarin to rabbits in shock.[107]

With the advent of the atomic age, concern has been manifested over radioactivity of marine substances exposed to test explosions of nuclear weapons. It has been found that elements of atomic weights 83 to 115, and 127 to 154, are produced in greatest concentration by nuclear fission. Studies are reported on the way on the uptake by seaweeds of radioactive Iodine, Caesium, Rubidium, Cobalt, and Selenium.[107,143] The potential effects of radioactivity on marine organisms are only too obvious.

C. *MISCELLANEOUS USES*

Not only have algae been useful in themselves, but they have also supplied some exotic products which have been highly prized by certain peoples. For example, in the distant Siberian province of Kamchatka, the natives employed dulse in the preparation of a potent alcoholic drink. Similarly the Alaskan Indians, in making an alcoholic concoction called "hoochenoo" (sic!), used the hollow stipes of *Nereocystis*—not for its intrinsic content, but for its mechanical attributes as a worm condenser to cool the distilled spirits.[144] The Irish, like the Alaskan natives, used dulse—washed, dried, then hand-rolled—as a sort of chewing tobacco. During World War II, considerable quantities of dried karengo, a species of *Porphyra,* were used by Maori soldiers serving in the Middle East; chewing karengo was asserted to be more thirst-quenching than chewing gum. On the island of Grand Manon, off the coast of New Brunswick, the reddish-purple leaves of dulse are still considered a great chewing delicacy.[145]

Pliny the Elder (first century B.C.) considered seaweeds valuable as dyes, as did the Roman ladies who used them for cosmetic purposes. They availed themselves of a rouge extracted from the *Fucus* wracks (the word *Fucus* is in fact derived from the Latin word for rouge.) Even today, a similar extract mixed with fish oil is in vogue with Kamchatkan women wanting to redden their cheeks.[146]

Since the seventeenth century, a seaweed glue called "funori" has been derived by the Japanese from the red alga *Gloiopeltis furcata.* Its main utility has been as an adhesive and as a sizing for paper, fiber, or cloth. In Europe, "kelp"—the burnt ash of certain brown seaweeds—was used in glazing pottery and manufacturing glass, as well as a source of soda.[147] The production of "kelp" was a big industry for a whole century, until the development of the saltpeter beds in Chile made it uneconomic.

IV. PLANKTON

A. ZOOPLANKTON

As PREVIOUSLY INDICATED, zooplankton is that variety of plankton which consists of minute animal forms.[148-150] It is collectible in a #2 bolting silk net, and it is thereby separable from the main phytoplanktonic organisms (diatoms and dinoflagellates), which pass readily through such a net.

The chief components of zooplankton are the copepods, prawns or krills, and fish eggs and larvae.[151-154] The most significant, in both bulk and nutritional value, are usually the copepods, which feed on phytoplankton.[155] There have been about 750 species of copepods identified, averaging 2.0 to 5.0 mm. in length. The *Calanus finmarchicus,* for example, is well recognized as an important food for many fish, and is very abundant during the summer around North Atlantic shores. The colder northern seas generally contain the largest quantities of copepods, though the Pacific and tropical water offer more varieties. The mysids and the euphasiids (krills) are the most noteworthy of the higher crustaceans; they comprise some 400 species. They are much larger than the copepods, the euphasiids occasionally reaching a length of 5.0 cm. As is commonly the case, these larger organisms live at deeper levels and come to the surface only during the spawning season. They are then so plentiful in Antarctic waters that the blue and fin whales feeding on them can attain a weight of sixty to seventy tons in two years.[3] G. O. Sars is cited by C. E. Lu[30] as having early discovered that most fish have planktonic eggs and larvae. Even herring, which lay their eggs on the ocean bed, contribute to the planktonic amalgam when their larvae leave the ocean floor and float to the surface.

The use of zooplankton as a nutrient has been discussed with increasing frequency since the latter part of the nineteenth century. Although both freshwater and marine zooplankton are potential sources of food, the oceanic varieties have excited greater interest.[156-158] A search of the recent literature on zooplankton reveals a repetitive pattern of reference to the same group of articles. One of the better summaries is that of Robinson and Bajkov[159] in their 1947 survey of marine plankton for the United States Army Air Force.

W. A. Herdman[160] is frequently credited with the first publication on the eating of zooplankton. His single paragraph communication to *Nature* in 1891 related that eight yachtsmen off the coast of Norway breakfasted well on cooked red copepods, probably *Calanus finmarchicus;* it also mentioned that the Prince of Monaco had already considered the nutrient potentialities of copepods for shipwrecked sailors. Yet even in 1888 Greely[161] had reported that men in his 1882 Arctic Expedition ate shrimp-like crustaceans so small that 700 weighed only an ounce. In 1908, J. Johnstone[162] reported Brand's chemical analysis in 1898 of dried copepod plankton as follows: protein 59 per cent, carbohydrate 20 per cent, fat 7 per cent, chitin 4.7 per cent and ash 9.4 per cent. Shortly before the first World War, Nansen[163] reemphasized the potential value of plankton to castaways. In 1939, Seidenfaden[164] wrote that sailors often considered prawns a delicacy.

In 1939, too, George L. Clarke,[2] of the Woods Hole Oceanographic Institute, wrote in *Science* that he had heard the Germans in Heligoland were studying the use of plankton as a food supplement; he said they considered zooplankton nutritionally equivalent to the best meat, and phytoplankton to rye flour. Clarke hypothesized that if all the organic matter in marine zooplankton were assimilable, it might yield four calories per gram. Thus 750 gm. plankton would be required to produce the 3000 calories needed daily to support a man. If, as he figured, one cubic meter of water yields only 0.1 gm. dry plankton, then 7500 cubic meters of sea water would have to be filtered to provide daily sustenance. This represents a heroic operation, but Clarke suggested using nets in the tidal flows of estuarial or interinsular waters.

The war situation spurred further interest. In 1940, W. A. Jaschnov[165] spoke in Russia of the desirability and the feasibility of using planktonic foods. His opinion was not based on theory alone. Between 1921 and 1937 he had studied the distribution and concentration of zooplankton in the Barents, White, Kara, Laptev, East Siberian and Chukotsk Seas, as well as in the adjacent polar areas. He examined 2500 samples and made 1300 quantitative determinations. From these he concluded that zooplankton contained over 90 per cent copepods, and that *Calanus finmarchicus* made up about 95 per cent of the latter. Like others, Jaschnov also found the highest concentration of zooplankton in the autumn. He calculated that there was an average of 65 tons of *Calanus* available annually per square kilometer of sea surface, equivalent to a biomass of 43 tons. The total biomass available yearly from the northern seas of the USSR was estimated to surpass 50 million tons—40 million tons from the Barents Sea, 5 million from the Kara Sea, 3 million from the Laptev Sea, 2 million from the East Siberian Sea, 1 million from the Chukotsk Sea, and one-half million from the White Sea. Berutski[166] also noted seasonal variations of copepods in the White Lake in Russia. Wiborg[167] classified many of the copepods in the Oslo fiord.

In 1941 Sir John G. Kerr suggested in the House of Commons and in the London *Times* that marine plankton be investigated as a food source.[168] Indeed, soon thereafter, Professor A. C. Hardy[3] enthusiastically urged in *Nature* that Clarke's tidal flow filtrations be tried in the plankton-rich sea lochs of western Scotland. In the following year, Hardy[169] also suggested freshwater plankton as a source of food. In 1493, Judy[4] reported in *Science* on the good aquatic food potentialities of Wisconsin lakes. That these war-inspired enthusiasms might be practical was further attested to by Dr. S. P. Chu, who wrote to Clarke in 1946 from Yunnan that Chinese peasants had long used marine plankton as food.[159] In the same year, too, Sir Hubert Wilkins wrote Bajkov that he had eaten plankton in the Antarctic with no ill effects.

In 1945, Knowlton and Irving,[170] of the Aero-Medical Laboratory, studied the effect of eating plankton upon the water requirements of men on life rafts. Their investigation was basically a physiologic analysis of the nutrients of copepods caught in Buzzard's Bay (Massachusetts) by Bajkov. On the basis of determinations made by Dr. A. Butler, the chloride concentration of the plankton was calculated as about 0.57 per cent, as compared with 1.9 per cent in sea water. The protein content was 51 per cent, while the fat was estimated to vary from 10 to 30 per cent. If 800 ml. drinking water are available daily (from solar still, desalination kit, and captured rain water), they concluded that over 300 calories could be derived from plankton before the limits of salt excretion were reached. With this amount of planktonic protein metabolized, the urinary nitrogen produced would be about 12.4 gm., also a serious consideration when water intake is limited.

Robinson and Bajkov mentioned in their 1947 report[159] that about twenty of their human subjects ate 50 to 100 gm. plankton at one meal without ill effect, and that one individual even consumed a full pound without harm. The plankton in these experiments came from Atlantic, Pacific, and Gulf of Mexico waters. In most instances, the plankton was considered quite palatable. The raw plankton tasted like raw oysters, while the cooked material resembled cooked shrimp. Variations in the smell of raw plankton were produced by admixtures of certain species of diatoms and dinoflagellates.

In the same report, Robinson and Bajkov gave the results of feeding plankton to rats. Knowing that Atlantic plankton could be digested and assimilated by rats (studies at Woods Hole Marine Laboratory, 1945), they successfully repeated the procedure with fresh water plankton from an Ohio lake, and also proved that it had a specific growth-producing value. Similar results were then obtained with Pacific plankton. Comparative analyses of several types of plankton were listed (Table II).

It is apparent that the composition of plankton varies with geography, climate, and the predominant species of organisms. Water comprises 75 to 85 per cent of marine copepod plankton. On a dry weight basis, 45 to 60 per cent may be protein; 4 to 31 per cent, fat; 18 to 23 per cent, carbohydrate; and 0.5 to 20 per cent, ash. Chitin is usually present in less

TABLE II

	Atlantic (Robinson and Bajkov)	*Pacific*	*Barents Sea* (Vinogradov)	*Arctic*
Protein	52.6%	49.0%	59.0%	62.65%
Fat	4.2	7.0	7.0	18.50
Carbohydrate	15.0	11.0	20.0	—
Total Ash	28.2	32.2	14.0	—
Chitin	—	—	—	3.49

than 5 per cent concentration in copepods, but is more abundant in higher crustaceans. It forms the exoskeleton of copepods. Although chitin is not digestible, it passes through the gastro-intestinal tract harmlessly as bulk.

That oceanic plankton can truly be an aid to the survival of drifting castaways has been spectacularly highlighted recently by the *Kon-Tiki,*[171] the *Heretique,*[172] and the *Seven Little Sisters*[173] adventures.

Thor Heyerdahl and five companions left Callao, Peru, on April 29, 1947, on the raft *Kon-Tiki,* seeking to prove that pre-Incan Indians could well have settled the Polynesian Islands after passively drifting westward on prevailing ocean currents. They landed 102 days later, physically none the worse, on the Raroia Atoll in the Tuamotu Archipelago. Throughout the 4300 mile journey, they collected plankton in a conical silk mesh net, often getting several pounds of "porridge" in a few hours. The catch was greatest at night and when the sea was cold. Heyerdahl vividly described the myriads of planktonic organisms netted, stressing especially their colorations and nocturnal fluorescence. Four of the six men really liked plankton. The taste varied with the composition of the day's catch; it ranged from that of shrimp paste to that of lobster, crab, caviar, and oysters.

Dr. Alain Bombard, a French physician and sailing enthusiast, became obsessed with the conviction that victims of shipwreck would have a better chance for survival if they became better acquainted with the resources the sea itself offered. To dramatize his thesis, he sailed his 15-foot rubber raft, the *Heretique,* alone from Tangier across the South Atlantic to the Barbados in 1952. Although the 65-day voyage was physically somewhat traumatic, Bombard sustained himself on plankton, fish, water derived from fish, occasional rain water, and small amounts of sea water.

Even more recently, William Willis, a sailor from the Bronx, followed the *Kon-Tiki* route as the solitary passenger on the balsa raft the *Seven Little Sisters.* He left Callao, Peru, on June 22, 1954, and arrived

115 days later at Samoa, 6000 miles away. Preliminary reports indicate that he, too, sustained himself to a fair extent on plankton.

B. PHYTOPLANKTON (MICROSCOPIC ALGAE)

1. *General Nature*

Phytoplankton or "plant plankton" conists chiefly of microscopic algae, which, with few exceptions, are aquatic organisms. These microscopic growths comprise mainly the Divisions Chlorophyta, Euglenophyta, Chrysophyta, Pyrrophyta, and Cyanophyta. Though thousands of species are represented, the diatoms and unicellular flagellates are predominant. They thrive in both fresh and salt water; however, a given species is specific for the one or the other.

Algae are the only plants that can flourish in the sea. They make available the vast amounts of dissolved minerals and other nutrients; thus neritic plankton (near the coasts) is richer because of the higher concentrations of salts. As previously indicated, phytoplankton provides—directly or indirectly—the basic food for all the ascending hierarchy of marine animals. Freshwater algae play a somewhat similar role in their own habitat. The total quantity of phytoplankton perforce exceeds that of zooplankton. However, effective collection of phytoplankton is very difficult because the individual organisms are so small. As Burlew[41] has so strikingly put it, one quart of moderately thin suspension of *Chlorella pyrenoidosa* contains *twenty billion* cells, a number equivalent to eight times the world's human population!

The color of the sea directly reflects the particulate matter therein.[174] When there is little marine growth, the water is deep blue, since the light penetrates to deep levels, and the yellow and red rays are absorbed. When planktonic growth is abundant, the water assumes the yellow, green, or brown shades of the microscopic algae. Seasonal abundance of certain reddish algal growths may produce the "red water" described since ancient times; to this may also be credited the coloration of the Red Sea and the Vermilion Sea. The nocturnal glow of some seas is attributable to the phosphorescence of certain planktonic organisms.

Reference is made in the literature to microscopic algal studies as early as 1779; Priestly[175] and Ingen-Housz[176] described experiments on the photosynthetic gas exchanges of the simple algae which contaminated their laboratory equipment. However, not until the latter part of the nineteenth century was there any strong interest exhibited in the precise nature of microalgae. These investigations were in three general directions:

1. The chemical constituents of algae (carbohydrates, pigments, and —more recently—the nitrogenous and lipid components).
2. Short-term biochemical experiments, e.g., photosynthesis.
3. The growing of algae in culture.

Cohen[177] first cultured algae in soil in the middle of the nineteenth century, while Famintzin[178] in 1871 was the first to raise them in aqueous medium. The initial extensive employment of microscopic algae was by Engelmann[179] in 1883, in his work on photosynthesis. In 1890, Beijerinck,[180] a Dutch bacteriologist, described the first isolation of algae in pure culture; he obtained *Chlorella vulgaris* and *Scenedesmus acutus* free from bacteria and all other organisms. In 1896 Molisch[181] published his work showing the mineral requirements of simple algae to be similar to those of higher plants. The study of Willstätter and Stoll[182] in 1913 pioneered our knowledge of algal pigments. In 1919 Warburg[183] recommended *Chlorella* as a system in which photosynthesis might be very simply studied; so thorough was he that in 1931 he received the Nobel prize for his observations on cellular respiration, much of them made on *Chlorella viridans*. Since then, *Chlorella* has become a standard organism for studies on photosynthesis. It has also pointed the way for many diverse uses of the byproducts of photosynthesis, as will be discussed later.

By 1920 many algae had been isolated in pure culture. In 1926, Roach,[184] investigating the effects of carbon compounds on soil algae, first introduced quantitative methods in the study of algal growth.

Another landmark was Barker's report[185] in 1935 on oxidative processes in the colorless alga *Prototheca*. Pearsall and Loose[186] demonstrated in 1937 that the metabolic changes in *Chlorella* growth are basically similar to those occurring in the leaf of a higher plant. The history of algal culture techniques was well summarized in 1946 by Pringsheim.[187]

Only during the past two decades have the diverse studies of algal metabolism been unified into a cohesive entity. The first truly comprehensive reviews of algal physiology and chemistry were those of Myers[188] and of Blinks[34] in 1951. In 1953, Schussnig's handbook on plant biochemistry[189] summarized many physiological data on phytoplankton, while

Fogg's monograph[40] offered an invaluable compilation of facts on the metabolism of algae. In the same year, the Carnegie Institution issued "Algal Culture: from laboratory to pilot plant"[41]; this volume, collected and edited by Burlew, made available extensive data on algal metabolism and culture. Additional data on the metabolic aspect of algal growth was provided in the "Growth of Protozoa,"[190] issued in 1953 by the New York Academy of Sciences.

2. *Metabolism, Culture, and Nutritional Values*

Even more so than with zooplankton, the recent intensive interest in phytoplankton has been sparked by the growing disparity between world population and food supply.[191,192] The attractiveness of phytoplankton as a food is enhanced by several inherent qualities: Practically all of it is digestible, in contrast with higher plants, which contain a large proportion of cellulose. The protein content is higher than in field plants, it is of low molecular weight, and it contains most of the required amino acids. Under controlled conditions, an excellent lipid yield can be achieved. Vitamins and minerals are present in abundance. And, best of all, algae can be cultivated and almost always be maintained at a maximal growing rate, whereas land crops utilize sunlight most efficiently only just before harvest. Interesting comparative figures are offered in Table III.

Microscopic algae have a basically simple cellular structure.[193-196] Like other pigmented plants, the algal cell usually comprises a nucleus, one or more chloroplasts, and a cytoplasm containing various formed structures. The pigments are generally concentrated in the chloroplasts, the "solar engines" which are now considered the main sites of photosynthetic activity.[197] Here the pigment enables algal cells to convert inorganic to organic substances by utilizing radiant energy.[198]

The key pigments involved in algal photosynthesis (as condensed from Fogg[40]) are shown at the top of page 31.

The chlorophylls and carotenoids are both fat soluble. The former embody a cyclic tetrapyrrolic nucleus with magnesium; the latter generally contain about 40 carbon atoms. The carotenes are hydrocarbons, while the xanthophylls are oxygen derivatives of the carotenes. The phycobilins differ from the other pigments in being soluble in water but not in fat solvents. They resemble the chlorophylls in possessing a tetrapyrrolic nucleus, but the latter is linear instead of cyclic, and is linked to a globulin instead of to a metal.

TABLE III.—*Elementary Analysis and Calculated Composition of Some Marine Algae, Leaves of Land Plants, and Freshwater Algae**

FORM	*ASH* % dry wt.	*C*	*H* % ash-free dry wt. by analysis	*N*	*R-value*†	*PROT.*	*CBH.* % ash-free dry wt. calculated composition	*LIPIDS*
MARINE ALGAE:								
Gigartini Agardhii	17.82	42.90	6.27	5.83	29.85	36.4	63.6	trace
Ulva sp.	18.72	44.36	6.09	4.87	30.53	30.4	69.6	trace
Amphipleura rutilans	46.24	46.52	6.48	5.67	33.67	35.4	62.8	1.8
Macrocystis pyrifera	37.66	46.02	6.82	6.60	32.24	41.2	57.6	1.2
Navicula torquatum	35.10	48.45	6.72	8.80	36.22	55.0	43.7	1.3
Egregria Menziesii	31.74	49.97	6.29	5.39	36.27	33.7	57.2	9.1
LAND PLANT LEAVES:								
Corn	5.39	46.69	6.04	2.02	31.88	12.6	82.0	5.4
Begonia	12.92	45.73	6.39	3.04	32.00	19.0	77.6	3.4
Pine	3.13	51.83	6.52	2.40	37.74	15.0	65.7	19.3
Alfalfa	10.31	49.90	6.86	7.39	38.00	46.2	44.9	8.9
Castor	13.62	51.12	6.58	5.63	38.02	35.2	51.9	12.9
Sunflower	14.09	51.04	6.29	9.87	38.37	61.7	33.9	4.4
Nasturtium	9.58	50.46	6.91	6.84	38.45	42.8	45.9	11.3
Tobacco	11.80	50.87	6.78	7.70	38.77	48.1	41.7	10.2
Flax	12.25	51.91	7.01	7.11	40.08	44.4	40.8	14.8
Cedar	5.53	53.58	7.08	2.91	40.75	18.2	56.0	25.8
FRESHWATER ALGAE:								
Chlamydomonas sp	4.74	47.46	6.82	5.81	35.27	36.3	58.2	5.5
Anabaenopsis sp	9.35	49.57	7.00	7.28	37.88	45.5	45.6	8.9
Oikomonas termo	5.08	51.76	7.60	5.36	40.85	33.5	45.8	20.7
Stichococcus bacillaris	6.50	52.65	6.99	9.96	41.44	62.3	25.8	11.9
Stichococcus bacillaris	11.24	57.14	8.12	3.61	46.51	22.6	38.5	38.9
Chlorella pyrenoidosa	3.45	49.51	6.78	9.31	37.92	58.0	37.5	4.5
Chlorella pyrenoidosa	3.46	70.17	10.53	1.43	63.33	8.7	5.7	85.6

* Synthesized from several tables cited by Milner[41]

† R-value, as conceived by Spoehr and Milner, is the level of reduction of the total organic matter contained in plant material. The scale of R-values ranges from 0 for CO_2 to 100 for methane.

From the R-value calculated on the basis of carbon, hydrogen, and oxygen analysis, and from the nitrogen content, one can estimate the proportions of protein, carbohydrate, and lipid. Protein is 6.25 x nitrogen.

Key Pigments in Algal Photosynthesis

CHLOROPHYLLS
- Types *a*, *b*, *c*, *d*, *e*

CAROTENOIDS
- *Carotenes*
  - Types α, β, ϵ
- *Xanthophylls*

Lutein	Siphonein	Diadinoxanthin
Zeaxanthin	Siphonoxanthin	Dinoxanthin
Violaxanthin	Fucoxanthin	Neodinoxanthin
Flavoxanthin	Neofucoxanthin	Peridinin
Neoxanthin	Diatoxanthin	Myxoxanthin
		Myxoxanthophyll

PHYCOBILINS
- *r* and *c* Phycoerythrins
- *r* and *c* Phycocyanins

The distribution of these pigments in algae is rather variable. The commonest chlorophyll is the type *a,* just as in higher plants. Chlorophyll *b,* also present in higher plants, occurs only in the Chlorophyceae and the Euglenineae. The other classes contain chlorophylls *c, d,* and *e.* Of the carotenes, the β-type is most abundant. The α-carotene, typical Chlorophyceae and the higher plants, is not found in some algae. In Bacillariophyceae, for example, it is supplanted by ϵ-carotene. Each algal class also has its own characteristic xanthophyll, as may be deduced from the number that have been identified. Phycobilins, on the other hand, occur only in the Rhodophyceae and the Myxophyceae, the former containing mostly phycoerythrin, the latter phycocyanin; the origin of these related pigments in either Rhodophyceae or Myxophyceae is indicated by the designation *r* or *c,* respectively.

Granick and his associates reported[199,200] in 1953 on the protoporphyrin precursors produced by *Chlorella* mutants. They have been particularly interested in the relationship between chlorophyll and hemin, seeking information on porphyria and blood formation in human beings. Della Rosa,[201] using C_{14}, demonstrated that *Chlorella vulgaris* was able to synthesize chlorophyll from acetate and glycine; he also showed that chlorophylls *a* and *b* are not interconvertible. Jorgensen[41] in Venezuela demonstrated to one of us (M.S.) the red blood wiggler *Chironomus* growing on the sides of his *Chlorella* culture containers; he suggested that there might here be a conversion of chlorophyll to heme. French and Young[202] indicated that phycocyanin may be an intermediate in the resonance transfer of energy from phycoerythrin to chlorophyll.

The photosynthetic role of the various pigments can be estimated by comparing the absorption spectrum with the "action spectrum" or photosynthetic capacity of light at various wave lengths.[40,41] Photosynthetic efficiency is usually measured in terms of oxygen production. The chlorophyll absorption bands are chiefly in the blue, blue-green, red, and infrared ranges. The yellow-orange color of the carotenoids results from absorption in the blue and green. The chlorophylls and phycobilins are often strongly fluorescent in solution. Tucker[203] has even used pigment extraction with acetone as a method of quantitating phytoplankton.

Despite all the emphasis on photosynthetic capacities, it must be noted that not all algae contain pigment. Beijerinck[180] long ago noted the natural occurrence of yellow and colorless colonies of *Chlorella variegata* on culture media. Bogorad and Granick[200] produced colorless, pale yellow, and light green mutant colonies of *Chlorella vulgaris* by radiation. Butler[204] exposed *Chlorella pyrenoidosa* to ultraviolet light and obtained colorless mutants without chlorophyll. He also noted that similar permanently colorless forms of *Prototheca* have been observed in nature, and that they were probably mutants from Chlorophyceae. Very recently Tolbert and Zill[205] reported photosynthetic studies with protoplasm extruded from *Chara* and *Nitella*. They found activity 12-15 per cent that of the whole cell, as measured by C_{14} fixation. Both protoplasm and cut cells reduced carbon dioxide in light to sucrose and hexose phosphate. Dark controls fixed C_{14} into products associated with plant respiration, but it is not clear whether they represented true photosynthetic pigments. In fact, the whole sequence of biochemical synthesis in the dark or without pigment remains rather obscure. This is particularly so in the light of Duysens' claim[206] that light energy is convertible only through chlorophyll *a* or *b* (he maintains that in diatoms, for example, carotene must be converted to chlorophyll *a*.)

Radiation and natural mutation are not the only methods of producing non-pigmented algae. Dubie[207] produced chlorophyll-deficient *Chlorella vulgaris* with streptomycin. And even strong radiation does not always effect alterations. Blinks[208] reported there was no significant change in composition or function of marine algae one year after the atomic explosions in the Bikini area. The only exception was an increase in catalase, possibly due to an increased hydrogen peroxide concentration secondary to radiation.

Algal cells generally do not grow old and die; instead, they produce more cells by binary fission. However, not all algae grow by cellular division. It has not been widely realized that sex may play a role even in this remote corner of the cosmos! Sexual union of two gametes to produce a zygote does occur, particularly among the Phaeophyta and Chlorophyta (*Chlorella* and *Scenedesmus* are notable exceptions.) Either or both gametes may be flagellated, or mating may be achieved by ameboid motion. Gilbert M. Smith[209] notes that algal sexuality was initially described in 1858 in *Oedogonium.* In 1926, Jollos demonstrated that the heterothallic green alga *Dasycladus* excretes sexual substances which influence the movement of gametes toward each other. Since then, much experimentation has been carried on with *Chlamydomonas eugametos,* a unicellular green alga. Motility is absent in cells grown anaerobically in the dark, but it is revived by filtrates from cells grown in light. The substance responsible is reported to be *crocin,* an ester of the sugar *gentiobiose* combined with the carotenoid pigment *crocetin.* There are also substances affecting maleness and femaleness; these are activated by exposure to light, weakened by dilution. The virilizing factor appears to be *safranal,* the feminizing one a *gentiobiose* of *safranal.* There are all degrees of sexual activity, depending upon the concentration of activating substances and exposure to light. There are also varying degrees of maleness and femaleness, even intermediary forms, depending upon the ratio of opposing substances.

Similar findings were reported by Sager and Granick[211] in 1954. They noted that depleting the nitrogen supply in culture media of *Chlamydomonas reinhardi* encourages the differentiation of ordinary cells to gametes. If nitrogen is added, the process is reversed. Light apparently acts only indirectly via photosynthesis, and it is not obligatory for zygote formation of dark-grown nitrogen-depleted cells.

Much has been learned about the metabolic characteristics ol algae by observations of cultures. *Chlorella* has been widely used in photosynthetic experiments especially because it can grow under such varied environmental conditions. The rate of algal growth is often measured by "generation time"—the time required to double the number of cells in a culture.[41] There are two major phases of growth, one chiefly of cellular multiplication, the other of storage of reserve materials. In the first phase, proteins predominate; in the second, fats and carbohydrates. The main factors influencing algal growth are: available radiant energy, carbon dioxide concentration, temperature, and composition of nutrient substrates. The exact channeling of the various nutrients may depend upon the amount of available nitrogen, or upon the presence of enzyme systems. *Chlorella* extracts, for example, have been shown to contain transaminase systems which aid the transfer of amino groups to either pyruvic or *a*-keto-glutaric acid.[212] The most active enzyme systems in algae are those of aspartic and glutaric acids, and alanine.

Spoehr and Milner[41] demonstrated that the composition of *Chlorella pyrenoidosa* could be varied specifically simply by controlling cultural conditions. In over 300 pure cultures, it has been possible to produce the following wide range of variations in composition and R-value:

Protein:	7.3 to 88.0%	Carbohydrate:	5.7 to 38.0%
Lipid:	4.5 to 86.0%	R-value:	37.92 to 63.33

The amount of radiant energy present markedly influences the rate of growth of a culture, either directly or by increasing the nitrogen absorption. The latter is certainly of critical importance, since a *Chlorella* cell can no longer undergo fission when the nitrogen content falls below 1.6×10^{-13} gm.

As has already been indicated, the protein content of microscopic algae may vary widely; its level may be grossly estimated from the nitrogen content. There is a definite similarity in the protein content of *Chlorella* and the leaves of many higher plants. Under optimal cultural conditions, a protein level can be achieved in *Chlorella* which exceeds that of the best vegetable substances used as animal feed. This includes such materials as Brewer's yeast, torula yeast, soybean meal, dried skimmed milk, and wheat.

Not very much is known of the specific nature of algal proteins, except that there is often a resemblance between their amino acid patterns and those of higher plants.[213-215] The nutritional value of *Chlorella* is based not merely on its high protein content, but also on the fact that it contains all the essential amino acids. Methionine is present in only small amounts, but this deficiency also occurs in many vegetable proteins. The essential amino acid Biological Index of *Chlorella vulgaris* has been calculated to be 62 in a pilot plant assay, as compared with an arbitrary value of 100 for whole egg protein. The Index of most animal proteins ranges from 80 to 90, while cereal proteins are usually between 60 and 80. Thus *Chlorella* is roughly comparable to white flour, corn gluten, and peanut meal. An amino acid assay of dried *Chlorella* is given in Table IV.

Although most algae are able to utilize wide varieties of basic inorganic nutrients and to synthesize from them the more complex metabolites required, some are not so versatile. They may not be able to synthesize all

TABLE IV.—*Amino Acid Assay of Dried Chlorella*

Nutrient	*Pilot Plant Sample*	*Laboratory Sample*	*Torula Yeast*
Crude protein......	44.00%	40.00%	—
Arginine...........	2.06	2.39	3.61%
Histidine..........	0.62	0.65	1.31
Isoleucine..........	1.75	1.69	3.75
Leucine............	3.79	1.99	3.57
Lysine.............	2.06	2.43	4.14
Methionine........	0.36	0.57	0.84
Phenylalanine......	1.81	2.14	2.41
Threonine.........	2.12	1.91	2.58
Tryptophane.......	0.80	0.41	0.66
Valine.............	2.47	2.67	2.98
Glycine............	—	2.20	0.22

the amino acids from ammonia and the requisite carbon fractions, and they must thus depend at least in part upon an exogenous source of some amino acids. The deficiency is not of the amino acid *per se,* but of the capacity to form amino groups from ammonia. Thus, for proper growth, *Euglena deses* requires aspartic acid, while *Chlamydomonas chlamydogama* needs histidine and aspartic acid.[40]

Nucleic acid has been demonstrated by staining reactions to be present in algal cells. Jeener[216] reported that *Polytomella caeca* had 6 to 10 per cent of its protein as ribonucleic acid in actively growing cells. He indicated that there was great variability in the synthesis of nucleic acid, and that this was unrelated to the rate of general protein synthesis and cellular multiplication. Szafarz and Brachet[217] indicated that formation of ribonucleic acid can proceed independently of the nucleus in *Acetabularia mediterranea.* Studies by Goryunova[218] have indicated the presence of mucins in *Oscillatoria.*

The assimilation of nitrogen by algae is important not only with respect to amino acids, but also in the synthesis of various carbon compounds. As in the case of certain bacteria, many autotrophic algae are characteristically capable of nitrogen fixation. Nitrogen may be absorbed as the element, or as nitrate or ammonia. Frank,[219] in 1889, first reported the possible nitrogen-fixing properties of certain algae, but this was not proven until 1928.[220] Since then, further corroboration has come from Kjeldahl determinations and tracer studies using nitrogen isotopes.[40] So far, over 20 species of Myxophyceae have been proven capable of fixing nitrogen. Aspartic acid, glutamic acid, succinamide, asparagine, and glutamine can serve as the sole sources of nitrogen for various green algae via deamination or transamination. It is probable that nitrogen enters the general metabolic pool most often in this manner.

Nitrogen-fixing algae can contribute to the fertility of the soil, especially in tropical areas. A notable example is the utilization of blue-green algae in the Usar area of northern India to decrease the excessive alkalinity of the soil.[15] The algae thrive during the rainy season and decrease the pH of the soil from 9.0 to as low as 7.0, and thus make possible the growing of field crops during the dry season. This resembles somewhat the employment elsewhere of large seaweeds as direct fertilizer.

The cultivation of rice is also aided by microalgae. The latter flourish in the wet rice paddies and, by fixing atmospheric nitrogen, enable perennial cultivation without additional fertilizer. Nitrogen fixation here may be greatly enhanced if bacteria are also present and contribute their fixation activities. In addition, the algae, by producing oxygen, help to aerate the rice plants. Watanabe[221] reported in 1951 that 13 species of 643 blue-green algae from Far East and South Sea area rice fields were capable of atmospheric nitrogen fixation, chiefly in warmer climates. Most of them belonged to the genera *Tolypothrix* and *Nostoc,* some to *Schizothrix, Calothrix, Anabaenopsis,* and *Plectonema.* Aspartic and glutamic acids and alanine were the systems mostly involved. A similar function was reported by Douin,[222] who noted a symbiosis between the *Cycadaces* plant and *Anabaena cycadeae,* which fixes nitrogen for its host.

Much less is known about the carbohydrates of microscopic algae than of the macroscopic.[40,41] Whereas the latter may contain large quantities of carbohydrates in the cell walls (chiefly alginic complexes), the microalgae have relatively scanty structural materials, and their storage products are lipid rather than starchy. Milner has isolated starch and sucrose from *Chlorella pyrenoidosa.* Broun found 17.8–20.2 per cent (dry weight) total carbohydrate in *Scenedesmus obliquus.* The substances included mainly insoluble polysaccharide; there were also small amounts of free reducing sugar, sucrose and other oligosaccharides, and water-soluble polysaccharides.

Healthy algal cells, particularly in older cultures, liberate various organic substances into their fluid environment. These consist primarily of nitrogenous substances and pentosans. It is probable that more specific knowledge of microalgal carbohydrates will emerge when higher carbohydrate contents are achieved by varying cultural environmental conditions.

The lipids represent one of the most important components of microscopic algae.[223] Many investigators have confirmed Beijerinck's observation in 1921 that fat droplets accumulate in diatoms as the cultures become old and the available fixed nitrogen decreases.[40,41] This occurs, as we have indicated earlier, because nitrogen deficiency limits growth and favors the building of reserve materials. The ratio of lipid to carbohydrate is variable; it depends far less on algal genetic traits than it does on environmental factors such as nitrogen supply and enzyme systems. Also noteworthy is the fact that algal fat metabolism is but little reflected in gas exchanges.

The lipid content differs not only from one species to another (the blue-greens have very little), but also may vary greatly within the same species with changes in cultural conditions. *Chlorella* generally contains 20–25 per cent fat, yet nitrogen starvation can increase the amount to as high as 86 per cent. Even during World War II, the Germans, culturing *Chlorella, Nitzschia,* and *Scenedesmus* as potential sources of needed fat, attained lipid yields of 40–70 per cent. These comprised chiefly the triglycerides of stearic, oleic, and linoleic acids.

Most of the fat in phytoplankton is unsaturated, with a high proportion in the 16 and 18 carbon series.[41] Detailed analyses of *Chlorella,* for example, show 54–67 per cent unsaturated C-18 and 18–29 per cent C-16 acids. Unsaturation ranges from 1.60 to 2.25 double bonds per molecule. Palmitic is the main saturated acid, while stearic constitutes less than 4 per cent of the total fatty acid. As the lipid content of *Chlorella* rises, the unsaponifiable fraction decreases and the fatty acid portion increases; at the same time there is a significant fall in the degree of unsaturation of the fatty acids. *Chlorella* fat resembles other unsaturated plant lipids and falls into the chemical classification of "drying oils."

Sterols have been obtained from most of the algal groups, excepting the blue-greens. Klosty and Bergmann[224] reported in 1952 that chondrillasterol is the principal sterol in *Scenedesmus obliquus,* while ergosterol predominates in *Chlorella pyrenoidosa.* Chondrillasterol may be remembered as having been favorably considered a few years ago as a starting point for cortisone synthesis. Table V offers analyses of the various lipids in several samples of *Chlorella.*

Phytoplankton, like the macroscopic seaweeds, has a high vitamin content. It has been determined that most of the known vitamins are present in *Chlorella* and related microalgae. The quantities are eminently adequate for the algae to be used as food (for man or animals), but not if

TABLE V.—*Analysis of the Lipids of Chlorella*

Analysis	*Chlorella* lot no. 1	2	3	4
1. Total lipid (% of *Chlorella*)	23.37	33.17	62.96	75.51
Composition of total lipid				
2. Fatty acids (% of lipid)	28.0	49.5	83.0	86.8
3. Unsaponifiable fraction (% of lipid)	12.0	7.7	3.3	3.3
4. Water-soluble, after saponification (% of lipid)	60.0	42.8	13.7	9.9
5. Calculated fat (% of *Chlorella*)	6.85	17.2	54.7	68.6
Analysis of total fatty acids				
6. Palmitic acid, C16 (%)	16.6	10.9	7.9	11.4
7. Stearic acid, C18 (%)	0.4	4.1	3.9	3.5
8. C16 unsaturated acids (%)	29.1	18.3	27.2	18.0
9. C18 unsaturated acids (%)	53.9	66.7	60.9	67.1
10. Total fatty acids (%)	100.0	100.0	99.9	100.0
11. Equivalent weight	269.5	273.6	272.7	274.1
12. Iodine number	163.1	143.8	143.6	125.3
Degree of unsaturation				
13. C16 plus C18 unsaturated acids	−4.4H	−3.6H	−3.7H	−3.2H
14. C16 unsaturated acids	−4.1H		−4.4H	
15. C18 unsaturated acids	−4.5H		−3.4H	

they are intended purely as a source of vitamin concentrates. In the latter case, synthetic methods are more economical.

Microscopic algae are the ultimate basic source of vitamins in fish, and the finding of large quantities of Vitamin A in fish livers was indeed the stimulus for much of the study of algal vitamins.[225] Several decades ago, it was shown that *Nitzschia closterium* and other phytoplankton were good sources of Vitamin A and its carotene precursors.[226] In the early 1930's, Jorgensen in Scandinavia considered using algal carotene to feed cows and to fortify margarine.[41] Later, in Venezuela, he used dried plankton from Lake Maracaibo as a source of carotene and related unidentified substances as a highly efficacious food supplement in the therapy of lepers.

Vitamin B_1 is generally necessary for algal growth, and it has been demonstrated in most species in which it has been sought.[40] Especially large amounts are found in *Chlorella*. Young cultures with actively-dividing cells contain more thiamin than do the older fat-storing ones. Thus, von Witsch obtained yields of 1.8 to 18.0 gamma per gram of dried substance from cultures of varying ages.

While *Chlorella* and related algae contain dehydrogenase for direct oxidation of pyruvic acid, there is also some alternative participation in the Krebs tricarboxylic acid cycle, as in vertebrate tissues. For this reaction, the co-carboxylase thiamin pyrophosphate must be present. An example is the great enhancement of pyruvic acid oxidation by the addition of thiamin to the thiamin-deficient *Prototheca.* If Vitamin B_1 is not present as such in the aqueous nutrient, it must be synthesized by the algae. Most are capable of so doing, but some cannot synthesize the thiazole, others the pyrimidine, radicles of thiamin. Thus the thiazole portion must be supplied to *Polytomella ocellata,* and the pyrimidine component to *Euglena gracilis* and to a radiation mutant of *Chlamydomonas moewusii.* The pyrimidine-thiamin requirement of the *Euglena* can be markedly decreased by the addition of glutamate. Both thiazole and pyrimidine must be supplied exogenously for the growth of *Polytomella caeca, Chilomonas paramoecium,* and *Prototheca zopfi.*

Riboflavin, like thiamin, has been demonstrated in practically all algae in which it has been sought. It is a component of the prosthetic group of the flavo-proteins, but its essentiality to algal growth has not been shown. Para-aminobenzoic acid is known to be necessary for the growth of only one alga, a mutant of *Chlamydomonas moewusii.* With the latter and with *Nitzschia, para-aminobenzoic* acid has the same antagonism to sulfonamide inhibition of growth as with many bacteria. Aniline may replace para-aminobenzoic acid as a growth factor for *Chlamydomonas* with an efficiency of one per cent, but it does not neutralize the sulfonamide effect.[40,227]

Plankton paste contains considerable quantities of cyanocobalamin or Vitamin B_{12}.[40,41,228] Traces of this substance are necessary for the growth of various Euglenineae as well as other classes, such as *Chlamydomonas chlamydogama* of the Chlorophyceae. Vitamin B_{12} may be concerned in the synthesis of desoxyribosenucleic acid.

Ascorbic acid is present in phytoplankton (especially *Chlorella*) in amounts comparable to those in lemon juice; however most of it is lost in drying and storage. Although little is said about the Vitamin D content of microalagae, feeding experiments have indicated good antirachitic activity. Indirect evidence comes from the potency of fish liver oils, which ultimately derive their Vitamin D from planktonic sources.[229] As with seaweeds, the vitamin activity of phytoplankton is based upon ergosterol content. Substances with coagulant effect have been demonstrated by Dam, who determined the Vitamin K content of *Chlorella* as six gamma per gram of dried material.[230]

Phytoplankton has a great potential as a source of vitamins in human nutrition. Though most of the studies have been with *Chlorella,* other microalgae are comparable. It has been calculated that a quarter pound of *Chlorella* will supply all the daily minimal human vitamin needs except for ascorbic acid. Table VI, taken from Burlew,[41] details the vitamin content of two samples of *Chlorella.*

TABLE VI.—*Vitamin Assay of Dried Chlorella*

Vitamin	*Pilot Plant Sample*	*Laboratory Sample*
Carotene (mg./lb.)	—	218.0
Thiamin (mg./lb.)	11.0	4.5
Riboflavin (mg./lb.)	26.2	16.3
Niacin (mg./lb.)	54.0	109.0
Pyridoxine (mg./lb.)	—	10.4
Pantothenic acid (mg./lb.)	3.6	9.1
Choline (mg./lb.)	—	1370.0
Biotin (mcg./lb.)	—	67.0
Vitamin B_{12} (mcg./lb.)	45.0	10.0
Lipoic acid (acetate units/lb.)	1.5	—

Algae require basically the same nutrients as higher plants—nitrogen, phosphorus, potassium, sulfur, magnesium, and iron. Unlike the higher plants, all except a few algae can dispense with calcium. Potassium and magnesium (and occasionally calcium) are important in the photosynthetic process because their bicarbonates can make available supplemental carbon dioxide. In addition, algae also have need for certain trace elements or micronutrients. Myers[41] has specified algal requirements for manganese, zinc, calcium, boron, and copper. The exact metabolic role of these has not been elucidated. Neither has it been for the following substances, which have also been identified in algal cells: antimony, arsenic, beryllium, cobalt, germanium, iodine, lead, manganese, molybdenum, nickel, rubidium, silver, strontium, tin, titanium, tungsten, uranium, vanadium, and yttrium. (Does not this listing of elements sound more like a roll-call of our present-day electronics and metallurgical industries, and are not the biologic and the mechanistic worlds drawing closer?) It has been determined that, because of selective absorption, the cellular content of manganese, strontium, uranium, and yttrium is above that of the surrounding media. The only known role of molybdenum is that of fixing nitrogen in *Anabaena.*

Even though their functions are obscure, the addition of trace elements to algal culture media has become standard operating procedure. An excellent means for providing continuously adequate micronutrients are the chelating or complexing agents. These are biologically inert cyclic organic compounds ("inner complex salts") resulting from the attachment of a group at two points to the same metallic atom. They prevent excessive absorption and precipitation. Ethylenediamine tetraacetic acid (EDTA) is the chelating agent most commonly used. When a given amount is added to a nutrient medium, it will permit the release of enough ions through mass action to provide for cellular needs. By forming easily reversible complexes of trace elements, it provides tonic concentrations at preferred levels throughout the life of a culture.

Besides the trace elements, there are other unidentified factors sometimes necessary for algal growth. It is common observation that some algae grow with ease in impure culture, yet do not flourish in bacteria-free media. *Gymnodinium* cannot be continuously subcultured without an organic factor extracted from soil. Similarly *Cryptomonas ovata, Synura ovella, and Gloeotrichia echinulata* demand the presence of certain soil factors. Also, *Ditylium brightwelli* requires, for good growth in artificial seawater, two organic substances from natural seawater. One of these is a specific sulfur grouping, the other a substance also found in yeast or algal extracts.[40] The exact nature of these various needed supplementary compounds will doubtless become clearer with more experience in algal culture.

Although mass culture of phytoplankton for food and special organic material has long been the subject of considerable discussional enthusiasm, little specific investigation was done until World War II.[231] Most of the experiments have been performed with some species of *Chlorella, Scenedesmus,* or *Nitzschia* because they grow so fast and tolerate a diversity of cultural conditions. *Chlorella pyrenoidosa* has been an especial favorite because of its previous wide use in studies on photosynthesis. As listed by Burlew in "Algal Culture, from Laboratory to Pilot Plant,"[41] the elements of a plant for growing algae on a large scale are relatively simple:

1. A container with a transparent upper surface.
2. A means of circulating the culture medium, so that the algae do not settle.
3. A means of controlling temperature.
4. A means of introducing carbon dioxide and other nutrients continuously.
5. A means of harvesting the algae almost continuously.
6. A means of preserving the harvest until used.

A great variety of apparatus has been employed to achieve the mechanical requisites. In 1951, Jorgensen and Convit in Venezuela grew freshwater algae in unglazed baked red clay bowls exposed to the open air and hot sun. The nutriment consisted of a commercial fertilizer suspended in water. Evaporation from the porous sides of the bowls kept the temperature at 26°C. even in direct tropical sunlight. Von Witsch and Harder in Germany utilized nutrient solutions in glass tubes 3 x 30 cm. and 6 x 150 cm.; these were illuminated by 300-watt water-cooled lamps and supplemented with an air stream containing 0.5 per cent carbon dioxide. At the Carnegie Institution in Washington, greenhouse culture of *Chlorella* was carried on in five-gallon bottles in 1947; illumination, temperature, and carbon dioxide supply were controlled. This study led the following year to activation of a successful pilot plant for continuous *Chlorella* culture at the Stanford Research Institute.

In 1949, Geoghegan in England tried out a variety of culture vessels: cylinders 15 x 1.75 inches; large tubes 4.5 feet x 2.75 inches; aspirator bottles 18 x 10.5 inches; and a tall outdoor plastic tank 4.5 feet high, 1.5 feet long, and 0.33 feet wide. He concluded that shallow horizontal tubes or troughs were preferable, and that sunlight is more economical than artificial illumination. Wassink and a group at the Agricultural University at Wageningen in Holland experimented with mass *Chlorella* culture from 1948 to 1950. Initially they used inverted half-liter glass-stoppered bottles with incandescent light. Later they tried outdoor sunlit square meter tanks with 300 liter capacity. The tanks were covered with glass to minimize contamination; carbon dioxide was supplied from a cylinder; and agitation was accomplished by a motorized stirrer. Algae were harvested after 5 to 7 days of growth. Subsequently they set up similar concrete tanks in a dark room and illuminated them with either daylight fluorescent bulbs or high-wattage incandescent lamps. They obtained a 12–20 per cent efficiency of light-energy conversion, and therefrom concluded that excessive illumination could decrease algal growth under either artificial or natural conditions.

Gummert and his colleagues, working in Essen in 1950–51, hoped to demonstrate that local conditions were suitable for large-scale outdoor and greenhouse culture that could take advantage of the huge amounts of waste carbon dioxide in the industrial Ruhr. They succeeded for the most part, but their deliberately nonsterile procedures resulted in troublesome contamination with foreign algae and protozoa. This could be overcome by using protozoa-resistant *Scenedesmus,* by altering the nutrient media, by increasing the amount of illumination, or, in fact, by doing anything that promoted faster growth of the desired algae.

Much was accomplished in America from 1950 to 1953 under the guidance of the Carnegie Institution of Washington. They helped sponsor such investigators as Spoehr, Milner, Davis, Myers, and Krauss, with much of the work done in the Department of Plant Biology at Stanford. Here studies were performed not only with various types of algae and nutriments, but also with differing forms of culture equipment. The latter included large bottles, rocking trays, and plastic and glass tubing. The culmination of the Carnegie studies was the establishment of a large pilot plant for *Chlorella* culture in Cambridge, Massachusetts, by Arthur D. Little, Inc., a consulting research and development organization. Three large-scale culture units were developed and studied. The last, and best, consisted of a massive plastic tubing in U-shape, set up on a roof-top with free exposure to sunlight. It was equipped with a centrifugal pump to circulate the culture, a measured inflow of carbon dioxide (5 per cent in air), a heat exchanger for cooling the culture, a harvesting system, and apparatus to freeze or spray dry the harvested material. From this study, it was concluded that with this type of pilot plant, 20 gm./sq. meter/day could be realized, equivalent to 17.5 tons *Chlorella* per acre per year.

Pilot plant studies were also carried on in Israel and in Japan. The former were begun in 1951 at the Hebrew University in Jerusalem under government sponsorship. The latter were done in Tokyo in 1952 at the Tokugawa Institute for Biological Research; Tamiya, the director of this group, spent some time as guest worker in the Carnegie Institution laboratories in Stanford through a fellowship supported by the United States Office of Naval Research.

Experiments on mass culture of algae are also being carried on by segments of private industry. An example is the reported work of Pruess and his colleagues[232] for a pharmaceutical concern on algal culture in carboys and in deep tank fermentations. Kindred to the latter is the growing of algae in so-called "oxidation ponds," with the double purpose of sewage disposal and formation of algal protein suitable for feeding. Renn[233] has estimated that domestic sewage from a community of 10,000 could produce nearly 1,400 pounds of protein daily if properly treated. Oswald and his colleagues[234] at the University of California are also enthusiastic proponents of this method. One eager college professor[22] has already designed a house with an algal pond on its flat roof. No comment is offered on the monotony of having to stick to one brand of sewage!

The enthusiasm for culturing phytoplankton for feeding is based on reports that it is innocuous and that it possesses great nutritional value. These have been well summarized by Fisher and Burlew.[41] Mention has already been made of Jorgensen's feeding plankton soups to lepers in 1941, with a resultant improvement in weight and energy. These comprised cultures of *Chroococcus, Homocystinea, Oocystis, Ankistrodesmus, Chlorella,* and *Scenedesmus*. More recently, Jorgensen told one of us (M.S.) that an algal broth, when fed to mice with skin cancer, brought about an appreciable decrease in the size of the lesions; inexplicably, however, test animals maintained on such a diet did not have a long life span. Currently Jorgensen is feeding his phytoplankton in rather palatable compressed pills.

Another early feeding experiment was that in 1949 at the Stanford Research Institute. Rats fed on a diet containing a third *Chlorella pyrenoidosa* had a weight gain only one-third that of a control group. This has been attributed either to the unpalatable flavor or to the mechanical difficulty resulting from the hygroscopic nature of the food preparation. Henry, at the National Institute for Research in Dairying, in England, fed freeze-dried *Chlorella* prepared by Geoghegan to young rats. No adverse effects resulted after four months on a 17 per cent *Chlorella* diet. In a four-week comparative study, the protein efficiency of *Chlorella* appeared slightly superior to that of dried brewer's yeast and peanut meal, but inferior to that of skim milk.

In 1951, Combs,[235] at the University of Maryland, tested New Hampshire chickens on *Chlorella* provided by the Carnegie Institution. A marked increase in growth and an improvement in feed efficiency resulted from adding 10 per cent *Chlorella* to a basal diet in place of soybean meal. This was attributed to the high vitamin content of the alga. However, a growth depressant effect was noted after extended feeding because of impacted beak conditions. This occurred much sooner with 20 per cent *Chlorella* feedings. Similar results were obtained in 1952 with chicks when vacuum-dried *Chlorella* was tried by a feed supply concern. Most of the difficulties encountered were apparently due to excessive water absorption by the algae. Other methods of preparation would seem to be indicated.

Tamiya[22,41] has included substantial amounts of *Chlorella* in preparing highly acceptable bread, noodles, soup, tea, and ice cream. He has also used hydrolysates of dried algae as a substitute for soy sauce. His students and even his colleagues admitted no ill effects whatever from repeated ingestion of these concoctions.

The palatability and acceptability of phytoplankton as a human food are dependent upon many factors. These include especially appearance and taste. These depend, in turn, upon whether the material is fresh or dried; also, whether it is eaten as such, or mixed (and diluted) with more familiar foods. Doubtless much improvement can and will be made in all these respects for a greater acceptability. Certainly these items must be worked out before plankton is really the "food of the future."[236]

3. *Medical Aspects*

a. *Water Supply and Sewage Disposal*

Despite all the hopeful reports on their merits, it must be emphasized that phytoplanktonic organisms can exert adverse effects upon the waters in which they flourish. These include not only changes in appearance, odor, and taste of the water, but also changes in other aquatic organisms.[237-243] Even more important are the varying degrees of intoxication and reaction in animals and humans who ingest or come in contact with these waters. The potential economic losses are self-evident.

Similar to the striking colorations of the seas previously mentioned (e.g., Red Sea, Vermilion Sea) are the alterations in appearance of fresh water bodies. "Water bloom" is present when extensive growths of microscopic algae blanket the surface of a lake, pond, or stream. In Europe the phenomenon has been variously called "Wasserblüthe," "fleur d'eau," and "flos aquae"; in some North Central parts of the United States, the ancestral Dutch "vasserbloom" designation is used. Under favorable circumstances, microscopic algae, especially the Myxophyceae, can grow so rapidly that the water assumes the color of the organism. And these colors are indeed variegated. The *Oscillatoria* and *Cylindrospermium* produce a green slime and a bluish soup, respectively. *Chlamydomonas* and *Pleurococcus* are a brilliant green. *Ceratium hirundinella* presents a rusty color and a powerful stench—perhaps the basis of the Biblical story (Exodus: 7) of the turning of the Nile into blood and the death of the fish therein. "Red water" and "red snow" in Australia are due primarily to Myxophyceae, but desmids (*Conjugata*) may be a factor. Both "red tide" and "yellow tide" have been reported along the coast of the Gulf of Mexico, while two major outbreaks of "red water" occurred in Gokasho Bay, Japan, in 1933 and 1934, from *Gymnodinium mikimotoi*.

Whereas the colorations are interesting and occasionally even weird, the odors arising from aquatic algal growths are more important, both esthetically and economically. They affect water supply and sewage disposal. Odors and tastes may arise from the oils of living algae; from products of active photosynthesis; from products of decomposition; and from the death of fish and other aquatic organisms. The oils of living algae are often unpleasantly odorous, each oil being characteristic of the generative organism. The smells may be fishy, aromatic, or grassy. Their disagreeable character may be heightened after planktonic death, when the frail oil globules are more widely dispersed. The odorous photosynthetic substances liberated during very rapid growth are ill-defined, but the offending materials present after algal decomposition are better known. They include hydrogen sulfide and other sulfurous compounds, phosphorous and nitrogenous substances, methane, ethereal sulfates, and other volatile gases. Fish and other aquatic organisms may die from suffocation, or from toxic substances such as hydroxylamine resulting from the breakdown of algal proteins. These dead animals contribute a liberal stench to the already odoriferous situation. Often enough, the chemical treatments applied to kill off offending algae aggravate the situation even further. The treatment, such as chlorination, kills the organism and liberates the taste and odor-producing materials, and chemical combination with chlorine may favor the formation of additional smelly compounds.

The most troublesome algae in lakes and reservoirs are the blue-green ones, which grow rapidly and float high in the water, forming surface scums. Greatest growth takes place in waters which are shallow and warm, and in those rich in nitrogen, phosphorus, and loosely-bound carbohydrates. The principal causes of bad odors and tastes are the Myxophyceae and Diatomales, which are less potent oxygenators than are other algae.

When unpleasant odors affect ordinary lakes or ponds they merely detract from the beauty of nature. But when they occur in reservoirs of water intended for daily drinking, they present a trenchant problem. Even mild alterations in smell or taste may be psychologically unacceptable to the average person. As Howard and Berry[238] have pointed out, odors caused directly or indirectly by living plankton or by the decomposition of vegetable growths are immediately ascribed by the layman to contamination with sewage. This is not too strange, since the decomposing plankton floats on the water, then is thrown onto the shore, where it forms odorous, blackish, discolored masses. It is difficult to convince people that the odor itself signifies no health hazard. When living algae are present in the drinking water, there may be a mild initial taste, but a bitter aftertaste; when the organisms are dead, there is a strong first taste, but practically no aftertaste. Measures to control algal growth[244-248] in reservoirs include periodic chemical treatment (with algicides such as copper sulfate), removal of accumulations from the shores before decomposition occurs, and restricting the food supply of the algae by removal of tree branches, leaves, and other floating debris.

While removal or eradication of algae is desirable in reservoirs, quite the opposite is true in sewage disposal units. Oxidation ponds have been widely used for secondary sewage treatment. As summarized by Oswald and his colleagues,[234] oxidative sewage treatment is based upon the fact that organic matter decomposes rapidly in water. When in solution, it is readily available to microorganisms and their enzymes. These convert complex organic compounds into simple substances such as carbon dioxide and ammonia. Because oxygen is the ultimate hydrogen acceptor in these rapid conversions, it may become so depleted that aerobic oxidation is halted. Thus the activated sludge and the trickling filter processes are so designed that atmospheric oxygen is forced into the liquid phase at an accelerated rate. Algal photosynthesis offers a completely different method of supplying oxygen. Here the availability of oxygen is independent of the physical laws normally governing aeration from atmospheric sources. Bodies of water containing green algae may attain supersaturation with dissolved oxygen up to three or four times the normal maximum of water in equilibrium with air. The organic waste material is changed by the cyclic activity of algae and aerobic bacteria into more algal cells. These are then disposed into natural streams or allowed to settle and then be digested anaerobically as bottom sludge. The particular merits of this method lie in the minimal odor, nuisance, and operating cost. It is therefore considered ideal for small communities situated in mild climates. Particularly in Texas and Southern California has there been reliance on the use of sewage lagoons and oxidation ponds for adequate and economical treatment of sewage. Algal purification of drinking water is also sometimes employed; on the other hand, uninvited algae often clog standard sand filtration plants.

As previously indicated (section on Culture), Renn[233] has recommended that oxidation ponds be used not merely for sewage disposal, but also to convert nitrogenous wastes into food protein. This can be accomplished by encouraging the growth of algae (especially *Chlorella, Scenedesmus,* and *Chlamydomonas*) which yield a high quality protein.

A relatively new problem in sewage disposal concerns atomic wastes.[251] Coopey[252] demonstrated radioactivity in all forms of life in the Columbia

River below the Hanford Atomic Energy plant in Washington. He estimated that about 570 tons of wet weight plankton passed the plant daily, with a resulting planktonic radioactivity about 0.6 curies per day. Analyses indicated that 5–30 per cent of the plankton's radioactivity came from isotopes with half-lives near to or longer than P_{32}, while the remainder was of shorter half-life. Some radioactivity persisted beyond 600 days' decay. Since algae can transmit their radioactive toxicity right up through the nutritional pyramid to higher food animals, it is clear that more than conventional disposal methods must be devised for atomic wastes.

An indirect application of algal attributes relates to mosquito control. Matheson[249] noticed that mosquitoes did not breed in certain pools where *Chara fragilis* thrived; he and Hinman then postulated that the large amount of oxygen from the algae either interferes with the larval food supply or gives the mosquitoes indigestion. However, Biswas,[250] over a decade later, insisted that algal flora provide not only food for the mosquito, but also shelter!

b. *Animal Intoxications*

The microalgal scums, odors, and tastes described in the last section produce chiefly esthetic problems. Of far greater import are the many harmful reactions reported to have occurred in both animals and humans.[253-273] Freshwater phytoplankton has been implicated since 1878 in the intoxication and death of domesticated animals, as well as waterfowl, shorebirds, and their mammalian or avian predators.

In Table VII we present a chronological summation of 38 incidents of animal intoxications by phytoplankton as culled from the literature. In most cases attacks occurred after the animals had drunk from lakes or ponds containing heavy algal growth, usually during hot weather. They have variously been suspected of being due to botulism, anthrax, or worm infestations; or to poisons such as lead, arsenic, copper, cyanide, or alkali; or, as in South Africa, to gallsickness (carried by ticks), lamsiekte, and poisoning by plants such as senecio, gousiekte, and gifblaar. Also confused with algal poisoning has been the African geel dikkop of sheep, produced by photosensitizing toxins.

The reported symptomatology of algal intoxications has varied, but the most striking clinically have been the involvements of the neuromuscular and respiratory systems. As described by Francis,[253] in the Australian outbreak due to *Nodularia spumigeria,* "the animals developed stupor and unconsciousness, falling and remaining quiet, as if asleep unless touched, when convulsions came on, with the head and neck drawn back by rigid spasm which subsided before death." Similar manifestations have been reported from Alberta,[269] Manitoba,[262] North Dakota,[266] Minnesota,[254] Ontario,[268] and South Africa.[258-260] In several of these, the animals also exhibited extreme photosensitivity,[266] with severe blistering of the skin,[258-260] and also jaundice.[258]

Fitch[254] tested laboratory animals on the *Microcystis and Anabaena* which had killed cattle in Minnesota. The characteristic symptoms in guinea pigs included restlessness, incontinence, deep breathing, sneezing, coughing, salivation, lacrimation, weakness in the hind quarters, clonic spasms, and death. Rabbits presented the same picture plus opisthotonus. Pigeons also developed opisthotonus, as well as rapid blinking and swallowing.

Mason and Wheeler[270] injected *Microcystis aeruginosa* extract into mice, rats, guinea pigs, and cats. After a latent period of 20–180 minutes, there appeared pallor, hypotension, tachycardia, hypothermia, hyperglycemia, respiratory difficulty, and death. Steyn[258-260] in South Africa reported general paralysis or strychnine-like convulsions following consumption of large quantities of *Microcystis* by cows; smaller amounts resulted in constipation, drop in milk yield, generalized weakness, and severe photosensitivity of the skin. If the animals survived longer, many developed jaundice and ascites. Some of Steyn's laboratory animals manifested symptoms after simply being drenched with the contaminated water. Smit[271] injected or fed *Microcystis* to rabbits. In acute cases, the animals developed anemia, restlessness, dyspnea, progressive paralysis, coma, and death within one-half to four hours. In chronic cases, the outstanding finding was cirrhosis of the liver, with restlessness, atonia, and ascites. McLeod and Bondar[262] in Manitoba found that *Microcystis* administered to rats and mice caused a loss of equilibrium and progressive paralysis; later there were clonic muscular spasms, dyspnea, and cyanosis, then death within twenty hours.

The pathological findings in phytoplanktonic intoxications have been most interesting. Autopsies made in 1930 and 1933 in Minnesota showed "no gross pathology"[254]; cultures, smears, guinea-pig inoculations, and tests for copper and cyanide were also negative. On the other hand, Mason and Wheeler,[270] after injecting *Microcystis* extracts, observed markedly congested livers and dilated right hearts, but little generalized venous congestion. Their terminal clinical findings included severe anemia and reduction in total serum proteins. Small amounts of the toxic extract

TABLE VI.—*Animal Intoxications by Phytoplankton*

Source	*Year*	*Locale*	*Victims*	*Organism*
1. Francis	1878	Australia	sheep, horses, dogs, pigs	*Nodularia spumigeria*
2. Porter	1882–83	Minnesota	cattle, horses, hogs	Blue-green algae
3. Porter, Arthur, Stalker	1884	Minnesota	cattle	*Gloeotrichia echinulata*
4. Ballad	1900	Minnesota	cattle	*Aphanizomenon*
5. Cotton	1914	Michigan	steer and cow	*Anabaena*
6. Fitch	1918	Minnesota	sheep, hogs, chickens	*Coelosphaerium*, *Anabaena*
7. Howard, Berry	1924	Ontario	cattle	*Anabaena*
8. *Wilmot Enterprise*	1925	South Dakota	stock	Blue-green algae
9. Gillam	1925	Alberta	hogs	Blue-green algae
10. Gillam	1925	Alberta	hogs	Blue-green algae
11. Gillam	1925	Alberta	horses, cattle, hogs, poultry, wild birds	Blue-green algae
12. Woodcock	1925	Michigan	livestock	Blue-green algae
13. Steyn	1927	South Africa	sheep, cattle	Blue-green algae
14. Fitch	1930	Minnesota	cattle	*Microcystis*
15. Fitch	1930	Minnesota	pigs	Blue-green algae
16. Fitch	1933	Minnesota	sheep, lambs, chickens	*Microcystis*
17. Fitch	1933	Minnestoa	turkeys, ducks, geese, cows, pigs, horses	*Microcystis*, *Anabaena*, *Aphanizomenon*
18. Fitch	1933	Michigan	cattle	*Microcystis*, *Anabaena*
19. Hinderson	1933	Finland	stock	Blue-green algae

Source	*Year*	*Locale*	*Victims*	*Organism*
20. Deem, Thorpe	1939	Colorado	fish, birds, horses	*Anabaena, Microcystis*
21. McLeod	1938–40	Manitoba	animals	Blue-green algae
22. McLeod	1939	Alberta	animals	Blue-green algae
23. Steyn	1942	South Africa	horses, cattle, sheep, mules, hares, water birds	Blue-green algae
24. Louw	1942–43	South Africa	cattle, livestock	*Microcystis*
25. Quin (Remer)	1943	Montana	sheep	Blue-green algae
26. Ophel	1945	Australia	sheep, cattle	Blue-green algae
27. McLeod, Bondar	1945	Manitoba	horses, calves, pig, cat	Blue-green algae
28. Brandenburg	1946	North Dakota	cattle	Blue-green algae
29. Mullor, Wachs	1948	Argentina		
30. Prescott	1948	Bermuda	cattle	*Aphanizomenon*
31. McLeod	1949	Manitoba	animals	Blue-green algae
32. Stewart, Barnum, Henderson	1949	Ontario	cattle	*Microcystis, Anabaena*
33. Olson	1950	Canada	ducks	Blue-green algae
34. Olson	1948–51	Minnesota	livestock	Blue-green algae
35. Olson	1945–50	Manitoba	livestock	Blue-green algae
36. O'Donoghue	1950	Alberta	cow	*Microcystis*
37. O'Donoghue	1950	Alberta	pigs, turkeys, geese, dogs, chickens, horses, gulls, ducks, blue-finch	*Microcystis*
38. McLeod, Bondar	1951	Manitoba	horse, spaniels	*Aphanizomenon, Anabaena, Microcystis*

caused systolic arrest of the isolated perfused frog heart, but the uterus of the virgin pig was unaffected. Oxygen consumption of various rat tissue slices also remained unchanged. Steyn[258-260] reported marked blistering of photosensitized animal skins, as well as extensive involvement of the liver and the central nervous system. Post-mortem examinations showed the lungs full of blood; the spleen enlarged; and the liver engorged and dark red to black. In chronic cases, the liver was either yellow and soft, or dark and brittle. Ashworth and Mason[272] were able to produce generalized cellular damage by administering *Microcystis* extract to rats. Especially severe injury was noted in the parenchymal cells of the liver. Successive stages included acute parenchymatous, hydropic, and fatty degeneration, followed by central lobular necrosis. If death did not ensue, complete restoration of normal lobular architecture—without fibrosis—took place in thirty days. The toxic alterations were comparable to those produced by chloroform, phosphorus, and epidemic hepatitis. Similar acute hydropic and necrotic changes were also seen in the heart and kidneys, while microscopic hemorrhages and edema sometimes occurred in the lungs. Ashworth and Mason felt that rapid death was due to shock and circulatory collapse, while delayed demise was consequent to hepatic insufficiency, renal failure, and diffuse cellular damage. Stewart and his colleagues[268] reported fresh pulmonary hemorrhages and hepatic and cardiac lesions in Ontario cattle dying after ingestion of *Anabaena* and *Microcystis* scum. They found the same material lethal to pigeons and rats. Pathological findings from Alberta were "dilatation of the heart" and "mottled liver."

As mentioned earlier, many non-algal poisons have been wrongly suspected. In several cases, botulinus antitoxin was administered, to no avail. Solutions of sodium nitrite and sodium thiosulfate given intravenously assertedly produced recovery of some poisoned animals. Although the various outbreaks are now known to have been algal in origin, the specific nature of the toxins involved has certainly not been clear. Fitch[254] determined that the substances from *Microcystis* and *Anabaena* are non-volatile, heat-stable when dry, stable in solution at ordinary temperatures, but unstable when heated to 100°C. They are not electrolytes, alkaloids, or toxalbumins. They are organic componds of low molecular weight, somewhat resistant to acid and alkaline solutions. The substances apparently disappear when the algae putrefy. Mason and Wheeler[270] were able to inactivate *Microcystis* extracts partially by heating with acid or alkali. Steyn[260] clamied that growing algae are poisonous, that they discharge their poisons into the water when they die, that the decomposition of masses of dead algae decreases the toxicity, and that boiling the contaminated water does not reduce its toxicity. He thought that *Microcystis* contains two active principles: a potent fucoin-type hepato-neurotoxin, and a fluorescent photosensitizing pigment, phycocyan. Wheeler, Lackey, and Schott[273] determined that *Microcystis* is less poisonous when fresh than when frozen or dried. They found the toxin dialyzable, soluble in alcohol, and able to withstand autoclaving in neutral solution. It also survives, unfortunately, the laboratory equivalent of a water purification process—alum coagulation, chlorination, and filtration. It is adsorbable on carbon, but only when this is used in comparatively large amounts. According to McLeod and Bondar,[262] in a Manitoba outbreak associated chiefly with *Aphanizomenon* (and some *Microcystis* and *Anabaena*), the toxic material was in the plants and not in the water. They found the toxin stable in the plant, not destroyed by air drying at 37°C., by freezing, or by ultraviolet irradiation. Shelubsky[257] claimed in 1951 that the *Microcystis* toxin is alkali-labile. In contrast with other investigators, Louw[263] has reported isolation of two alkaloids from *Microcystis*. The first is an inactive one with a probable formula of $C_{10}H_{19}NO_2$. The other, without stated molecular structure, was isolated as a picrate with a melting point of 165°C. It is supposedly the active hepatotoxin, and it has been effective in experimental rats. Somewhat similar findings with marine algae from the South Pacific have recently been described by Habekost,[274] who isolated substances toxic to mice.

c. *Human Intoxications*

Algal poisoning has not been limited to animals. During the past 25 years, phytoplankton has also been incriminated in human reactions. These have presented as three general clinical pictures: dysenterial disorders, systemic allergic reactions, and local allergic eruptions. Table VIII summarizes the reported human cases.[275-281] (Poisonings from ingestion of plankton-nourished fish will be discussed separately later.)

The epidemic intestinal disorders of the early 1930's involved many thousands of people, in areas as widely separated as the National Parks in the Northwestern United States[275] and Charleston, West Virginia.[276] Symptomatology consisted of nausea, vomiting, diarrhea, and sometimes abdominal pain. It was self-limiting and usually resolved in a few days. These outbreaks were of no known specific etiology, but, significantly enough, were always concomitant with the presence of extensive algal blooms in the local water supply. Doubtless many other similar outbreaks have been neither studied nor reported.

TABLE VIII.—*Human Reactions Associated with Phytoplankton*

Author	*Year*	*No. Cases*	*Locale*	*Symptomatology*	*Etiology*
1. Spencer	1930	95	National Parks, Northwest U.S.	Nausea, vomiting, diarrhea, abdominal pain	algal bloom
2. Tisdale	1930	8,000–10,000	Charleston, West Virginia	Same	algal bloom
3. Olson	1930–31		Anacostia Reservoir area near Washington, D.C.	Dysentery-like epidemic	algal bloom
4. Heise	1934–35 1944	57 yr. old man	North Lake, Waukesha County, Wis.	Itching, conjunctivitis, blocked nares, bronchial astham	*Oscillatoraceae*
5. Heise	1945–46	39 yr. old woman	Lake Keesin, Waukesha County, Wis.	Swollen eyelids, blocked nose, generalized urticarial eruption	*Oscillatoraceae*
6. Sams	1947	67	Southern coast of Florida	Acute urticarial-papular "seabathers' eruption"	marine algae
7. Ayres	1948	1	Gulf of Mexico	Acute urticarial-papular eruption	"red tide"
8. Heise	1951	Office patients		Positive skin tests	*Microcystis*
9. Cohen	1953	6 yr. old child	Lake Carey, Pa.	Erythematous papulovesicular dermatitis of exposed areas	*Anabaena* (*phycocyanin*)

Heise[277] described two cases in which systemic hyperergic pictures appeared following swimming in a Wisconsin lake. In the one, the chief manifestations were itching, conjunctivitis, blocked nares, and bronchial asthma; in the other, swollen eyelids, nasal stuffiness, and a severe generalized urticarial eruption were the presenting findings. In both cases, *Oscillatoraceae* (Myxophyceae) were proven to be the causative organisms. Heise also obtained many positive skin reactions to *Microcystis* among his office patients.

Local allergic phenomena have been recorded more frequently of late. Sams[279] described an acute urticarial papular eruption—locally known as "seabathers' eruption"—in swimmers off the southern coast of Florida. Ayres,[280] in 1948, had a patient who swam in the Gulf of Mexico during the "red tide," then developed an acute urticarial papular eruption. And as recently as 1953, Cohen and Reif[281] reported phycocyanin, the blue pigment in *Anabaena,* as the cause of an erythematous papulo-vesicular contact dermatitis in a six-year old child who had bathed in a Pennsylvania lake.

Microscopic algae have also been assigned a more indirect role in some human disease states. Mariani[282] reported on his and Redaelli's work with *Prototheca portoricensis.* This alga, isolated from the stools of tropical sprue patients in San Domingo, produced a transitory local caseous granulomatosis in guinea pigs when injected intramuscularly or intraperitoneally. No specific sensitization or immunization could be achieved even with repeated injections. A related alga, *Blastocystis enterocola,* was isolated by Newiadomski from human, rat, and mouse feces, and was considered by him capable of passing through a Seitz filter. Mariani could not confirm the filtrability of *Blastocystis,* nor did he prove any pathogenicity. Neither could he corroborate Szendy's assertion that algae contributed to infections in metapneumothorax.

Although algae have been isolated from the skins of animals, none has yet been reported resident on the intact human epidermis.[283] However, in 1946, de Almeida and his colleagues[284] in Brazil isolated *Chlorella* from lesions of three patients with mycoses. The first had perirenal actinomycosis; the second, pulmonary and lingual paracoccidiosis (*brasiliensis*); and the third, lymphatic and dermal paracoccidiosis. The *Chlorella* was not considered a contaminant, but the authors debated whether it might be pathogenic either alone or in association with the fungi. They wondered, too, whether the pathogenicity of the fungi might not even have been lessened by the action of "chlorellin," an antibiotic substance isolated from

Chlorella. The activity of this material (effective against Gram positive and negative bacteria) has been shown to result from photo-oxidation and splitting of long carbon chains of the unsaturated fatty acids in *Chlorella.*[285] The antibacterial substance dubbed "phyceine" by Lesage[92] is probably similar to "chlorellin." Related antibiotic agents in *Stichococcus* and *Protosyphon* have been reported by Harder and Opperman.[286]

An interesting medico-legal application of planktonic distribution relates to the diagnosis of death by drowning.[287-290] Attempts have been made to prove death resulted from submersion by demonstrating algae in the lungs or the gastro-intestinal tract. It has even been proposed that the time and exact place of the drowning might be determined from the nature and type of algae present. It is clear that inherent faults exist in this method, since the algae may be difficult to identify, and since algal distribution in any area may vary from hour to hour.

A striking illustration of indirect algal effect is found in human intoxications resulting from ingestion of certain fish. Since 1530, there have been recorded many episodes of fish poisoning involving thousands of people.[291-301] Fifty-eight of the best documented have been summarized in Table IX (pages 57-59).

The most familiar intoxications are the acute "paralytic" shellfish poisonings resulting from eating mussels and clams. The manifestations are primarily neurologic: prickling and numbness of the mouth and fingers, ataxic gait, muscular incoordination, ascending paralysis and ultimately respiratory failure. Death may take place in two to twelve hours. Outbreaks have occurred in many parts of the world, predominantly during the summer months. For years there was considerable groping—superstitious, epidemiological and toxicological—for the underlying cause or causes. It was not until 1928 that Lindner[296] suggested they might reside in the food supply of the fish. This has indeed been proven since then, especially through the work of the groups at La Jolla and the Hooper Foundation.[292,294, 296,297] They showed that intoxications result only when shellfish ingest large quantities of the marine dinoflagellate *Gonyaulax* (Dinophyceae). These algae can grow so profusely during warm weather that the sea turns a deep rusty red during the day and brightly phosphorescent at night.

It has been demonstrated that the *Gonyaulax* contains an alkaloidal type of poison resembling in effect such substances as strychnine, muscarine, and aconitine. It is heat-stable in acid or neutral solution, but it is slowly destroyed by boiling with alkali. It is readily soluble in water and alcohol, but insoluble in ether or chloroform. One-millionth of a gram will kill a

TABLE IX.—*Human Poisoning Following Ingestion of Fish*

Author	*Year*	*Locale*	*Time of Year*	*Fish Implicated*	*No. Sick*	*No. Dead*
1. Martyr	1530	Virgin Is.		*Ciguatera* group	—	—
2. Kaempfer	1690	Japan		*Tetraodon* (puffer)	—	—
3. Anderson	1776	Central Pacific			—	—
4. Vancouver	1793	British Columbia		Mussels	3	1
5. Kraus et al.	1799	Peril Way, near Sitka, Alaska		Mussels	200	150
6. Combe	1827	Leith, Scotland	June	Mussels	30	2
7. British Medical Journal	1857	Victoria Dock, England	August	Mussels	(?)	1
8. Crumpe	1872	Tralee, Ireland		Mussels	3	3
9. Crumpe	1872	Tralee, Ireland		Mussels	1	—
10. British Medical Journal	1872	Liverpool, England	Oct.	Mussels	2	1
11. Meyer et al.	1870(?)	Mendocino, Calif.	August	Mussels	—	"Band" of Indians
12. Virchow & Wolff	1885	Wilhelmshaven, Germany	Oct.	Mussels	19	4
13. Schmidtmann	1887	Wilhelmshaven, Germany	Sept.	Mussels	3	1
14. Permewan	1888	Liverpool, England	August	Mussels	3	1
15. Cameron	1890	Dublin, Ireland	June	Mussels	7	5
16. Todd	1891		Sept.		1	—
17. Thesen	1901	Christiania, Norway	May	Mussels	5	2
18. Carr	1903(?)	Timber Grove, Calif.			12	5
19. Rolfe	1904	Avonmouth, England	June	Mussels	2	1
20. Netter & Ribadeau-Dumas	1907	Calais, France	May		13	2
21. Neale	1909	Barry, South Wales	August		19	1

TABLE IX (*continued*).—*Human Poisoning Following Ingestion of Fish*

Author	*Year*	*Locale*	*Time of Year*	*Fish Implicated*	*No. Sick*	*No. Dead*
22. Tahara et al.	1911(?)	Japan		Pufferfish (Fugu Poison)	—	—
23. Meyer et al.	1915	Santa Cruz, Calif.			4	0
24. California State Board of Health	1917	Santa Cruz, Calif.	July	Mussels	13	2
25. Kofoid & Vaughan	1918(?)	La Jolla, Calif.	September	Mussels	1	0
26. Olson	1924	Koenigsberg Haff, East Prussia, Germany	Winter Autumn	Eels & Fish Livers	800 appr.	6
27. Sommer & Meyer	1925(?)	Subtropical American Waters		Local Fish (*Ciguatera*)	—	—
28. Sommer & Meyer	1927	Central Calif. Coast	July	Mussels	102	6
29. Sommer & Meyer	1929	Central Calif. Coast	Summer	Mussels & Clams	56	1
30. Sommer & Meyer	1930	Central Calif. Coast	July	Mussels	2	—
31. Sommer & Meyer	1932	Central Calif. Coast	June & July	Mussels & Clams	37	1
32. Olson	1932–33	Koenigsberg Haff, East Prussia, Germany		Fish or Fish Livers	125	—
33. Sommer & Meyer	1933–34	Gokasho Bay, Toba, Japan	Nov., Dec., Jan., Feb.			
34. Sommer & Meyer	1933	Central & North Calif. & Oregon	July & September	Mussels	19	1
35. Sommer & Meyer	1933	Oregon	October	Mussels & Clams	3	—
36. Sommer & Meyer	1934	Alaska	July	Mussels	12	2
37. Sommer & Meyer	1936	Southern Calif. Coast	May	Mussels	3	2
38 Murphy	1936	Bay of Fundy, Nova Scotia	July	Mussels	5	2
39. Halstead	1938			Amberjack	1	—

Author	*Year*	*Locale*	*Time of Year*	*Fish Implicated*	*No. Sick*	*No. Dead*
40. Wolczynska & Conrad	1939	Bruges-Zeebrugge Canal Belgium	Summer		—	—
41. Olson	1939	Calif. Coast			—	—
42. Olson	1940	Lake Ysmen, Sweden		Eels and other fish livers	Small epidemic	—
43. Halstead	1941–45	Micronesia			400	(?)
44. Lively	1944	Gilbert Is., Pacific	September	Red Snapper	4	—
45. Lewis	1944	Line Is., Pacific	August	Red Snapper	6	—
46. Halstead	1944	Honolulu, Hawaii, Midway & Christmas Is.	November & December	Sea Bass	38	—
47. Von Fraenkel & Krick	1945	Mariana Is., Pacific		Barracuda	30	—
48. Medcoff	1945	New Brunswick, Canada		Mussels	28	—
49. McCann	1945–46	Palmyra Islands, Pacific		Red Snapper	32	14
50. Cohen et al.	1946	Mariana Is., Pacific			51	—
51. Ross	1946–47	Fanning Is., Pacific		Fish—type not stated	95	—
52. Medcoff	1948	Le Boules, Quebec, St. Lawrence River, Can.			2	2
53. Sapeika	1948	Capetown, South Africa		Mussels	Many	1
54. Olson	1948	Lake Ysmen, Sweden		Eels and other fish livers	11	—
55. Halstead	1949	Saipan, Pacific	May	Black Moray Eel	57	2
56. Martin	1950–51	Johnston Is., Pacific	Summer		20	—
57. Halstead	1953	Marshall Is., Pacific	March	Black Moray Eel	6	1
58. Tennant et al.	1954	Metis Beach, St. Lawrence River, Can.		Mussels	7	2

mouse on injection, while a few milligrams orally can be fatal to a human being, especially when taken on an empty stomach. It has been estimated that 4,000 algal organisms are required to produce human toxicity. The shellfish store the poison chiefly in their "livers" or digestive organs; thus the "dark meat" of the infested bivalves is much more toxic than the "white" portions. The fish themselves are unaffected by the toxin, and they will in fact excrete it in several weeks if *Gonyaulax* disappears from the marine diet.[297,298,302,303]

Post-mortem examinations of animals and human beings who have died of acute paralytic shellfish poisoning have, surprisingly enough, revealed no structural changes. However, in chronically poisoned experimental animals, distinct alterations are noted in the medullary ganglion cells and in the large cells of the ventral horn of the spinal cord.[302] The Golgi apparatus of small and medium-sized spinal ganglionic cells is also damaged. Neural mitochondria remain normal, but those in the convoluted tubules of the kidney show definite damage. In concert with the latter finding are the clinical nephropathies reported by the French in human shellfish poisoning.[304,305]

It is probable that other algae besides *Gonyaulax* contain toxins. Some have been associated with *Ceratium* and *Prorocentrum,* but no direct extraction or isolation has yet been accomplished. It is also quite logical to expect that fish other than mussels and clams could have improper planktonic relations and thus transmit their toxins to predators. This is indeed so, and—except for the puffer fish and possibly barracuda—it is likely that poisonousness is directly dependent upon periodic ingestion of noxious plankton, rather than being due to inherent toxicity. Planktogenic fish toxicity can occur during any time of the year, while puffer and barracuda may be most lethal during the reproductive season. Epidemics have been unrelated to spoilage or bacterial infection.

Poisonous fish are found through the world. They are most numerous in the warm seas (especially in the Caribbean and Central and South Pacific), but even certain arctic sharks have been reported to be toxic. During World War II, fish poisoning represented quite a problem, since epidemics were encountered wherever American forces were stationed in the Pacific theater. The Japanese, too, estimated that over 400 of their military men died in Micronesia from eating toxic fish. Halstead and Lively[300] have made a partial listing of fish most often found to be poisonous: surgeonfish, triggerfish, pompano, porcupine fish, wrasse, snapper, filefish, surmullet or goatfish, moray eel, parrot fish, sea bass or grouper, barracuda, and puffer. They have divided fish poisoning into four clinical groups, the last three of which are probably planktogenic:

1. *Tetraodon* (puffer) poisoning
2. *Gymnothorax* (Moray eel) poisoning
3. *Ciguatera*
4. *Scombroid* poisoning

Tetraodontoxin is the only ichthyosarcotoxin about which anything specific is known. Japanese studies indicate it is a white hygroscopic powder, soluble in water but not in the ordinary organic solvents. Although it is thought to have a formula of $C_{16}H_{31}NO_{16}$, little else is known except that it is not an alkaloid or a protein or a protamine. It is a most potent poison, with symptoms occurring within thirty minutes of ingestion. Initially there are numbness and tingling of the face and extremities, then nausea, vomiting, headache, dizziness and overwhelming weakness. Next come speech impairment, dyspnea, and generalized muscular paralysis. Death from respiratory paralysis may occur in 1–24 hours. The mortality rate is about 60 per cent.

In *Gymnothorax* poisoning, similar neurotoxic manifestations are seen at first, but then the patient develops motor incoordination and violent convulsions, laryngeal spasm, coma, and respiratory paralysis. Here the mortality rate is only about 10 per cent, and, in the non-fatal cases, major symtomatology disappears in about ten days. With *Ciguatera* (originally considered only Caribbean), the attacks are milder than with *Gymnothorax* and usually come on more slowly. The chief symptoms include sensory disturbances, myalgias, arthralgias, and severe weakness. Only 2–3 per cent of the victims die, but recovery may take many months. *Scombroid* poisoning results from eating various tropical tuna-type fish. The manifestations are mostly histamine-like: headache, flushing of the face, conjunctivitis, giant hives, erythema, and gastric upset. Recovery usually takes place in 8–12 hours.

Haffkrankheit—erroneously designated "Haff's Disease" by some enthusiastic eponymist—was first described in 1924[295] among fishermen in Koenigsberg Haff ("harbor") in East Prussia. It follows contact or ingestion of fish (primarily eels) and fish livers. It is characterized by a sudden onset of weakness, severe myalgia, myoglobinuria, and some digestive upset. Pathological findings include fatty degeneration of the liver and kidneys, and damage to the anterior horn cells of the spinal cord. Over 1,000 people have been reported afflicted, some fatally. Similar episodes have been recorded at Lake Ysmen in Sweden. Originally Haffkrankheit was considered to be caused by arsines in sewage. However, the correlation of outbreaks with climatic changes, and the predominantly neurotoxic nature of the manifestations, suggests that this disease, like the several other categories of fish poisoning, is dependent upon periodic planktonic intoxication.

No method of testing suspected fish is known, other than feeding it to animals (kittens have been a favorite). Neither does any specific antidote exist. Treatment has consisted primarily of rapidly emptying the gastrointestinal tract by mechanical means, apomorphine, and laxatives. Administration of charcoal as an adsorbent may be helpful. Also important are supportive measures such as stimulants, parenteral fluids, oxygen, and artificial respiration. Intubation and tracheotomy may be necessary with respiratory distress. Anti-convulsants and opiates may have their indications, while some assign a special merit to intravenous calcium gluconate. To us it would seem logical also to employ the adrenal steroids, either to combat anaphylactoid reactions or to diminish end-organ response to toxins. Anti-histamine agents, too, might play a part in treating the *Scombroid* type of poisoning. Doubtless the wisest management of fish poisoning consists of avoiding those fish with a known toxic potential; and, in the case of shellfish, avoiding them when the *Gonyaulax* or other suspected algae are flourishing.

V. DISCUSSION

After combing thousands of articles on algae and plankton, we are in firm agreement with Conway Zirkle,[306] who laments the fact that our age of specialization has produced a "splintered learning." He points out that many important developments are hopelessly buried because of the compartmentalization of knowledge, the specialized jargons of the various sciences, and the overwhelming volume of new publications. Because of a limited lateral diffusion of information, many workers remain unaware of pertinent findings even in kindred fields. This is especially true in the realm of medicine. With the tremendous mass of studies that physicians must continually digest, their meager knowledge of algae and plankton is readily understandable. The rare medical articles on these subjects have usually dealt only with some special point of interest—or a courtesy reference has appeared in a botanical, nutritional, or public health journal. The present assemblage of data brings into focus much diversified material, a good portion of which deserves closer integration into the field of medicine.

Interest in algae and plankton has varied over the years. From ancient times, the predominant emphasis was on the seaweeds; at the turn of this century, it shifted to zooplankton; and, since World War II, it has centered chiefly about phytoplankton. The traditional medicinal use of seaweed has declined sharply, as has its role as a human nutrient. Its value today resides primarily in the contained minerals and vitamins, and its greatest utility is as fertilizer and animal provender. There is also much merit in the alginates, and a good potential exists for other seaweed derivatives such as the antibiotics and anticoagulants. Consideration of plankton has for the most part been focused on its use as food.[307-309] Zooplankton is nutritionally far richer than seaweed, but it does not possess the significance its more enthusiastic proponents confer upon it. Despite the fact that the Thais[21] have reportedly collected 5,000 tons in one year, it should not be forgotten that there are tremendous variations in quantity and quality of yield in different oceans and at different seasons. While zooplankton may be considered an excellent emergency ration and food supplement, its extensive use as a food staple awaits the development of far more efficient collection techniques.

Since it is still much too early to envision extensive *in vitro* photosynthesis[197] without the intermediary of plants, it is the phytoplanktonic organisms which offer the most exciting prospect for nutritional gain. They not only supply controllable proportions of protein, fat, vitamins, minerals, and calories—they can also be cultured selectively on a large-scale basis. The marvelous adaptability of algae like *Chlorella* gives man another round in the crucial Malthusian battle of population versus food supply. In the past three centuries, the world population has risen from a half billion to two and one-half billion.[25] If fission and fusion do not permanently put an end to both mankind and its problems, the population will be four billion in 1980, and eight billion in 2050. Selective fresh-water mass culture of organisms (we suggest the term "aquaculture" as more descriptive than "hydroponics" and less restrictive than "mariculture"[29]) like *Chlorella* in specially built plants hold great promise for large quantities of valuable foodstuffs. It is more practical than "manuring" the sea with salts and minerals—"three-dimensional farming"—to increase the growth of phytoplankton, whether one hopes to harvest the phytoplankton as such, or whether it is planned to support a greater fish population. The latter is, in fact, a wasteful process, since it has been calculated that a pound of codfish ultimately represents 100,000 pounds of marine phytoplankton![7] Before phytoplanktonic feeding becomes commonplace procedure, there will have to be long-term human experiments, with careful observation of possible toxicities and biochemical changes, especially with respect to electrolytes, hemic elements, and proteins. It is also a certainty that a re-education of food tastes will have to take place before the acceptability of planktonic foods reaches a high level.[310]

Some other applications of phytoplankton reach almost into the realm of science fiction. Bassham,[311] at the University of California, has been working for the U.S. Navy on adapting microscopic algal cultures to control the oxygen and carbon dioxide in atomic submarines which would be submerged for long periods. It has been calculated that one kg. fresh weight of *Chlorella* (equal to 100 liters of growing algal suspension) can easily supply the 25 liters per hour oxygen required by a 70 kg. man, as well as utilize the carbon dioxide exhaled by him. In a similar fashion, the Department of Space Medicine of the Air Force[22] is considering using algae in space ships, with the intriguing extra notion of complete recycling of biological elements: the algae are also to be used as food by the spacemen, while their excreta will serve as nutriment for the algae!

From the strictly medical standpoint, the number of disease syndromes attributable to algae comes rather as a surprise and raises some interesting questions. The most obvious afflictions are the allergic dermatitides resulting from bathing in water contaminated with algae. Also understandable are the respiratory irritations from water-borne or inhaled algae; in fact, many cases of allergic rhinitis (and its corollary, chronic sinusitis) in coastal areas might be traceable to algae instead of being blamed on "damp climate" or that handy favorite of allergists, "dust." Then there are the nephropathies, and the striking epidemic gastroenteritides, so blandly ascribed to the "24-hour or 48-hour virus."

By all odds, the most dramatic algal intoxications are the acute neurotoxic and myotoxic ones typified by paralytic shellfish poisoning and some other ichthyosarcotoxicoses. Although pitifully meager long-range studies have been made, it seems incredible that such potent toxins should not produce related chronic sequelae in victims who have survived acute attacks, or who have ingested smaller amounts of toxin over prolonged periods. One cannot help but wonder whether chronic algal intoxication may not play a part in causing some of our idiopathic neuropathies such as multiple sclerosis, the muscular dystrophies, and amyotrophic lateral sclerosis. Interestingly enough, very recently a U.S. Public Health Service team has reported[312] an extensive survey of amyotrophic lateral sclerosis among the Chamorros of Guam and other islands of the Mariana group. They made the study because the incidence of the disease there is 420 per 100,000 population, a rate 100 times higher than in the rest of the world; at any one time, one per cent of the adults are affected, and eight to ten per cent of adult deaths are produced by it. No cause was discovered, except perhaps some tenuous genetic relationship to the Chamorro tribe. The investigators have apparently given little consideration to the water contacts or fish-eating habits of these people. It may be more than coincidence that epidemics of fish poisoning—likely due to toxic phytoplankton—have been particularly plentiful in the Marianas. That algae have not been implicated in the nerve afflictions may be due to the difficulty in demonstrating them, or simply to their not even having been considered.

It is interesting, if hazardous, to speculate also on a possible algal role in another neurologic disease—acute poliomyelitis. The viral etiology is certainly proven beyond any doubt, yet the epidemiology is anything but clear. Is it possible that a symbiotic virus-alga state exists, favoring viral transport or even viral multiplaction? The thought is suggested by the fact that the highest incidence of poliomyelitis is during the warm summer months, when algal blooms are most prolific; also, that infection occurs frequently following swimming. In the latter case, the victim, besides ingesting virus, might also decrease local tissue resistance by contact with algal toxins. However, we are not yet prepared to suggest *Chlorella* instead of monkey kidney as a medium for culturing polio virus!

A more definite chronic entity is "swayback,"[82,83] the neurologic disorder seen in lambs fed on seaweed. Whether a toxin disturbing copper metabolism resides in the seaweed itself, or whether it originates in contaminating microscopic algae, has not been established. Also worth considering are the hepatitis and cirrhosis seen in animals suffering from "waterbloom" toxicity. Their occurrence raises the question of whether all human hepatitis classified as "viral" is really so. Certainly these various diseases indicate that much might be gained from studying the results of controlled administration of toxic algae to animals.

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