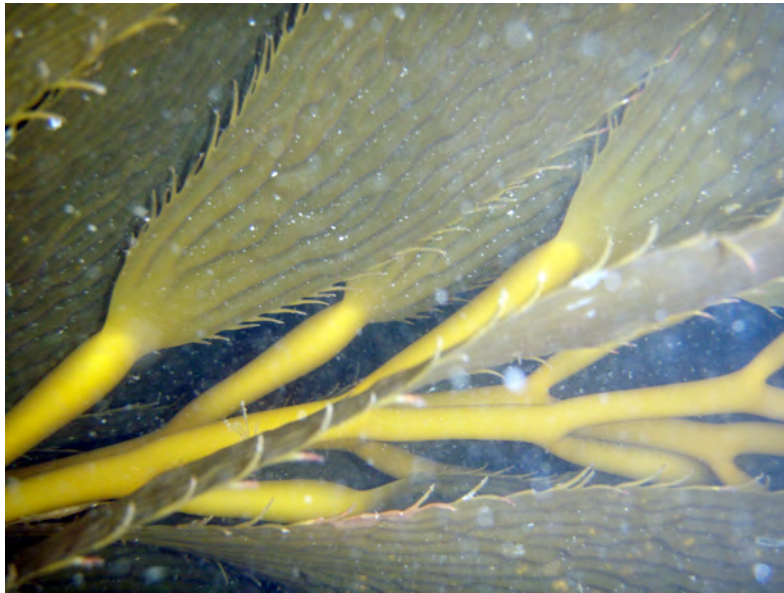


March 31, 2008

Techno-Economic Feasibility Analysis of Offshore Seaweed Farming for Bioenergy and Biobased Products

Independent Research and Development Report
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California kelp macroalgae seaweed underwater (Copyright: Jane Thomas, IAN, UMCES)

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ABSTRACT

The purpose of this study is to provide an initial assessment of the technical and economic feasibility of cultivating seaweed offshore to produce biofuels. This report reviews the seaweed industry and the higher value products that could improve the economic attractiveness of seaweed biofuel production process. We review previous attempts at offshore seaweed culture for biofuels, the technical and economic challenges faced by those projects, and the lessons learned. Progress in offshore seaweed farming technology is also examined.

We propose a concept for offshore seaweed cultivation that positions large seaweed farms in natural nutrient upwelling areas. This concept greatly simplifies prior proposals based on artificial upwelling of deep ocean waters for nutrient supply. We conclude with a technology road map that recommends future activities to move offshore seaweed culture from the present concept and vision to a future commercial reality.

For the context of this report, “offshore” or “open ocean” growing conditions refers to growing seaweed in waters that are generally too deep for even giant kelp to survive on their own and that are free from the direct influence of land. Nearshore refers to habitats of sufficiently shallow depth to enable such seaweeds to attach and grow or which provide a sheltered environment for aquaculture operations.

This report documents the long history of using seaweeds to meet human needs. The economic value of seaweeds worldwide is currently about \$6 billion USD, primarily as food products, and also as hydrocolloids for the food and pharmaceutical industry, soil conditioners, animal feeds, and cosmetics. The total seaweed harvest is reported at 15.7 million metric ton wet weight (about one million ton dry weight) per year, of which almost 90% is produced by nearshore aquaculture production. Thus, seaweed farming is already a significant industry, with a sophisticated technological basis, ranging from the biotechnology to aquaculture, processing, and marketing of the many products derived from these plants.

As the need for renewable energy continues to grow, seaweed farming has the potential to help meet future energy needs. The oceans cover over 70% of the Earth’s surface. Use of just 1% of that along the ocean margins could supply about 3.5 billion dry ton of new biomass annually, if the production rates already achieved in coastal seaweed farms in countries like China could be projected for open ocean systems. This is three-times the maximum amount of terrestrial biomass that can be reasonably collected annually in the U.S. Such systems would not compete

with the availability of fresh water, land, and nutrients needed to sustain terrestrial agriculture.

Large-scale open ocean seaweed farming for biofuels production was attempted in the 1970s and 1980s, but was not technically successful. However, the lessons learned from that earlier attempt, together with advances in open ocean engineering and the current energy economics, provide the basis and incentive to develop a novel approach to open ocean farming. Indeed, exploratory R&D activities in Japan, Korea, Denmark, Germany, and the United Kingdom, among others, are already pioneering new efforts in this area. Large-scale open ocean farming could be used to produce the next generation biofuels, in particular butanol, for which historical precedence exists, and also to increase the supply of higher value animal feeds and bioproducts.

The technical and economic viability of seaweed biomass production for conversion into biofuels requires an understanding of the factors that limit their growth in nature and under managed aquaculture operations, the evaluation of processes for converting the biomass into biofuels, and a determination of the risk factors in a seaweed-to-energy pathway. As noted above, we propose a concept for offshore seaweed cultivation, which we call the “Offshore Seaweed Farm”. This would be based on one-km² (100 hectare) dynamically positioned floating seaweed production platforms. A Marine Biorefinery would take the seaweed biomass and process it into biofuels and other products.

Recommendations

The following activities are identified as the first-order questions that need to be answered to allow this field to move forward, and thus merit attention in the near term:

- Identify appropriate seaweed species for open ocean culture and demonstrate optimized growth characteristics under simulated onshore conditions consistent with cultivation in open ocean culture environments.
- Initiate studies on bioprocessing to optimize production of desired products such as animal feeds from digester residue and, for renewable biofuels, a focus on butanol production.
- Conduct an updated economic assessment that includes the economics of seaweed products and growing seaweeds in offshore farms.

In conclusion, macroalgae, i.e., seaweeds, represent an unrealized biomass potential to meet future societal needs for renewable energy and biobased products.

CHAPTER 1

BIOLOGY OF SEaweEDS

General biology of seaweeds

Marine plants are generally divided into three groups: microalgae that can occur as phytoplankton in the open ocean and as benthic or sediment-dwelling forms; macroalgae, also known as seaweeds, which are multi-cellular plants that generally anchor to hard surfaces; and rooted plants, or angiosperms that include seagrasses, with roots in soft substrates that deliver nutrition to the plants. Micro- and macro-algae are continuously washed with seawater and gain their nutrients directly from the water (Mann, 1973). All marine plants need a carbon source, dissolved nutrients, including nitrogen and phosphate compounds, trace elements, and other compounds from the seawater plus sunlight to grow and thrive. The growth of marine plants is generally controlled by the availability of sunlight and nutrients; the lack of one or another will limit the rate of growth, and production of biomass.

The general structure of a typical seaweed such as brown alga like kelp is composed of the leafy blade or lamina (also referred to as the frond), the stem-like stipe, and the holdfast that anchors the plant to a hard substrate (Figure 1). The entire plant is referred to as the thallus.

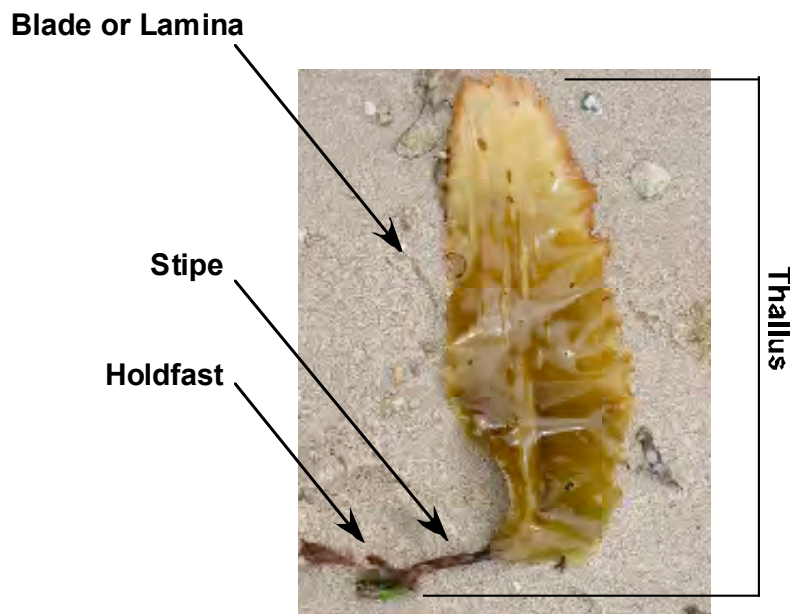


Figure 1. A small kelp plant with major structures identified.

There are three major groups of seaweeds: chlorophytes or green algae, rhodophytes or red algae, and phaeophytes or brown algae. The brown algae, in particular the larger species in the Order Laminariales, are generally referred to as kelp. These are of great interest in the present report. Seaweeds are also referred to as macroalgae, to differentiate them from the microalgae, though there is a close relationship between the two types. Each group of seaweeds has a different evolutionary history, characterized by distinguishing features that include photosynthetic pigments, cellular storage materials, reproductive strategies, life cycles and natural habitats. These factors play a role in determining the ease with which each species within an algal group can be raised, the resiliency to physical and chemical stress, and resistance to disease of each seaweed, and the nature of the energy source and additional products that may be generated (McHugh 2003). There are, of course, major differences between species within groups.

Because most seaweeds require a hard substrate to anchor their holdfast, their growth is restricted to shallow coastal waters, or areas where an artificial hard surface can be provided. Exceptions occur in that some are free floating. Most green algae are small and delicate, and only a few species have any significant commercial value. Of the roughly 20,000 known marine macroalgal species, 221 are of commercial importance (Zemke-White & Ohno, 1999). Cultured seaweed accounted for over 52% of worldwide seaweed production in dry ton, and four genera representing species of *Porphyra*, *Laminaria*, *Gracilariaria*, and *Undaria* comprised 93% of production from cultured seaweeds (Zemke-White & Ohno, 1999).

Seaweed life cycles

Seaweed life cycles are complex in many species, with annual and perennial species and sexual and asexual reproductive modes, resulting in isomorphic or heteromorphic life history forms, commonly referred to as alternation of generations. Understanding the complex and diverse life cycles of different seaweeds is of practical significance in controlling growth and reproduction for optimal plant husbandry. An example in which our increased understanding of life cycles had clear economic impact was the identification of the conchocelis, originally considered a separate organism, as a one of the diploid stages of *Porphyra* spp. (Drew, 1949; Drew, 1954). This discovery revolutionized the culture of a genus that includes one of the most important commercially cultivated seaweeds in Japan, China, and Korea. The conchocelis

became the seed stock source for artificial propagation of this seaweed (Choi *et al.*, 2002). An understanding of life cycles facilitates improvements in cultivation practices and strain selection for desirable traits such as faster growth, resistance to environmental factors, and enhancing economically important properties of seaweed-derived products.

Seaweeds, like many plants can reproduce both asexually, by simply dividing vegetative parts to produce new plants and sexually, thus strengthening the gene pool.

Algal life cycles can generally be categorized as gametic, zygotic, and biphasic (Bold & Wynne, 1985). Gametic (also called diplontic) life cycles are characterized by diploid adults and haploid gametes. Zygotic (also called haplontic) life cycles are characterized by haploid gametophytes that produce haploid gametes and diploid zygotes that arise from union of two haploid gametes and undergo meiosis to produce haploid spores that grow into new gametophytes. Biphasic life cycles are characterized by cycling between separate, free-living, and independent haploid gametophyte and diploid sporophyte phases as depicted in Figure 2 (Lee, 1999). The species with biphasic life cycles display alternation of generations with dominant forms that are either haploid or diploid and isomorphic or heteromorphic.

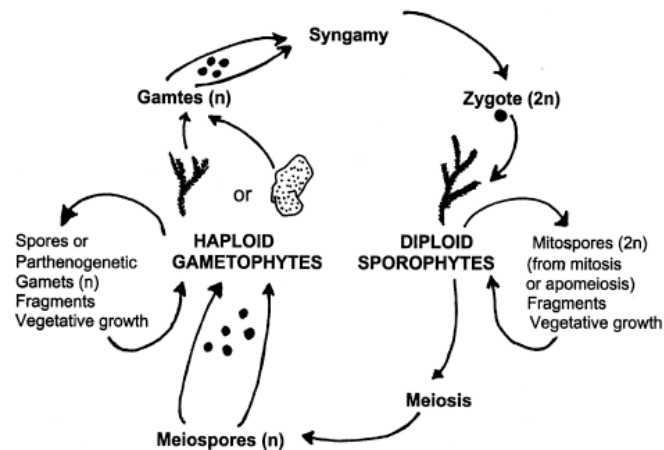


Figure 2. Generic representation of alternating life cycle of seaweeds (Collado-Vides, 2001).

Most red algae have a modified biphasic life cycle, wherein the zygote differentiates into a short-lived diploid carposporophyte that is attached to the female gametophyte and quickly divides to form carpospores (Thornber, 2006). These are released to the water column, settle, and develop

into the sporophyte. While these life cycles are representative of the different reproductive strategies associated with the major groupings of macroalgae, i.e., the red, brown, and green seaweeds, whether the macrophyte form is haploid or diploid is genus specific.

Also of practical significance are the modifications to the stereotypical life cycles described above, which result in the diversity of reproductive strategies seen in the seaweeds. Of note is the asexual looping (Thornber, 2006) that enables gametophytes to produce new gametophytes, bypassing the sporophyte stage, and sporophytes that directly produce new sporophytes, bypassing the gametophyte. Thus, in *Porphyra* spp. for example, the blades and conchocelis can replicate asexually through the formation of spores (Li, 1984; Nelson *et al.*, 1999; Nelson & Knight, 1995) that can germinate and develop directly into more blades or conchocelis' as a modification of the life cycle. Clonal growth, which can arise through various processes depicted in Figure 3, also occurs in the red, brown, and green algae. This diversity in asexual modes of reproduction provides options that can be exploited in culturing practices used by the seaweed culture industry. Clonal seaweeds, for example, are capable of regrowing from thallus fragments, a feature that is heavily used in the seaweed industry for propagation of some species.

The characteristics associated with both sexual and asexual reproduction in the seaweeds, thus, provide the basis for current cultivation practices. Advances in understanding seaweed life cycles and reproductive strategies are currently contributing to improvement of cultivation practices and strain selection.

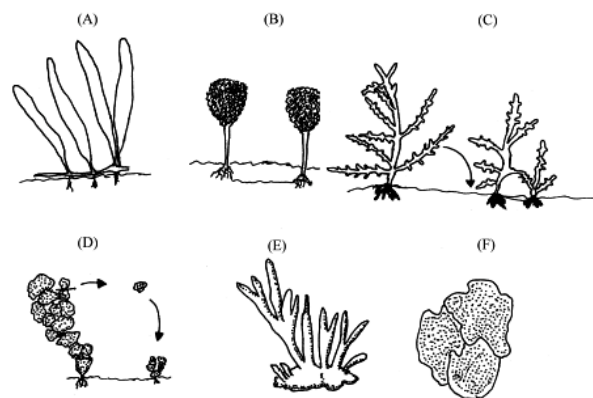


Figure 3. Variation in clonal in seaweeds. (A) stoloniferous growth, (B) two thalli from a buried stolon, (C) new growth from attachment of branch to bottom, (D) fragmentation, (E) fronds growing from a holdfast, (F) crustose growth (Collado-Vides, 2001).

CHAPTER 2

THE SEAWEED INDUSTRY

Worldwide production of seaweeds

There is no single source for reporting worldwide harvest and culture of seaweeds. Estimates can be gleaned from global aquaculture yields, and extrapolations can be made by assigning conversion ratios for biomass and weight. Challenges in accurately calculating the current production of seaweed include inconsistent reporting of yields as wet and dry weight, as well as a lack of clarity of species identification in reporting.

The total global production of all aquaculture products in 2004 was 59.4 million metric ton with a total value of \$70.3 billion (Chen, 2006). Of this almost a quarter by weight, but only a tenth by value (\$6.8 billion) were aquatic plants; 99.8% of which were farmed in Asia and the Pacific Region. Seaweed farms worldwide are estimated to produce 13.9 million metric ton wet weight per year (Table 1). Wild seaweed harvest is about 1.8 million metric ton of wet weight plants per year [based on 300,000 metric ton dry weight and 85% moisture (www.surialink.com)]. Most of the wild harvest is brown seaweeds harvested for production of marine colloids (mostly alginates) (Figure 4). An additional 800,000 metric ton per year are coralline red seaweeds (e.g. *Lithothamnion corraloides*), known as ‘Maerl’, and harvested as dead calcareous skeletons dredged from European waters and used as soil conditioners.

Production data by country is provided Table 2 for four genera of brown seaweeds, three genera (and an unspecified category) of red seaweed, which together represent most of the seaweed produced in the world. Six Asian countries (China, Japan, South Korea, North Korea, Philippines, and Indonesia) produce almost 99% of the world’s farmed seaweed. A more detailed examination of Chinese seaweed culture is warranted, as that nation is the dominant force in the industry, as it is in most types of aquaculture.

Traditional uses of seaweed products

Seaweeds have been gathered for centuries for food and for the chemicals they contain, many of which are produced at industrial scale today (Neushul, 1987). Brown seaweeds are grown for both human consumption and extraction of marine colloids, while red seaweeds, with

Table 1. FAO figures for world farmed seaweed and *Spirulina* production in 2004*Courtesy R. Subasinghe, FAO*

Country	Common name	Taxa	Metric ton
Bulgaria	Chlorella- unicellular green alga	<i>Chlorella vulgaris</i>	...
Cambodia	Aquatic plants nei	<i>Plantae aquaticae</i> ¹	16,840
Chile	Gracilaria seaweeds	<i>Gracilaria</i> spp.	19,714
China	Aquatic plants nei	<i>Plantae aquaticae</i>	3,230
China	Aquatic plants nei	<i>Plantae aquaticae</i>	2,535,130
China	Dark green nori	<i>Enteromorpha prolifera</i>	3,280
China	Euचेuma seaweeds nei	<i>Euचेuma</i> spp.	97,820
China	Fusiform sargassum	<i>Sargassum fusiforme</i>	131,680
China	Japanese isinglass	<i>Gelidium amansii</i>	1,150
China	Japanese kelp	<i>Laminaria japonica</i>	4,005,640
China	Laver (Nori)	<i>Porphyra tenera</i>	810,170
China	Spirulina nei	<i>Spirulina</i> spp.	41,570
China	Wakame	<i>Undaria pinnatifida</i>	2,196,070
China	Warty gracilaria	<i>Gracilaria verrucosa</i>	888,870
Fiji Islands	Euचेuma seaweeds nei	<i>Euचेuma</i> spp.	45
France	Harpoon seaweeds	<i>Asparagopsis</i> spp.	12
France	Wakame nei	<i>Undaria</i> spp.	25
Indonesia	Red seaweeds	<i>Rhodophyceae</i>	12,606
Indonesia	Red seaweeds	<i>Rhodophyceae</i>	397,964
Italy	Gracilaria seaweeds	<i>Gracilaria</i> spp.	...
Japan	Aquatic plants nei	<i>Plantae aquaticae</i>	15,968
Japan	Green seaweeds	<i>Chlorophyceae</i>	...
Japan	Japanese kelp	<i>Laminaria japonica</i>	47,256
Japan	Laver (Nori)	<i>Porphyra tenera</i>	358,929
Japan	Wakame	<i>Undaria pinnatifida</i>	62,236
Kiribati	Euचेuma seaweeds nei	<i>Euचेuma</i> spp.	3,904
Korea, Dem. People's Rep	Gelidium seaweeds	<i>Gelidium</i> spp.	...
Korea, Dem. People's Rep	Gracilaria seaweeds	<i>Gracilaria</i> spp.	...
Korea, Dem. People's Rep	Japanese kelp	<i>Laminaria japonica</i>	444,295
Korea, Dem. People's Rep	Laver (Nori)	<i>Porphyra tenera</i>	...
Korea, Dem. People's Rep	Wakame	<i>Undaria pinnatifida</i>	...
Korea, Republic of	Aquatic plants nei	<i>Plantae aquaticae</i>	...
Korea, Republic of	Aquatic plants nei	<i>Plantae aquaticae</i>	142
Korea, Republic of	Brown seaweeds	<i>Phaeophyceae</i>	22,814
Korea, Republic of	Green laver	<i>Monostroma nitidum</i>	11,514
Korea, Republic of	Japanese kelp	<i>Laminaria japonica</i>	22,510

Korea, Republic of	Laver (Nori)	<i>Porphyra tenera</i>	228,554
Korea, Republic of	Wakame	<i>Undaria pinnatifida</i>	261,574
Madagascar	Aquatic plants nei	<i>Plantae aquaticae</i>	...
Malaysia	Aquatic plants nei	<i>Plantae aquaticae</i>	30,957
Mali	Aquatic plants nei	<i>Plantae aquaticae</i>	90
Micronesia, Fed.States of	Eucheuma seaweeds nei	<i>Eucheuma</i> spp.	0
Mozambique	Elkhorn sea moss	<i>Kappaphycus alvarezii</i>	92
Mozambique	Spiny eucheuma	<i>Eucheuma denticulatum</i>	...
Namibia	Gracilaria seaweeds	<i>Gracilaria</i> spp.	67
Peru	Gracilaria seaweeds	<i>Gracilaria</i> spp.	...
Philippines	Caulerpa seaweeds	<i>Caulerpa</i> spp.	4,252
Philippines	Elkhorn sea moss	<i>Kappaphycus alvarezii</i>	44,814
Philippines	Gracilaria seaweeds	<i>Gracilaria</i> spp.	389
Philippines	Spiny eucheuma	<i>Eucheuma denticulatum</i>	85,754
Philippines	Zanzibar weed	<i>Eucheuma cottonii</i>	1,069,599
Russian Federation	Brown seaweeds	<i>Phaeophyceae</i>	...
Russian Federation	Brown seaweeds	<i>Phaeophyceae</i>	216
Saint Lucia	Eucheuma seaweeds nei	<i>Eucheuma</i> spp.	1
Saint Lucia	Gracilaria seaweeds	<i>Gracilaria</i> spp.	...
Solomon Islands	Eucheuma seaweeds nei	<i>Eucheuma</i> spp.	120
South Africa	Aquatic plants nei	<i>Plantae aquaticae</i>	2,750
South Africa	Gracilaria seaweeds	<i>Gracilaria</i> spp.	95
Taiwan Province of China	Aquatic plants nei	<i>Plantae aquaticae</i>	...
Taiwan Province of China	Laver (Nori)	<i>Porphyra tenera</i>	7
Taiwan Province of China	Warty gracilaria	<i>Gracilaria verrucosa</i>	9,085
Taiwan Province of China	Warty gracilaria	<i>Gracilaria verrucosa</i>	72
Tanzania, United Rep. of	Eucheuma seaweeds nei	<i>Eucheuma</i> spp.	6,000
Tonga	Zanzibar weed	<i>Eucheuma cottonii</i>	1,195
Un. Sov. Soc. Rep.	Brown seaweeds	<i>Phaeophyceae</i>	...
Venezuela, Boliv Rep of	Elkhorn sea moss	<i>Kappaphycus alvarezii</i>	...
Venezuela, Boliv Rep of	Spiny eucheuma	<i>Eucheuma denticulatum</i>	...
Viet Nam	Gracilaria seaweeds	<i>Gracilaria</i> spp.	30,000
TOTAL			13,927,067

¹Plantae aquaticae is designation for unidentified aquatic plant.

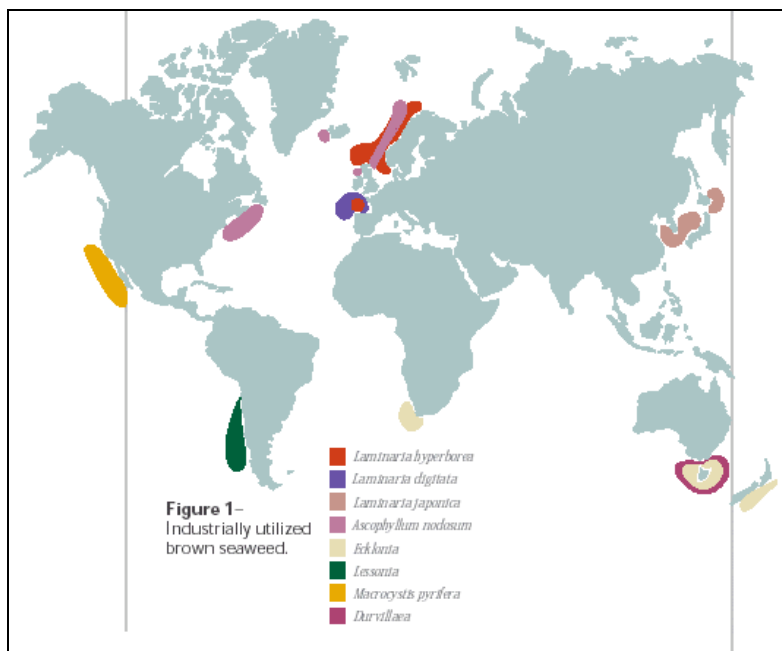


Figure 4. Industrially utilized brown seaweed resources.

Source: FMC Corporation, Biopolymer Division

(<http://www.fmcbiopolymer.com/PopularProducts/FMCAlginates/Origins/tabid/801/Default.aspx>).

Table 2. Annual production in metric ton dry weight of the main farmed seaweed genera by country in 2004 (Wu & Pang, 2006)

	China	Philippines	S. Korea	Japan	N. Korea	Indonesia
Brown seaweeds						
<i>Undaria</i>	2,196,070	0	261,574	62,236	0	0
<i>Laminaria</i>	4,005,640	0	22,510	47,256	444,295	0
<i>Sargassum</i> ¹	131,680	0	0	0	0	0
<i>Plantae aq.</i>	2,535,130	0	0	15,968	0	0
Total	8,868,520	0	284,084	125,460	444,295	0
Red Seaweeds						
<i>Porphyra</i>	810,170	0	228,554	358,929	0	0
<i>Eucheuma</i>	97,820	1,155,353	0	0	0	0
<i>Kappaphycus</i>		44,814	0	0	0	0
<i>Gracilaria</i>	888,870	0	0	0	0	0
<i>Unspecified</i>		0	0	0	0	410,570
Total	1,796,860	1,200,167	228,554	358,929	0	410,570
Grand total	10,665,380	1,200,167	512,638	484,389	444,295	410,570
% World production	76.7%	8.6%	3.7%	3.5%	3.2%	3.0%

¹FAO reports this as '*Fusiform sargassum*' the Chinese now classify this seaweed as *Hizikia fusiformis*

the exception of *Porphyra*, are farmed mostly for the production of agar and carrageenan. *Porphyra* is farmed exclusively for food and is commonly referred to as 'Nori'. The other marine colloid produced from seaweed is alginate, which is produced from brown seaweeds that are mostly harvested from natural populations. Some seaweed-derived protein is used for animal feed, including fish feed, and is a subject of great interest to many countries. While seaweeds have not yet been cultivated for production of fuels, many cultures have used seaweed biomass for small-scale heating and cooking processes (Neushul, 1987). Also, during WWI, the giant kelp *Macrocystis pyrifera* harvested off the California coast was used to produce potash and the organic solvent, acetone, which were needed to make cordite, an indispensable commodity for the war effort (Neushul, 1989).

Seaweed farming

Harvesting seaweeds from wild populations is an ancient practice dating back to the fourth and sixth centuries in Japan and China, respectively, but it was not until the mid-twentieth century that methods for major seaweed cultivation were developed (McHugh, 2003). Since that time, seaweed farming or marine agronomy has grown rapidly due to demand that has outpaced the productivity of natural populations. Today almost 90% of seaweed for human use comes from cultivation, rather than wild harvests (Zemke-White & Ohno, 1999). Four genera representing species of *Porphyra*, *Laminaria*, *Gracilariaria*, and *Undaria* comprise 93% of the cultured seaweeds (Santelices, 1999a).

Seaweeds have traditionally been grown in nearshore coastal waters, with some smaller operations on land. Offshore systems are an emerging seaweed culture technology, and the focus of the present report.

Traditional nearshore systems

The key components of the Chinese seaweed farming industry are shown in Figure 5 (Chen, 2006);. The Chinese have developed techniques and overcome significant challenges to achieve the current yields.



Figure 5. Seaweed farming in China (Chen, 2006).

The kelp *Laminaria* does not reproduce well vegetatively; Chinese aquaculturists have developed and perfected the difficult and costly technique of producing kelp plantlets in hatcheries. The need to produce plantlets could be a barrier to wide scale expansion of kelp farms, particularly offshore.

Most seaweed in Asia, including China, are grown on ropes slung between mooring structures, a technique known as ‘the floating raft method.’ Seaweed plantlet from a nursery are attached to the ropes as shown in Figure 6. This method is labor intensive and would be unsuitable for deep water systems.

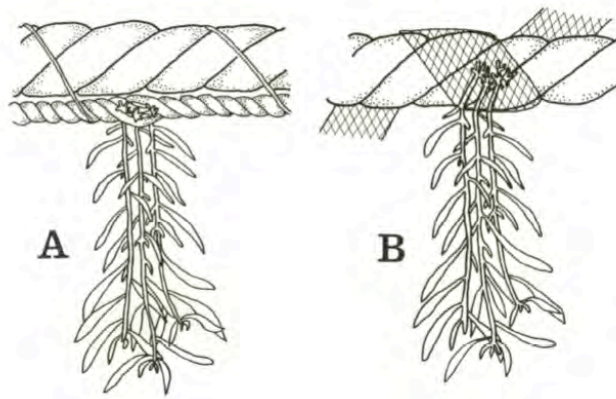


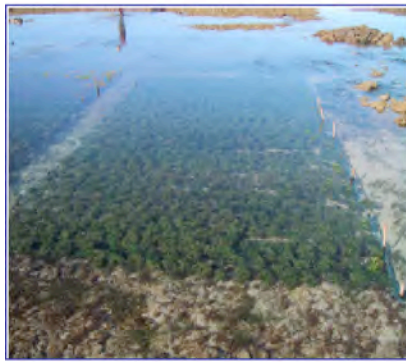
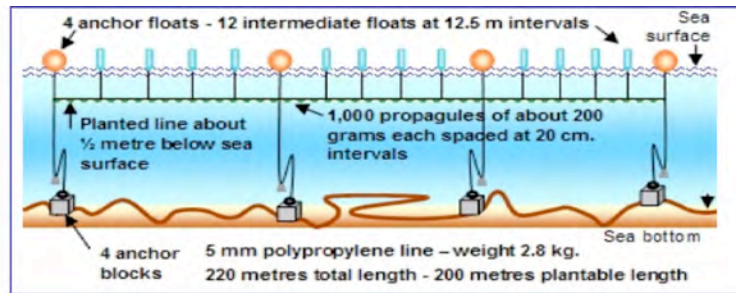
Figure 6. The floating raft method.

Production of kelp in China in 2004 was about 0.8 million metric ton dry weight from just over 40,000 hectares of farms (Chen, 2006). FAO’s wet weight figure for the same year is 4 million metric ton, indicating a wet to dry ratio of 5:1.

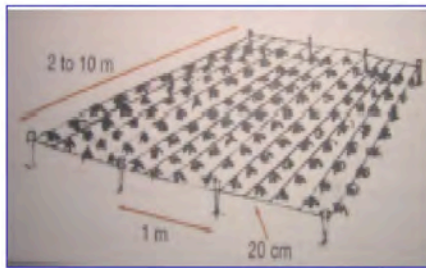
The production of all species of seaweed in China in 2004 totaled almost 100 metric ton (97.4 metric ton) wet weight per hectare (Chen, 2006). This production level can be compared with the yields of 300 metric ton per hectare per year of giant kelp *Macrocystis pyrifera*, projected 30 years ago by the U.S. Department of Energy Marine Biomass Program (see below for discussion of the Marine Biomass Program). High yields are thought possible with giant kelp because it grows fast under good conditions and because, unlike the Chinese kelp, can be ‘coppiced’ i.e. cut at intervals, allowing regeneration of plants and several sequential harvests (similar to an alfalfa field) (Chen, J., Yellow Sea Fisheries Research Institute, Qingdao, China;

personal communication). Harvesting Chinese kelp is very labor intensive, involving untying of the ropes, loading the kelp into sampans and then transporting the biomass to shore (FAO, 2008).

The red seaweeds, most notably *Porphyra*, are farmed in China, Japan and Korea. Red seaweeds other than *Porphyra* are farmed using different methods and are almost exclusively grown for the production of colloids. Figure 7 illustrates farming methods for *Eucheuma*, referred to generically as *Spinosum*, and *Kappaphycus*, which at one time was considered a species of *Eucheuma*, often referred to as *Cottonii*.



Eucheuma amoldii
Philippines



Green and brown *Eucheuma Spinosum* varieties



Kappaphycus alvarezii (*Cottonii*)

Figure 7. Illustrations and photographs to show red seaweed farming for marine colloids (Critchley & Ohno, 2006).

The largest seaweed crop farmed in China is the Japanese Kelp (*Laminaria japonica*) (Table 2). *L. japonica* is not native to China but was introduced from Japan in 1927, with large-scale kelp farming established in China in the early 1950s. For large scale *Laminaria* farming three key challenges had to be met (FAO, 2008): provision of dissolved nutrients, provided with synthetic fertilizers, breeding of summer rather than autumn plantlets, allowing for a longer growth season; and the movement of commercial cultivation into the southern provinces of Liaoning, Shandong, Jiangsu, Zhejiang, and Fujian, where the temperature is higher.

Nearshore seaweed cultivation shows some parallels with terrestrial agriculture. Terrestrial agriculture has succeeded in large part from the introduction of non-native plants and their selective breeding for new locations. Similarly the species of seaweed that have emerged as the preferred and dominant cultivars are not native to regions in which they are now most widely farmed. It can be reasonably argued that without introductions of non-native species in the past, agriculture would not be able to provide the amounts of food it does. Yet, these days, the introduction of exotic species, even for cultivation, is widely condemned and this may prove to be a significant barrier to any future seaweed bioenergy program. Almost by definition, any seaweed to be mass cultured in open ocean farms will be non-native to that environment. The use of genetically modified seaweeds must be evaluated in the same light.

The use of artificial fertilizers in Chinese seaweed cultivation paralleled the use of such fertilizers in the Green Revolution on land. Some coastal waters suffer from excess nutrient inputs, from sewage effluents and other sources, causing algal blooms.

Seaweed farms in China were frequently limited in their ability to produce plantlets. The multiphasic life cycles that characterize many seaweed species are more complex than simple seed production in most terrestrial plants. This is illustrated in Figure 8 for *Laminaria japonica*. Consequently, the production of plantlets for some seaweeds is time consuming and requires the sophisticated hatchery facilities shown earlier in Figure 5.

In China the cost of plantlets is \$50 to \$60 per ‘plantlet curtain’ each holding about 40,000 to 50,000 plantlets. However, large-scale seaweeds farms are unlikely to need hatchery-produced plantlets because a large plantation of seaweed should be able to perpetuate itself (Chen personal communication).

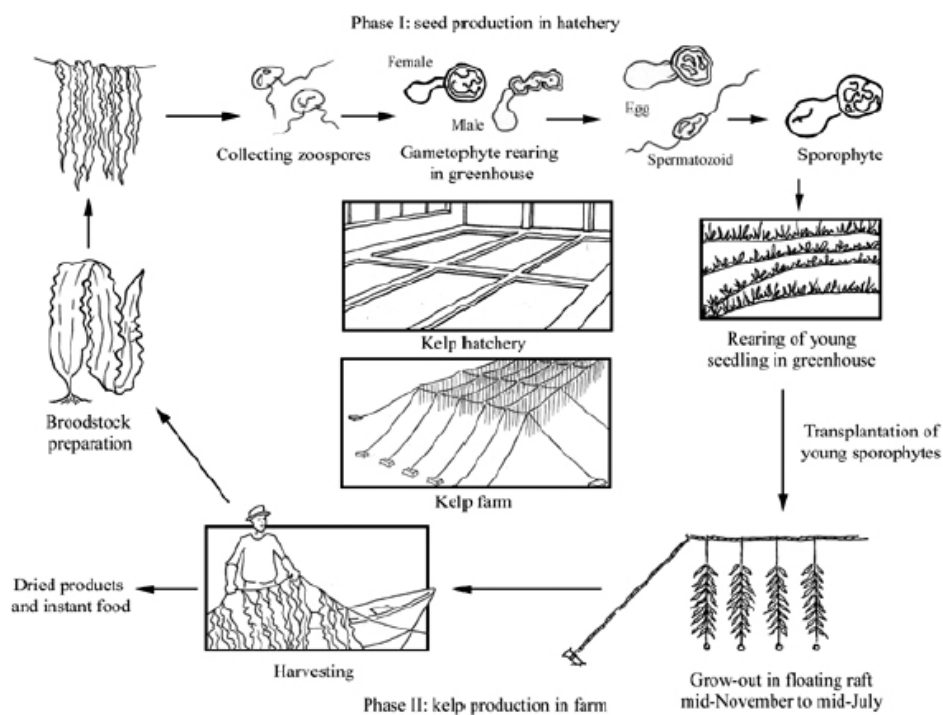


Figure 8. Production stages for the farming of Japanese kelp *Laminaria japonica* in China (FAO, 2008) (http://www.fao.org/fishery/culturedspecies/Laminaria_japonica)

Land-based systems

Land-based production of seaweeds affords greater control over farming methods, making it possible to grow seaweed at higher production densities than in near-shore farms. Land-based production also allows for the cultivations of seaweed species not well suited to oceanic farming, such as free-floating plants. One of the first studies of on-shore seaweed farming was carried out almost 30 years ago at Harbor Branch Oceanographic Institution, Fort Pierce, Florida, on red seaweed, *Gracilaria tikvahiae*. The seaweed was produced in small free-floating cultures with average annual productivity of 80-91 dry metric ton per hectare per year, equivalent to about 500 metric ton wet weight per year (Hanisak & Samuel, 1987), which exceeds yields assumed in the U.S. Marine Biomass Program for *M. pyrifera*.

More recently, a yield of 635 metric ton per hectare per year wet weight of *Ulva* spp. was obtained from land-based cultures growing downstream of a fish farm in Israel (Figure 9). Irish moss, *Chondrus crispus*, has been farmed commercially for over ten years (Figure 9), but little

information on this operation is available. Commercial onshore seaweed culture is also currently being carried out in South Africa, but, again, no production data are available.



Figure 9. Examples of land based seaweed production (Neori & Shpigel, 1999).

CHAPTER 3

BIOBASED SEAWEED PRODUCTS

Chinese and Japanese aquaculturists have been growing seaweeds for food and other uses in nearshore waters at a small scale for centuries (Tseng, 1981). There are occasional reports of farms in deeper water, but the practice has never been wide-spread, due to the difficulties of operating in open water conditions (Doty, 1982).

The extraction and use of chemicals from seaweeds, both harvested from the wild and produced in near-shore aquaculture facilities, is a modest industry in the U.S. (though domestic seaweed products still amount to over a billion dollars in sales in finished form) and a significant industry worldwide with a total economic value of \$5.5 to 6 billion USD (McHugh, 2003). The markets for seaweed products are at this point almost entirely for specialty food items and for extraction of functional polysaccharides – alginates, agar, carrageenans, which are used as gums and thickening agents mainly in foods and feeds, but also have some modest industrial applications (e.g., textiles, printing). Seaweeds have also been important as fertilizers, in particular for potash production.

Anaerobic digestion and extraction of valuable coproducts

Anaerobic digestion of seaweeds for methane, an energy product, is a microbial process in which complex organic materials are converted to simple sugars via hydrolysis, then further degraded via acidogenesis to volatile fatty acids (primarily acetate, propionate and butyrate) and hydrogen and carbon dioxide. The volatile fatty acids, other than acetate, are then converted to acetate, hydrogen and carbon dioxide by a process called ‘acetogenesis’ before being transformed to methane and carbon dioxide in a process called ‘methanogenesis’ (Figure 10).

Significantly, the major structural polymer complexes of seaweeds that are the raw material for this process are made up of mostly complex polysaccharides such as algin, laminarian, mannitol and fucoidan in the brown seaweeds and agar and carrageenan in the red seaweeds, and not lignin as is found in terrestrial plant biomass. This is significant because lignin resists anaerobic degradation, which has lead some to suggest that marine biomass may therefore be a more suitable substrate than terrestrial biomass for bioconversion to methane. However, unlike lignin, many of these complex polysaccharides have alternative commercial values, suggesting

that there is an opportunity cost that must be factored in to any overall assessment of economic

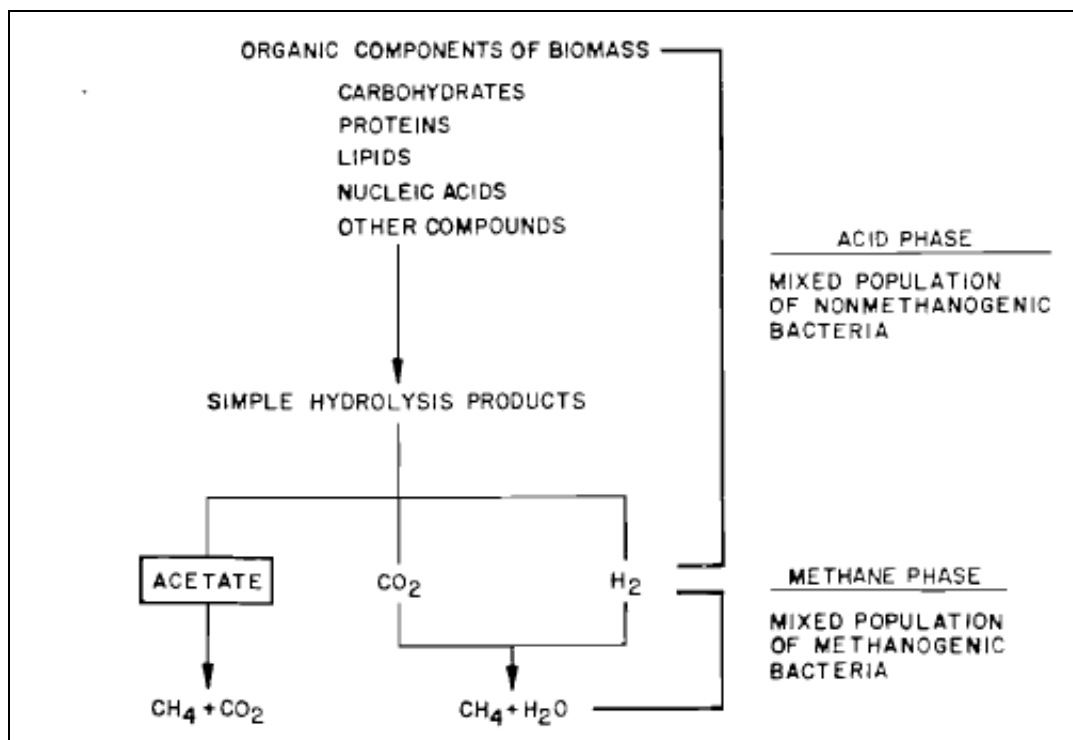


Figure 10. A simplified schematic of bioconversion of complex carbohydrates to methane (Forro, 1987).

feasibility. The same is true of the use of sugar from sugar cane or starch from corn in the production of ethanol by fermentation. Moreover, because anaerobic digestion is a relatively crude and unselective process, other seaweed constituents such as proteins and fats will almost certainly be degraded as well, as evidenced by the finding of Hanisak (1987) that ammonia comprised 40% to 70% of the total nitrogen content of the residues from digestion of *Gracilaria tikvahiae*. The residual solids obtained from anaerobic digestion of the kelp *Macrocystis* are rich in protein, with estimates exceeding 50 percent protein (Tompkins, 1983). Most of this protein is in the microbial biomass produced during methanogenesis. Its value could be similar to protein-rich Distillers Dried Grain with Solubles (DDSG), a product of corn fermentation, which is used as animal feed. However, the quality and processing required to make the seaweed protein suitable as animal feed have yet to be determined.

Anaerobic digestion of seaweed will yield methane, a sludge containing bacterial biomass (methanogens), plus ammonia and other products derived from the protein degradation. This prompts questions of whether the nutritional value of digester residues can be improved. Additionally, a bioprocessing scheme that extracts high value products before anaerobic digestion may be feasible. In considering bioenergy production from brown seaweeds, it was noted that *Ascophyllum nodosum*, a brown algae that is harvested in Norway to produce alginates contains the products shown in Table 3, some of which have potential commercial value.

Table 3. The chemical composition of *Ascophyllum nodosum*. Water content is given as a percentage of the fresh weight. All other components are given as the percentage of the dry weight. (Horn, 2000).

Component	[%]	Comments
Water	67 - 82	Decreased with salinity and lowered during the spring
Ash	18 - 24	Increased from autumn to spring
Alginic acid	24 - 29	Fluctuations during the year
Laminaran	1.2 - 6.6	Increased from spring to late autumn
Mannitol	6.8 - 10.4	Increased from early spring to early autumn
Fucoidan	4 - 10	
Other carbohydrates	10	
Protein	4.8 - 9.8	Increased from autumn to spring
Fat	1.9 - 4.8	Increased from early spring to late autumn
Fibre (cellulose)	3.5 - 4.6	Almost constant throughout the year
Polyphenols	0.5 - 14	Lowered during the spring and increased greatly with salinity
Iodine	0.06 - 0.09	Highest during the summer
K	2 - 3	
Na	3 - 4	
Ca	1 - 3	
Mg	0.5 - 0.9	
S	2.5 - 3.5	
P	0.1 - 0.15	

* Except for water content all other values in the table are expressed on a dry weight basis.

Thus, it may be advantageous to extract such products first and subject what is left to anaerobic digestion or fermentation as shown in Figure 11.

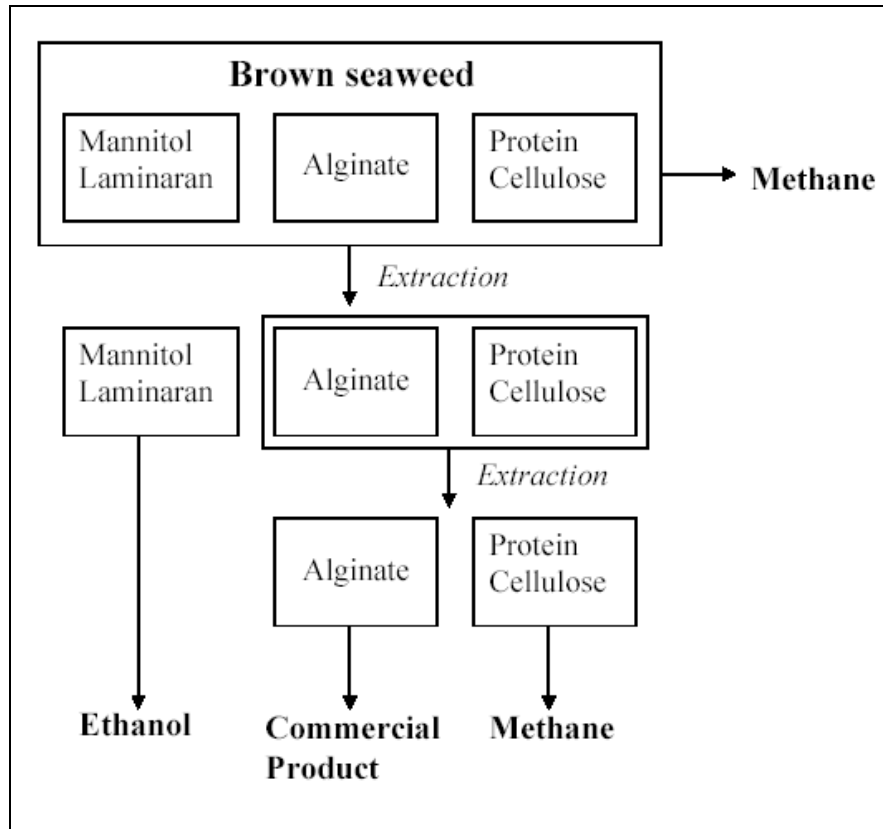


Figure 11. Pathway for processing brown seaweeds for fuel and other commercial products (Horn, 2000).

The economic merit of such an approach depends on processing costs and scale. A similar idea was considered during the Marine Biomass Program when one proposed production scheme assumed that 15% of harvested kelp would not be converted into methane, but would be processed into products of higher value instead (Tompkins 1982). The economic significance was diminished by the huge scale contemplated, which was made necessary by an exclusive focus of the program on energy production. The concept was not developed further.

Nutritional value of energy coproducts

Protein is degraded in digesters, producing ammonia, dissolved organic nitrogen, and bacterial biomass, none of which are suitable for incorporation into animal feeds to any extent. Therefore, it would be advantageous to recover more of the protein before it is degraded. Fermentations, such as ensilage (lactic acid fermentations), already practiced in making hay and

related fodders, could preserve the seaweed biomass, and in particular its protein, in a form suitable for animal feed. Of course, the choices of seaweed species cultivated will be a major factor in the co-production of animal feeds. The high content of carbohydrates in seaweeds suggests that the best option would be to extract the protein prior to the anaerobic digestion of the residual biomass. Extraction of leaf-protein, such as from alfalfa and other grasses, has been studied for many decades, but has not achieved commercial applications (except for xanthophylls-protein extracts used in chicken feeds). However, seaweeds may be more amenable to such fractionation and a better candidate for protein extraction, with the residues containing 80% or more of the organic material of the biomass available for biofuels processing anaerobic digestion. This topic has not been sufficiently investigated in studies conducted to date.

In most cases, seaweeds are used in human and animal foods for their minerals or for the functional properties of their polysaccharides and are rarely promoted for the nutritional value of their proteins (Fleurence, 1999). The protein content is not insignificant. In brown seaweeds, for example, proteins contribute between 3-15% of the dry weight and, in red and green seaweeds, between 10% and 47% of dry weight. In most cases, the protein content is calculated as the total nitrogen content multiplied by 6.25, a rough conversion factor used for meat and grains, but not always applicable to novel food and feed sources such as seaweeds due to the presence of nonprotein nitrogen.

Amino acid composition, a more direct measure of protein, varies considerably among seaweed species and with season (Table 4); with red seaweeds appearing to be an interesting potential source of feed and food protein (Marrion *et al.*, 2005). However, as stated above, seaweed protein is not readily digestible due to inhibition by phenolic molecules and/or polysaccharides (Table 5). *In vitro* digestibility tests on *Palmaria palmata* and *Gracilaria verrucosa* showed that protein digestibility, as compared to a casein control, was 4.9% and 42.1% respectively (Marrion *et al.*, 2005).

Seaweed meals fed to some aquaculture species have beneficial effects or, at least, do no harm. Valente et al (2005) reported that inclusion of meals made from *Gracilaria bursa-pastoris* and *Ulva rigida* at 10% in diets for sea bass had no deleterious effect on fish performance, while 10% inclusion of a meal made from *Gracilaria cornea* led to reduced digestibility and growth. Though not suggested by the authors, this may have had something to do with the fact that the *G.*

Table 4. The amino acid composition of some seaweed proteins (Fleurence, 1999).

Amino acids	<i>Ulva armoricana</i> (green seaweed) [15]	<i>Ulva Pertusa</i> (green seaweed) [2]	<i>Palmaria palmata</i> (red seaweed) [3,7]	<i>Porphyra tenra</i> (red seaweed) (2)	Leguminous plant [24]	Ovalbumin (2)
Histidine	1.2–2.1	4.0	0.5–1.2	1.4	3.8–4.0	4.1
Isoleucine	2.3–3.6	3.5	3.5–3.7	4.0	3.6	4.8
Leucine	4.6–6.7	6.9	5.9–7.1	8.7	7.3	6.2
Lysine	3.5–4.4	4.5	2.7–5.0	4.5	6.4–6.5	7.7
Methionine	1.4–2.6	1.6	2.7–4.5	1.1	1.2–1.4	3.1
Phenylalanine	5.0–7.1	3.9	4.4–5.3	3.9	2.4	4.1
Threonine	4.5–6.8	3.1	3.6–4.1	4.0	4.0	3.0
Tryptophan	—	0.3	3.0	1.3	1.6–1.9	1.0
Valine	4.0–5.2	4.9	5.1–6.9	6.4	4.5	5.4
Alanine	5.5–7.0	6.1	6.3–6.7	7.4	—	6.7
Arginine	4.3–8.7	14.9	4.6–5.1	16.4	13.0–14.0	11.7
Aspartic acid	6.0–11.8	6.5	8.5–18.5	7.0	4.7–5.4	6.2
Glutamic acid	11.7–23.4	6.9	6.7–9.9	7.2	6.4–6.7	9.9
Cysteine	—	—	—	—	1.1–1.3	—
Glycine	6.3–7.5	5.2	4.9–13.3	7.2	—	3.4
Proline	5.0–10.5	4.0	1.8–4.4	6.4	—	2.8
Serine	5.6–6.1	3.0	4.0–6.2	2.9	—	6.8
Tyrosine	4.4–4.7	1.4	1.3–3.4	2.4	2.3–2.6	1.8

Table 5. Relative digestibility of some seaweed proteins (Fleurence, 1999).

Seaweed species	Pepsin % Digestibility*	Pancreatin % Digestibility*	Pronase % Digestibility*
<i>Ulva pertusa</i> [2] (green seaweed)	17.0	66.6	94.8
<i>Undaria pinnatifida</i> [2] (brown seaweed)	23.9	48.1	87.2
<i>Porphyra tenera</i> [2] (red seaweed)	56.7	56.1	78.4
<i>Palmaria palmata</i> [7] (red seaweed)	—	56.0	—

*Relative digestibility is expressed as a percentage compared with casein digestibility basis (100%).

cornea meal contained substantially less protein (11%, versus 30.2% and 29.5% for the other seaweeds respectively) and therefore, presumably, contained more polysaccharides. Several other papers cited by Valente et al suggest beneficial or ‘no effect’ results with small quantities (5%) of seaweed meals fed to other fish species including Tilapia and red sea bream. On the other hand, inclusion levels of 16% and 33% in diets for mullet suppressed growth and feed utilization (Davies et al, 1997 cited by Valente, 2005). In terms of other nutritional effects, Casas-Valdez et al. (2006) found that when a meal made from *Sargassum* spp was included at 4% in feeds for brown shrimp, it reduced cholesterol levels in the shrimp without affecting growth.

Pre-treatment of seaweed meals to improve digestibility, followed by determination of their nutritional value in aquaculture feeds at higher levels of inclusion than 5%, might provide useful information. Separation or extraction of seaweed protein before digestion might provide a substantial additional revenue stream for a bioenergy program, as well as making an important contribution to global animal and human nutrition. In addition, digestion might allow for the decreased degradation and recovery of valuable nutrients before the residue is used as fertilizer for seaweed production.

An alternative approach is to chemical pretreatment of seaweeds to liberate fermentable sugars as substrate for appropriate inoculum, such as yeast or *Clostridium*, for production of ethanol and other products such as butanol. These products have a higher fuel value than methane in most circumstances and would produce a byproduct analogous to the Distillers Dried Grain with no Solubles (DDG) and DDGS from corn fermentations. This represents a potentially large new market for seaweed byproducts, although DDG and DDGS have a value not much above that of the fuel value of fossil fuels.

Alginates and other chemicals

As the market for large-scale seaweed production for energy emerges, the economic role of these products might be significant. By examining the analog of terrestrial agriculture, it is likely that there is a range of still-to-be discovered products of value (Tompkins, 1983). A summary of data presented by Tompkins is provided in Table 6.

Tompkins looked only at the potential co and byproducts that could be produced from giant kelp *M. pyrifera*. Such an analysis for other species of seaweed might yield quite different results and other seaweed species may in fact be better overall candidates than *M. pyrifera* for large-scale biomass production.

Table 6. Selected data on coproducts and byproducts from giant kelp methane production (Tompkins, 1983)

Basis: Farm production 500 wet ton (2000 lbs) per day, 365 days per year.

Composition: Water 87.5%, Organics 7.5%, Inorganics 5.0%

	% Dry wt	Recovery %	Prod'n – ton/yr	Price \$/ton	Revenue - \$/yr millions	Annual U.S. demand (ton)
<u>Methane</u>			20,139	473	9.5	
<u>Coproducts</u>						
Organics						
						7,800
Algin	17	72	2,792	6,000	16.8	N/A
Mannitol	15	70	2,395	6,000	14.4	N/A
L-Fraction	5	65	740	2,000 – 6,000	1.5-4.4	N/A
Fucoidan	1.5	60	208	6,000	1.25	N/A
Inorganics						
Iodine	0.3	65	42	14,500	0.6	4,450
Potash	26	60	3,558	60	0.21	7,000,000
Bromine	0.1	65	15	1,200	0.02	N/A
<u>Byproducts</u>						
Carbon dioxide	N/A	90	4,039	35	1.4	2,500,000
Sulfur	N/A	90	322	120	0.04	N/A
L-Fraction	N/A	65	4,603	2,000 - 6,000	9.2 – 27.6	N/A
Bacterial protein	N/A	80	11,220	70	0.8	N/A
Potash	N/A	60	22,441	60	1.35	7 million
Iodine	N/A	65	288	14,500	4.2	4,450 ton
Bromine	N/A	65	115	1,200	0.1	N/A

1. The data are Table 6 from 1982 and earlier updated market values are needed.
2. Coproducts are assumed to be taken before digestion for methane production; byproducts are recovered from digester residues.
3. Though these figures show substantial potential contributions to the total value of raw or digested kelp, a farm that produces 500 metric ton wet weight per day would have contributed less than 0.01% to the Nation's natural gas consumption at the time.

4. Carbon dioxide and hydrogen sulfide are formed during the process of digestion. While carbon dioxide and hydrogen sulfide are not considered to be valuable resources; lessening their release into the atmosphere would be advantageous
5. L-Fraction is that which remains in the digester, also referred to as ‘phenolic compounds’ and analogous to the lignin structure in terrestrial biomass. Methanol extraction could be used for purifying the L-Fraction from digester residues. At the time (1982) it was thought that L-Fraction might have an application in plastics, adhesives and time-release dispersants but no market had actually been established for it. Therefore the potential revenue contributions noted by Tompkins are speculative.
6. Bacterial protein, which is kelp protein that is not denatured during the digestion process plus bacterial biomass generated during digestion, was considered as a possible ingredient in animal feeds, as also was raw kelp protein. Based on *in vitro* digestibility tests, amino acid analysis and limited feeding trials, Hart et al (1976) developed some estimates of value for kelp in cattle and poultry diets (Table 7).

Table 7. Estimates for amount of kelp in cattle and poultry diets.

Ration	<u>Whole kelp</u>		<u>Press Cake</u>	
	\$/ton	% of ration	\$/ton	% of ration
Beef	57-59	7-9	67.50-69	4-9
Poultry	-	-	10	16

7. The value and size of the markets for potash and iodine clearly make it worthwhile recovering these materials from digester effluents, at least in the early stages of development of a marine biomass energy program, if this can be done at reasonable cost.

CHAPTER 4

CHAPTER 4. OFFSHORE SEAWEED FARMS FOR BIOFUEL PRODUCTION

Past attempts at offshore seaweed farming

During WWI a chemical industry was established in Southern California to produce acetone and butanol, a by-product, by acetone-butanol bacterial fermentation of *Macrocystis pyrifera*. Using about 400,000 ton of this seaweed annually, it provided the major source of acetone required by the British Navy for smokeless gunpowder. After WWI, the facility was no longer economic to operate and was closed. However, it demonstrated that it is possible to ferment seaweeds to produce organic chemicals and fuels; butanol is now touted as the next generation biofuel, superior to ethanol. It also serves as an example of the role that seaweeds can play in fuel production should the need arise. The recent history of offshore seaweed farms is intertwined with the production of biofuels from seaweeds, driven by a search for alternatives to petroleum during the energy crisis of the 1970s.

The first concentrated effort to grow seaweed as a source of energy in offshore waters began in the early 1970s with small-scale research efforts funded by the U.S. Navy and carried out at several universities, including studies of floating, non-tethered seaweed farms (Neushul & Harger, 1987). With the energy crises of the 1970s, the Marine Biomass Program was initiated in the U.S., with mostly government funding. The objective was to mass culture the giant kelp *Macrocystis pyrifera* in large open ocean farms, and produce biogas as a replacement for natural gas, which was thought at the time to be vanishing rapidly. While the Marine Biomass Program added considerably to our knowledge base on seaweed cultivation, the program was not technically successful due to the loss of the floating farm equipment under open ocean conditions, the loss of attached algae, and problems with cultivation (e.g. epiphytes, fish predations, etc.) (Neushul, 1985). During these initial attempts, made largely in the Pacific Ocean off California, the plants were fertilized by pumping nutrient-rich water from depth. Other seaweed species, including *Laminaria*, *Gracilaria* and *Sargassum*, were also tested during the Marine Biomass Program. In the following sections we discuss in more technical detail the biological and engineering issues of proposed open ocean seaweed-to-energy systems. Specifically we address systems analyses and experimental work on open ocean seaweed

biofuels production from about 1974 to 1984 in the U.S., the Marine Biomass Program and related work (Jackson, 1988; Neushul, 1987; North, 1987; Richard, 1992).

Initial concept for biofuels from open ocean seaweed farms

The first concept for an open ocean biofuels process using macroalgae suggested that juvenile *Sargassum sp.* be released about 500 miles offshore at the latitude of the U.S.- Canada border, where the plants would be carried by existing currents south to the latitude of the U.S.- Mexico border, where they would be harvested by waiting ships and transported to onshore anaerobic digesters for conversion to methane (Szetela *et al.*, 1978). Although this proposal did not lead to further research on open ocean *Sargassum* farming off the California coast, it provided the inspiration for the “Ocean Food and Energy Farm Project”, a multi-product floating seaweed farm proposed in the late 1960s’ by Howard Wilcox, then with the San Diego Naval Undersea Center. As originally conceived, the production of fuels was not seen as the most valuable product of the offshore farm. The concept envisioned large floating rafts that would surround a nutrient upwelling pipe. Figure 12 shows the general schematic of the proposed

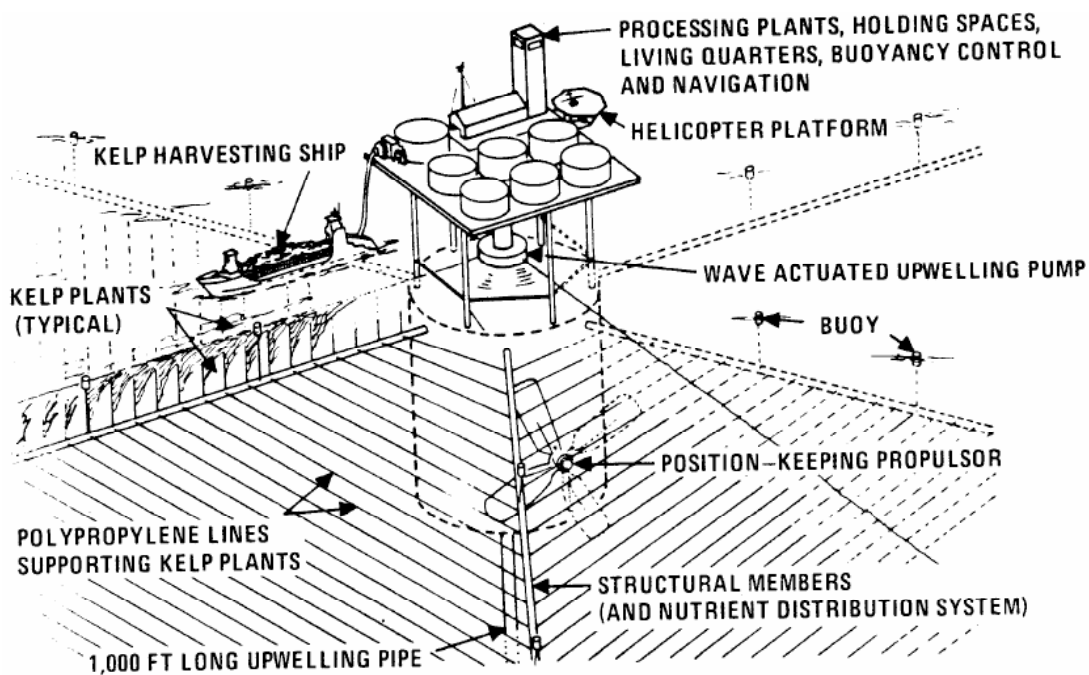


Figure 12. Initial concept for an Ocean Food and Energy Farm (Wilcox, 1975)

system. Wilcox obtained funds from the Navy and carried out the first attempt to establish such a facility in 1974, at a deep ocean location off the St. Catalina Islands (see Chapter 5, Figure 16). Because of energy shortages at the time, the priority was redirected to the production of biofuels, specifically methane production. By the mid 1970s the U.S. natural gas industry (American Gas Association, AGA), and the U.S. Energy Research and Development Administration (ERDA), and their successors, the Gas Research Institute (GRI) and the U.S. Department of Energy (via the Solar Energy Research Institute) took over sponsorship and management of the project from the U.S. Navy. The General Electric Company was selected as the prime contractor to carry out the project.

The open ocean farm concept – the ISC report

At the time of these initial developments, a detailed engineering and cost analysis of the multi-product ocean farm concept was commissioned by the Energy Research and Development Administration (ERDA), the predecessor of the U.S. Department of Energy. The analysis was carried out by a private contractor, Integrated Science Corp. (ISC), and published during 1976-1977 in a seven volume set (Budharja, 1976).

The process proposed and analyzed by ICS envisioned the cultivation of the giant kelp *Macrocystis pyrifera* native to the California Coast in 100,000 acre (400 km²) size farms, comprising 10 acre (4 hectare) triangular modules with large buoys at each corner, connected with large tension lines, and provided with a dynamic positioning system. A grid of “substrate lines”, at a depth of about 15 meters or so, would provide attachment sites for the plants, with smaller buoys spaced every 100 feet (30.5 meters). The modules were to include large upwelling pipes with wave power-actuated pumps to provide nutrient-rich deep ocean water to fertilize the floating plants. Buoyancy control would allow the entire system to sink below the waves in case of storms. (In the initial analysis only 100 meter deep upwelling pipes were specified. In retrospect, these needed to be extended several hundred meters, as nutrient concentrations are generally only high enough at much greater depth, closer to 500 m).

Figure 13 shows the design proposed by the ICS report. The concept was that once a single plant was planted in such a 10-acre system it would quickly propagate and fill the entire structure

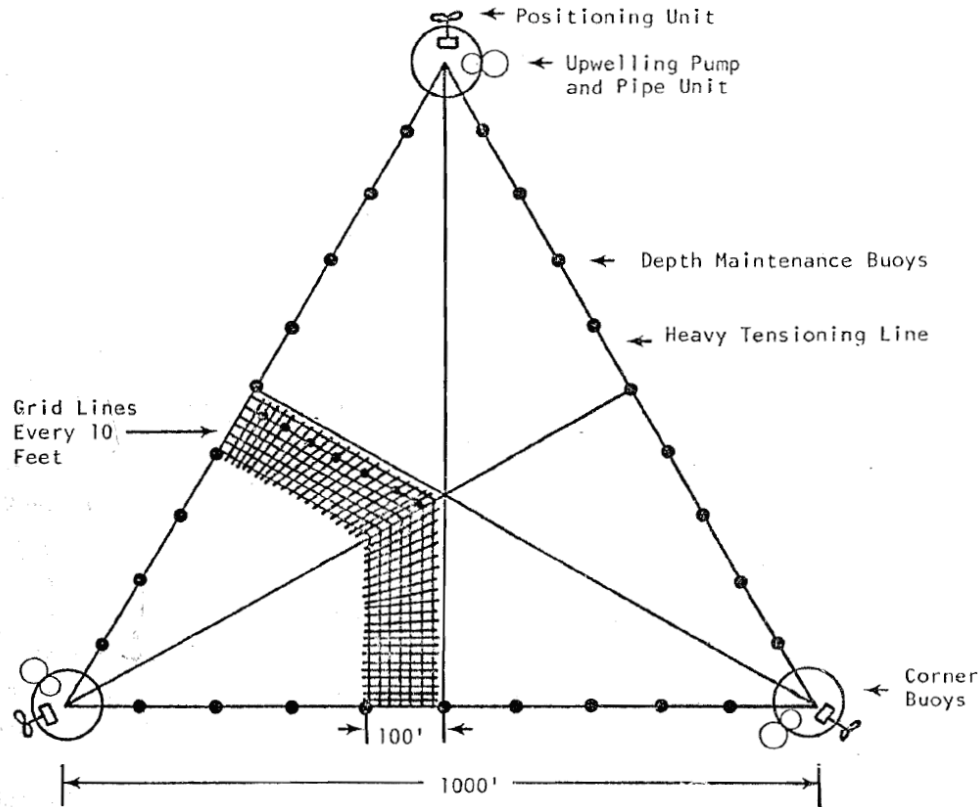


Figure 13. Design of the 10 acre Ocean Farm Module (Budharja, 1976).

(one plant per 10 m²), over a period of four years, at which point harvesting could commence. Power units (diesel, though also fueled by methane) were to be provided at each corner for dynamic positioning of the farm unit. The latter would allow the farm unit to adjust to the local current, keeping its lines and substrate line taut and in position, preventing damage to the plants. The upwelling was to be achieved with wave actuated pumps, backed-up with diesel/methane pumps.

A harvest yield of 55 metric ton of volatile solids per hectare per year was assumed. The amount of nutrients required was based on a N content in the seaweed (on an ash-free dry weight basis) of about 5% and a nutrient utilization efficiency of 70%. From this it was estimated that upwelling of 1.5 m³ of water per day, containing about 0.4 mg/l of N-nitrate, through the seaweed canopy would supply all the required N, assuming that some nutrients would be recycled. (Note: ash typically is 30 to 50% of the dry weight of these seaweeds, and there is a great deal of confusion about wet, dry and ash-free dry weights in many of the reports.)

The ISC reports provided much detail on the design of the system, such as of the tension ropes and substrate lines, the buoys and wave pumps, the pipes and harvesting machinery, etc. Harvesting was to be carried out by ships and the biomass anaerobically digested to produce methane gas at a rate of about 10 million BTU/ metric ton ash-free dry weight. The reports included many engineering estimates, such as drag coefficients for the farm, tension on the ropes and support structures, energy required for pumping water, harvesting rates, manpower for planting the seaweeds.

The capital costs were estimated in 1978 at about of \$570 million for the 100,000 acre system, not including start-up and working capital of about \$190 million and annual operating costs of \$61.4 million. If we assume that the farms could produce 2.4 million ash-free dry ton per year, the cost of 100 ash-free metric ton of biomass would be \$63. At 10 mmBTu/ton of net gas production, this would suggest a cost of \$12/mmBTu of gas. The ISC reports also estimated a one third reduction in cost to just under \$8/mmBTu of gas from byproduct credits. A more sophisticated financial model suggested a methane cost of only \$5/mmBTu that could be reduced further by about 25% by including a mariculture component. This addition added less than 10% to the overall capital costs, but was projected to more than double total revenues from the byproduct fisheries. The mariculture (i.e., the “food”) part of the process provided additional revenues at minimal costs, offsetting some of the high capital and operating costs. (For current \$ figures multiply these by about three). Although these were rather high cost projections they were of interest for renewable energy resources. It was concluded that the concept “offers a long-term promise for supplying large quantities of energy and food. The cost and productivity estimates make it roughly competitive with other energy systems.”

The Dynatech report

The Dynatech report (Ashare *et al.*, 1978), commissioned by the Department of Energy, reviewed in detail the feasibility of deriving energy from aquatic biomass, including open ocean seaweed cultivation. The Dynatech team reviewed the assumptions made in the ICS report and others and carried out independent calculations on many of the physical and biological parameters involved. For example, with regard to the critical issues of nutrient supply and distribution, the Dynatech analysis included calculations of the energy required to upwell water, the wave energy available, the dispersion and nutrient dilution of the upwelled water due to

currents, the rate at which colder upwelled water will sink when brought to the surface, and the mass transfer limitations of upwelled water contacting the plant surfaces. They pointed out that, based on fundamental research (Jackson, 1977), a major limitation of mass culturing seaweeds is the low mass transfer coefficients in seawater, limiting the supply of CO₂, nitrate and phosphates, and possibly other nutrients. Large energy inputs would be required to mix seawater and break down diffusion barriers near the surface of the seaweed blades.

Mixing, the intensity of which is measured as power density (W/m³), is provided in natural near-shore kelp beds by natural wave action and water currents. Even there, productivity is often limited more by diffusion than actual supply, though of course the biomass concentrations are quite low. Diffusion limitations are expected to be a greater problem in open ocean systems than in near shore aquaculture activities, where natural currents, in terms of power dissipation, are greater. Thus in open ocean environments where a high intensity cultivation process is required, both CO₂ limitation and in particular N (and P) limitations can be anticipated. This would significantly limit the productivity of any such scheme. Turbulence, requiring high power inputs for mixing, is a major limitation to productivity of seaweeds in on-shore systems and is even more likely to be so in open ocean processes. They also concluded that wave power does not appear to be sufficient for upwelling the deep nutrient rich water for operating the plant, at least not during the most productive summer months. They concluded that the system would require too much fuel to operate. However, most problematic, was the economics of the system. Figure 14 summarizes their costs (\$1978) vs. productivity estimates.

The entire spectrum of design assumptions and issues reviewed by the Dynatech study concluded that the earlier assumptions were unsustainable or at least overly optimistic. However, their economic analysis was not that far from the earlier ICS report, with a cost of \$170/ton or \$32 /mmBTu for Dynatech vs. \$120/ton and \$8/mmBTu for ICS. The high cost per mmBTu of the Dynatech report was due to the high ancillary energy use, and the total lack of by-product credits.

The Dynatech report concluded that that even with very favorable assumptions the economics “were above any practical costs to be considered for energy”. Nevertheless, the Department of Energy funded the Marine Biomass Program.

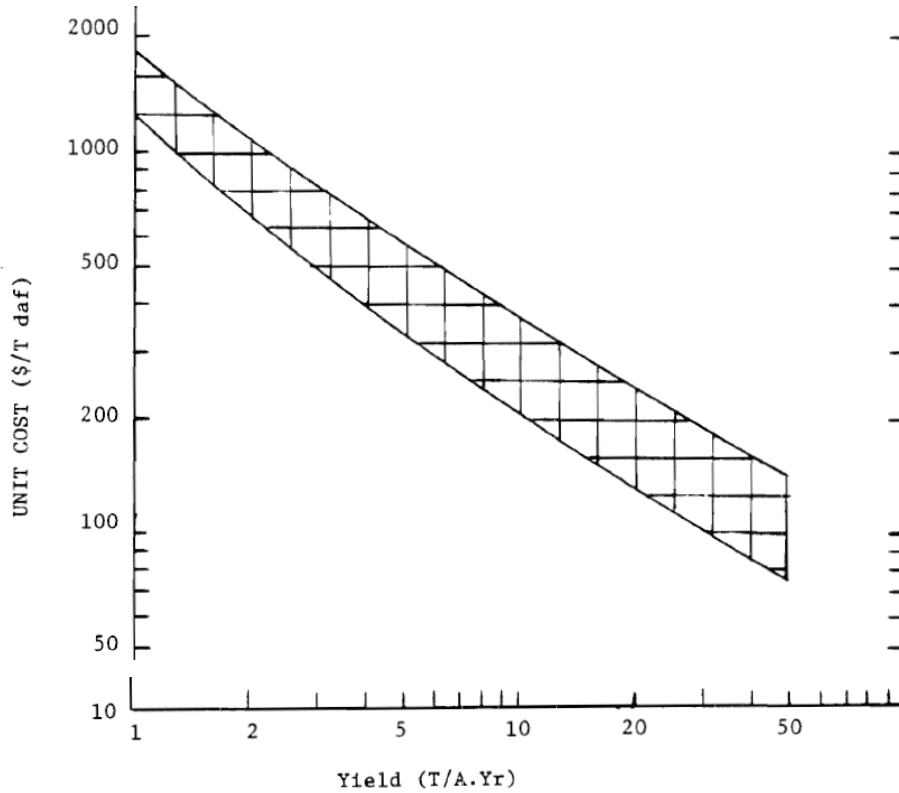


Figure 14. Costs ranges for open ocean farm *Macrocystis* production (units: \$/T daf, dollars (USD) per ton delivered at frontier; T/A.Yr, Ton per Acre per Year) (Ashare *et al.*, 1978)

The Marine Biomass Program

The Marine Biomass Program was one of the largest, if not the largest, single investment made by the U.S. Department of Energy during the period from about 1979 to 1983, exceeding \$30 million dollars (roughly \$100 million in 2008 USD). The Marine Biomass Program continued on the path outlined by the ICS Report, with the major focus being on the Ocean farm trials of several floating platforms that were to grow California’s giant kelp *Macrocystis pyrifera*.

The first farm unit was installed in September of 1978. During December the protective curtain came loose, the plants were lost, then the farm was destroyed. A second system was deployed, but the results were again disappointing with little demonstrable success in growing the seaweed plants for any length of time. By 1981 it became clear that the open ocean

environment was too harsh. Plans were made to move the operation close to shore, when the program was terminated in 1983.

Despite these setbacks, extensive data were collected during this entire period, from the early 1970s, to the end of the program, by Prof. Wheeler North of the California Institute of Technology, and later by others, in particular Prof. Michael Neushul at University of California Santa Barbara. Research on the provision of nutrients to the plants, in particular the use of upwelled seawater, was one major focus of the work. In general the Marine Biomass Program made significant advances in understanding some of the complexities of the biological system. Growth data were also collected, but the difficulties of working in the open ocean environment, of managing plants, and extrapolating laboratory data to the field, limited the ability to predict full-scale field productivity that might be achieved.

Studies with land-based systems have shown that achieving productivity of 50 to 70 metric ton ash-free-dry-wt per hectare per year, a level of seaweed growth assumed by the ICS and Dynatech reports, is possible (Hanisak, 1981; Hanisak, 1987; Hanisak & Samuel, 1987; Ryther *et al.*, 1984). However, these studies also suggest that these levels of productivity require a relatively large energy input to reduce diffusion limitation by CO₂ and other nutrients. This is a central issue that must be addressed.

Conclusions regarding open ocean seaweed biomass production

The history of the actual field trials is not encouraging: the first test farm, with 100 transplanted kelp, broke loose and was destroyed soon after installation, the second sunk and the third lasted less than one month. Only one upwelling experiment was ever carried out. The test farms were plagued by storms, accidents and most importantly, great difficulties in managing the plants. The initial laboratory work on growing seaweeds on upwelled water suggested a limiting nutrient or toxic effect. Although this was later resolved it was clear that much needed to be learned. However, much useful information was learned. The research demonstrated that giant kelp could survive, for a time, and even grow, when tethered to an open ocean structure, using upwelled nutrients. Thus it can be concluded that it would be possible to grow such seaweeds if sufficiently supplied with nutrients. However, the question of the productivity of such systems and the practicality of such a process has yet to be demonstrated.

It is the combination of the many biological, engineering and economic issues that together makes open ocean mariculture such a daunting challenge: even overcoming one or two or half a dozen, will not be sufficient. Consider the following major issues:

1. *Containment, Protection and Distribution.* It is clear that no engineering design has thus far been demonstrated that can contain the plants and provides them protection from storm and other damage. Concepts, such as proposed in Japan, of using large open pens enclosed by nets would most likely suffer from great difficulties in avoiding the plants bunching together.
2. *Productivity.* Productivity may not be a limiting factor, given the large surface areas of available open ocean. The more important issue is the creation of an extensive engineering design (lower per hectare cost) able to accommodate lower plant density and, thus productivity. If the system is affordable and robust, the level of productivity matters less.
3. *Nutrient Supply and Uptake.* The concept of nutrient upwelling is one that has yet to be demonstrated to be technically feasible, considering all the issues of plume dispersion, sinking and even CO₂ releases. If not supplied with upwelled nutrients the plants must be artificially fertilized, in part possibly with recycled nutrients. However here the efficiency of nutrient assimilation would become critical, but it is unclear if high nutrient utilization is possible in open ocean systems.

The development of open ocean seaweed farming systems was premature in the 1970s. The Marine Biomass Program did not gather sufficient experience to overcome offshore challenges of open ocean forces and balance them with the engineering needed to successfully site a seaweed farm. The Marine Biomass Program moved towards nearshore systems to gain needed experience. The current situation has changed with pertinent experience gained through oil and gas exploration, oceanographic and atmospheric surveillance of ocean conditions and weather prediction, and major improvements in tensile strength and weight of materials that can be used at sea.

CHAPTER 5

STRUCTURES AND TECHNOLOGIES FOR OFFSHORE FARMS

There have been relatively few offshore farms designed and operated solely for the cultivation of seaweed; much of the experience in offshore aquaculture comes from fish farming and more recently from the advent of offshore wind farms, which are now being integrated with aquaculture operations.

Floating versus anchored platforms

Offshore aquaculture systems are designed as floating, anchored, or as a combination design. Each has distinct advantages and disadvantages but most have yet to be tested in large-scale operations. Several small-scale pilot projects have been mounted, mainly in Europe and Japan. There are also numerous patents applied for and pending on offshore systems, both in the US and other countries, and a number of review articles and assessments have appeared in the scientific and technical literature. Here we discuss some of the two major types of systems that have been considered – free- floating enclosures and anchored platforms.

Floating offshore aquaculture farms drift at the mercy of ocean currents, wind and waves. They are designed to move easily up and down in a vertical motion that can range up to several meters under normal sea conditions and tens of meters during a storm. Although these systems can be positioned at depths that avoid storm damage, a major challenge of such floating systems is the uncertainty of where the farms may end up after a storm, and the depletion of nutrients (including C) due to the limited water flowing past the fronds under normal weather conditions.

There has been renewed interest in floating farms in recent years as platforms and technologies have improved to withstand open ocean conditions. Almost all that were moderately successful have combined a floating platform with a tethered or anchored component (Buck & Buchholz, 2005).

Most aquaculture systems are tethered to the sea floor through anchors, floats and lines. Cultivation in deep offshore waters raises the complexity and cost of such systems, due to the cost of materials and labor working at increased depth, and the need to periodically withstand severe oceanic conditions. Anchored systems have the advantage of certainty of location, and that ocean water is constantly passing through the plants, breaking down diffusion barriers,

providing nutrients (if available) and washing away waste products. If the surrounding ocean water is low in nutrients, the anchored systems can support complex fixed structures such as nutrient upwelling pipes. However, the design of anchored systems that can operate under the normal vertical motion of ocean water, and are able to withstand severe conditions, is challenging.

The first tethered seaweed cultivation systems, specifically for the purpose of marine bioenergy production, were installed off the Southern California coast in the early 1970s to grow *Macrocystis pyrifera*, the giant California kelp. This was a precursor project (the “Marine Farm Project”) to the later Marine Biomass Program, mentioned above and discussed further below). The initial system was anchored at a depth of 50 to 150 m, about 1 km offshore, with the plants held at a depth of 12 m (Figure 15). Strong currents, occasionally enriched with nutrients, swept past the juvenile plants that grew only slowly. Dispersing water from a depth of 30 to 50 meters, which had higher nutrient concentrations, increased their growth. However, further experiments with deeper water were inconclusive, with some growth inhibition noted, and research, by Prof. North, retreated for some time to the laboratory to study plant growth in deep ocean water.

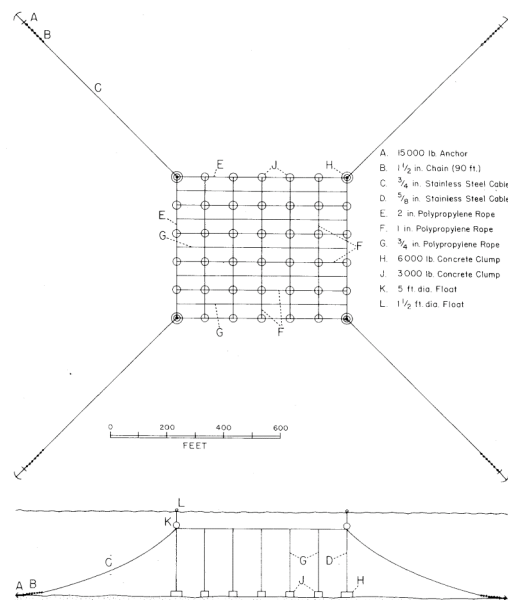


Figure 15. Diagram of the fixed grid seaweed farm installed by the US Navy in southern California in the early 1970s. The grid was designed to hold up to 1000 *Macrocystis* plants, but only 130 were actually installed (North, 1987).

Nevertheless, this first test of the open ocean farm concept provided a great deal of information and lessons (such as not to let plants come in contact with the grid) for the work later carried out under the Marine Biomass Program.

The next major development was the Offshore Test Platform (OSTP), an anchored platform designed to test the concept of growing seaweed for energy, built as part of the Marine Biomass Program during the late 1970s (Figure 16). The OSTP resembled an upside down umbrella, with

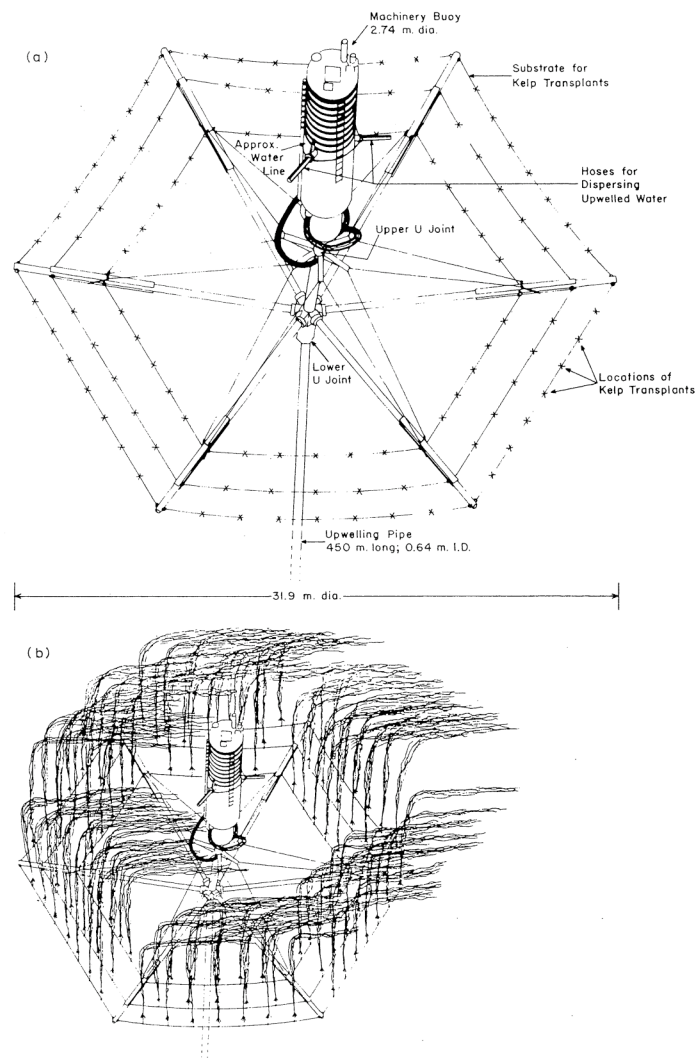


Figure 16. Diagram of the floating seaweed farm (the Quarter Acre Module) anchored off Newport Bay, California. The farm held up to 100 plants. Diesel-powered pumps brought nutrient-rich water from depth to fertilize the plants (North, 1987).

a cold-water upwelling pipe bringing deep water to the surface to supply the *Macrocystis* plants with nutrients. The module used to grow the plants, was designed to be a quarter acre (or 0.1 hectare) in size, dubbed the “QAM”. The QAM was used with several different moorage systems before the project was abandoned in the early 1980s (Bird & Bensen, 1987). Subsequent refinements of the Marine Biomass Program platform included an extensive grid design to support multiple growing areas, as well as a variation that included a four-point anchor system (Figure 17).

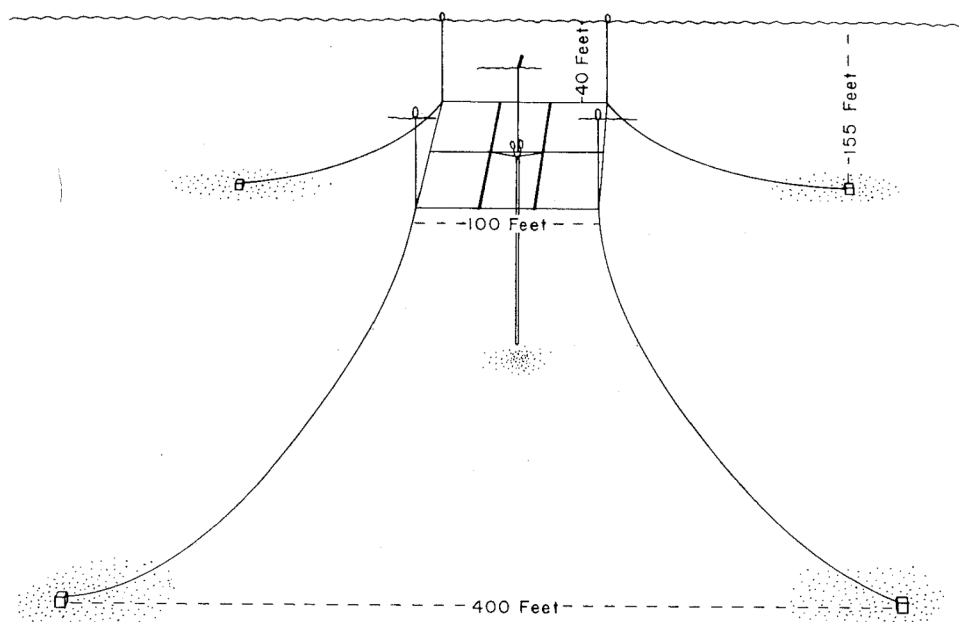


Figure 17. Diagram of a four-point moorage system used with OSTP (North, 1987).

More recently, several successful aquaculture platforms were deployed in the Mediterranean and the North Sea (Lisac, 1997); (Buck & Buchholz, 2004). These systems took several very different approaches to withstanding ocean conditions. The “Ocean Spar System” is based on several previous U.S.-designed models that were anchored with four vertical spars, between which an aquaculture cage was suspended (Loverich & Goudey, 1996). This design allowed the cage to be moved vertically, submerging it below the surface during severe ocean conditions (Figure 18). The rigidity of this system was useful in areas of high current velocity but could be detrimental above certain wave and wind conditions. The “Tension Leg System” was derived from this concept, using rigid spars at the corners and rigid components at depth. It allows the near-surface structure to be small, flexible and loose in order to absorb wave and

current action (Figure 19). This system has been shown to be seaworthy, easy to maintain and operate, and less costly than several other options. Tests in the Mediterranean proved moderately successful, although long term deployment has not been completed.

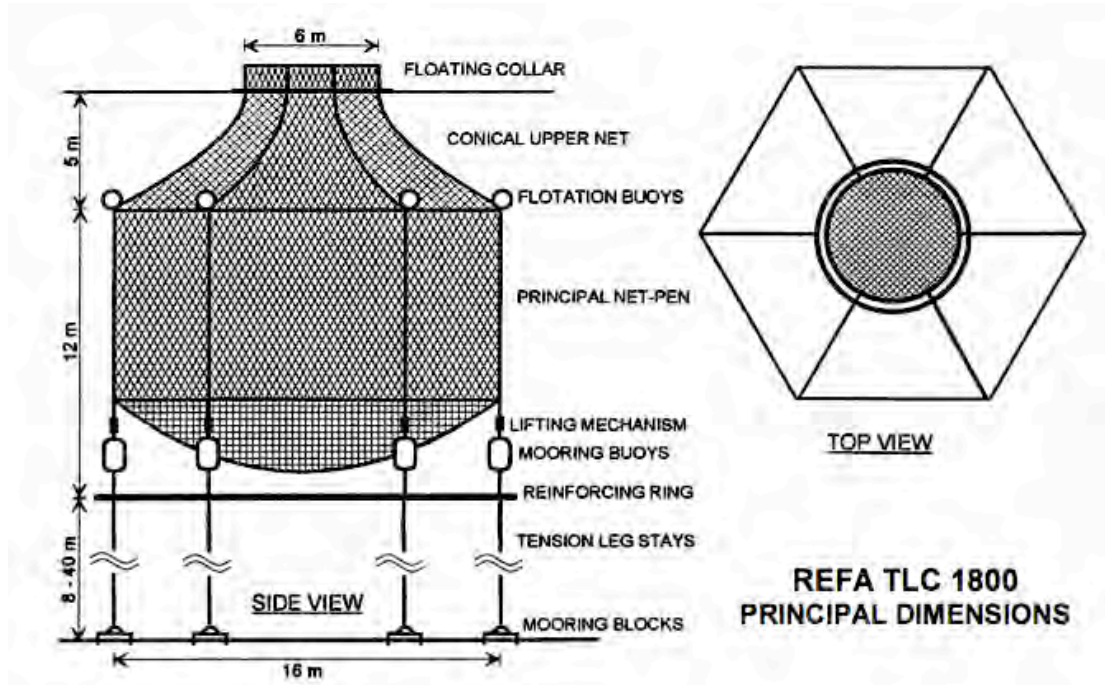


Figure 18. Ocean Spar System – The rigid system allowed the cage to be submersed when needed. The system is still in its trial stages (Lisac, 1997).

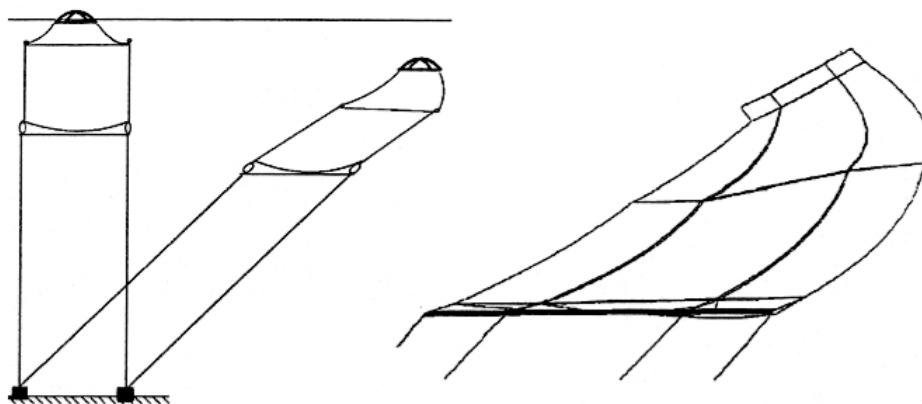


Figure 19. Tension Leg System – The cage and mooring are shown in profile; the cage is at the surface during normal ocean conditions (left) and submerges and deflects high wave and current conditions without intervention (right two drawings) (Lisac, 1997).

A series of offshore seaweed cultivation platforms were tested in the North Sea (Buck & Buchholz, 2004) including a long-line system that allows for integration of multiple aquaculture species (Figure 20). The system that proved most successful at growing *Laminaria* and surviving inclement ocean conditions was the offshore ring developed in Germany (Buck & Buchholz, 2004). The offshore ring system consists of a submerged ring with culture lines descending from it, surface flotation and anchoring system (Figures 21 and 22). The rigging is adjusted to maintain the growing lines at one to 1 ½ meters below the surface at the optimum light depth for growth. This system continues to undergo tests and improvements and should be watched as a potential large-scale seaweed cultivation platform (Reith, 2005).

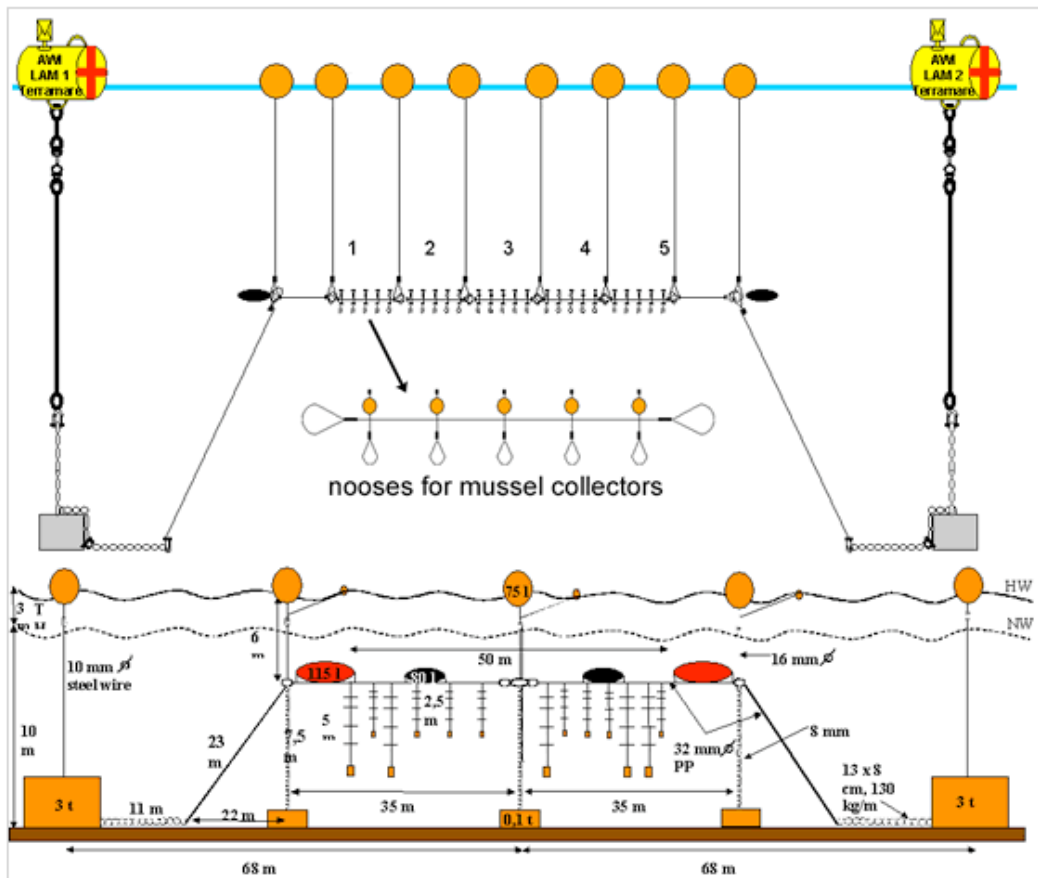


Figure 20. Long Line System. The system allows for culture of shellfish (in mussel collectors) as well as seaweed growing on ropes suspended from the surface line (Buck & Smetacek, 2006).

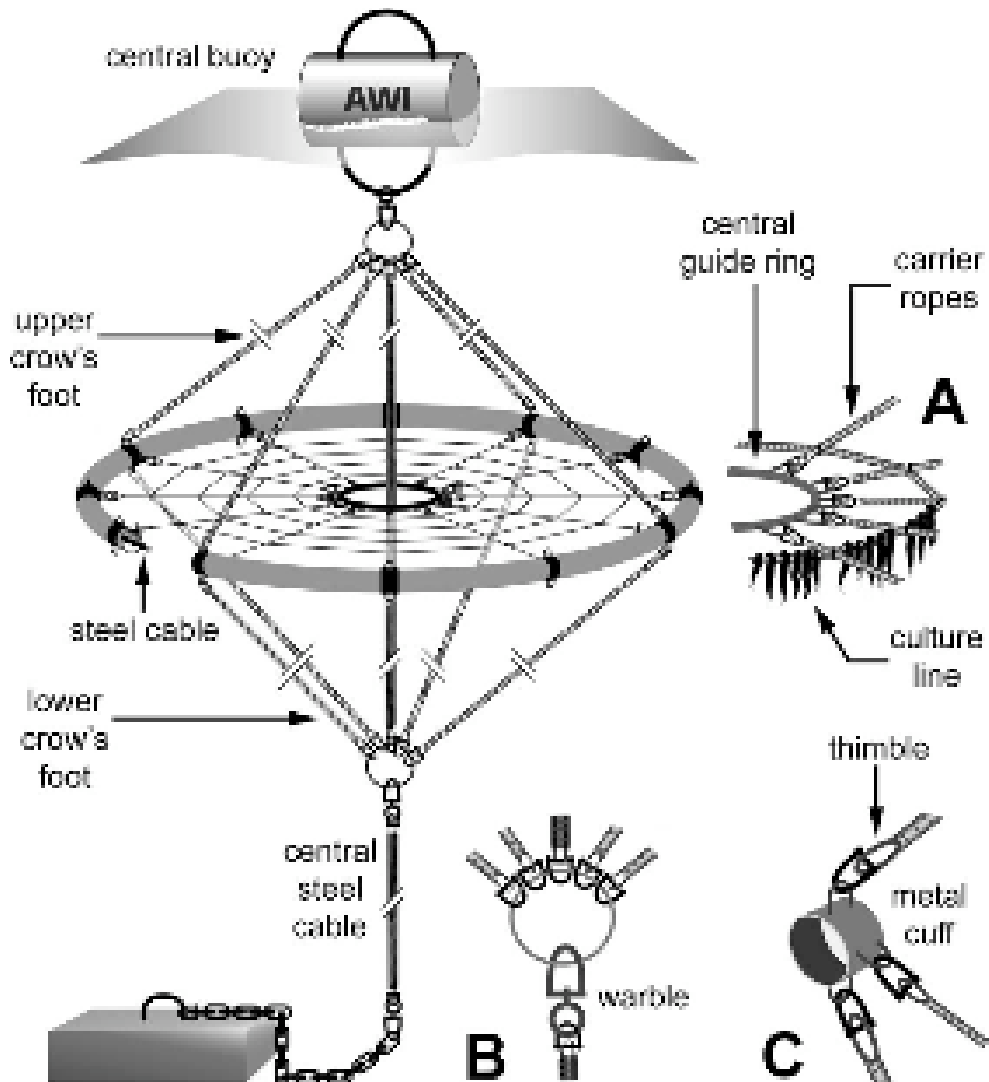


Figure 21. Offshore Ring #1 – The ring system can be completed rigged on-shore then towed to the location and anchored, decreasing the need for costly construction at sea (Buck & Buchholz, 2004).



Figure 22. Offshore Ring System #2 – *Laminaria* grown on ring system in North Sea (Buck & Smetacek, 2006).

Integrated aquaculture operations

Most fish farms are located close to the coastline, usually in sheltered bays and estuaries. Increasingly, pressure has been mounting to move operations offshore where the effects of land-based contaminants are lessened, and negative perceptions of aquaculture operations as polluting, threatening to native species, and resulting in unpleasant sights, odors and industrial activity, can be avoided.

Offshore platforms for culturing fish have taken two forms – scaled-up net pen operations that resemble those nearshore, and large cages that can be lowered below the ocean surface. Several integrated systems have been proposed and operated at a small scale to combine seaweed aquaculture and fish rearing (Reith, 2005). In most cases the impetus for the integration came as a means of cleaning the waste products (often nutrients) from the fish farms; these nitrogenous wastes provide seaweed cultivation operations with much needed nutrients when the seaweed operation is placed downstream but close to fish net pens (Troell & N. Kautsky, 1997). While these operations have been most successful nearshore and in land-based systems, there is reason to examine the idea of an integrated system for offshore areas as well (Reith, 2005).

Different groups of seaweeds have differing light requirements, so that green, red and brown seaweeds can be grown at differing depths in a seaweed farm with green algae grown closest to the surface and red and brown algae thriving at deeper depths, allowing for integration of crops intended for different purposes through a layered growth of algae (Figure 23).

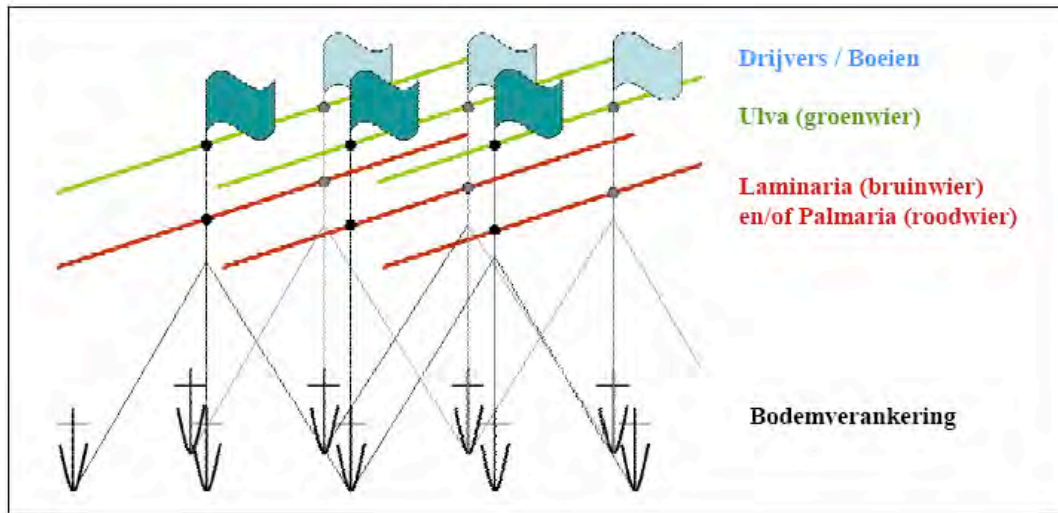


Figure 23. Layered Growth of Algae – Different groups of seaweed can be grown at different depths in response to differing light levels, with green (groenwier) near the surface and brown (bruinwier) and red (roodwier) deeper (Reith, 2005).

Aquaculture operations in conjunction with wind farms and other infrastructure

The challenges of withstanding severe conditions in the open ocean, combined with the need to produce energy from alternative sources, have led the EU to examine the efficacy of combining aquaculture operations with offshore wind farms (Reith, 2005); (Buck & Buchholz, 2004). The large infrastructure needed for siting wind farms in the ocean could provide excellent anchorage and protection for integrated aquaculture operations which might include seaweed cultivation as well as raising of shellfish and finfish (Reith, 2005). A simple version of an integrated farm is shown in Figure 24, depicting the Wind Farm with Ring concept. Such combined structures would lower investment and maintenance costs for the seaweed operation. Aquaculture operations might also help to alleviate public concerns about the installation of wind farms in areas where fears of conflict with fisheries occur (Buck & Buchholz, 2004). Infrastructure for wind farms would also allow for the cultivation of several species of seaweed at different depths, under different light regimes, as well as cages or platforms for small batches of specialized marine products (Reith, 2005). Conceptual models of a wind farm/aquaculture operation have been considered in an integrated offshore farm (Figure 25).

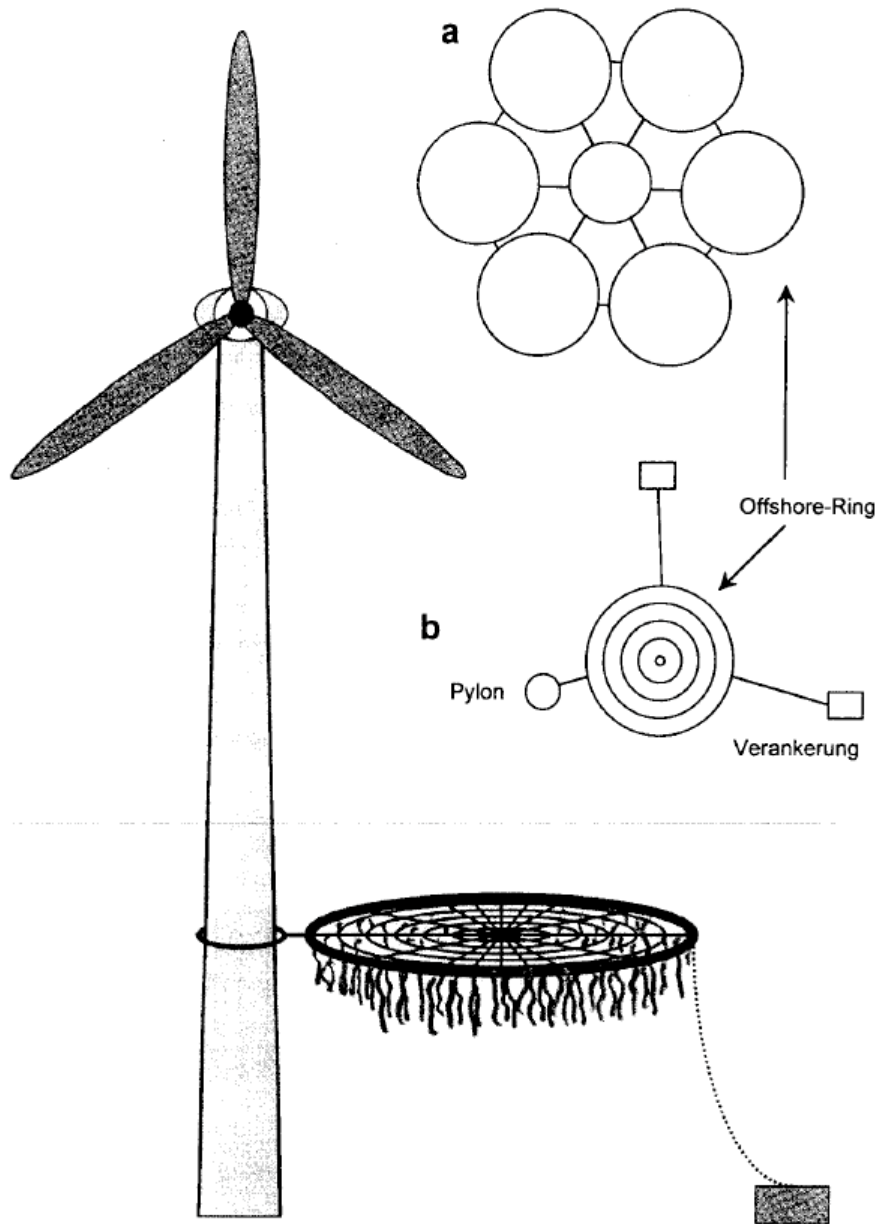


Figure 24. Wind Farm with Ring – The infrastructure needed for a wind farm could easily accommodate additional structures such as seaweed cultivation platforms (Reith, 2005).

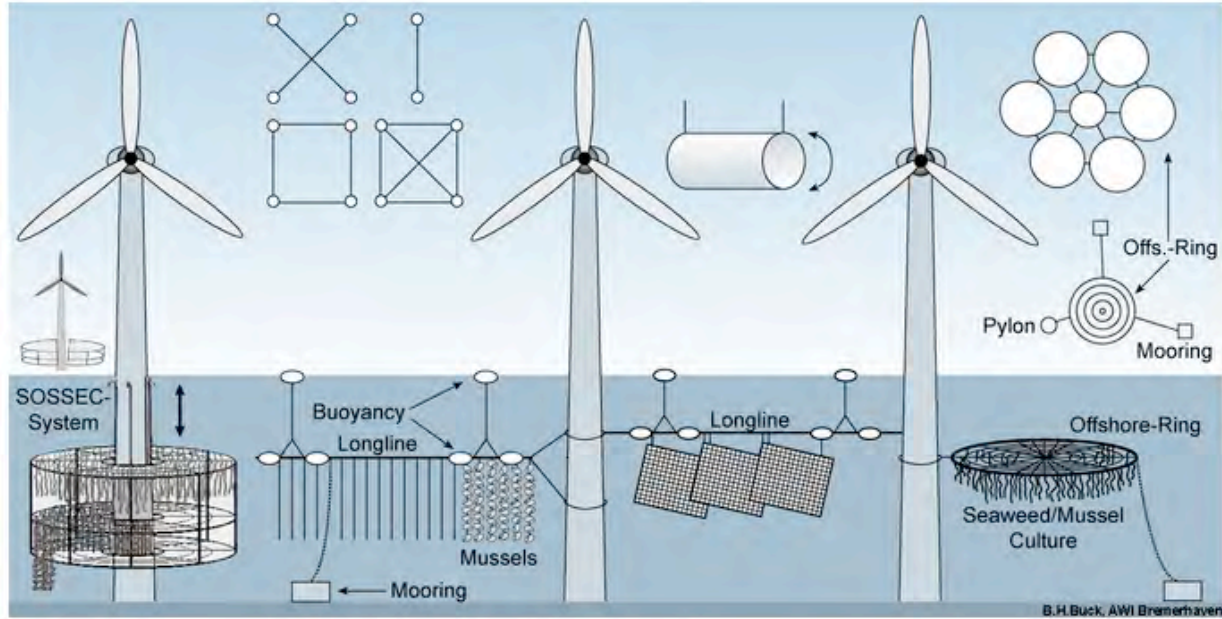


Figure 25. Integrated Offshore Farm – Potential multifunctional use of fixed underwater structures for wind farms. Many other configurations for aquaculture operations are possible within this configuration, including cultivation of seaweed on ropes, and submersible cages and rings for growth (Buck *et al.*, 2006).

Selection of the seaweed species for culture

Systematic examination of seaweed species that could be suitable for large-scale biomass production in offshore waters was already undertaken as part of the Marine Biomass Program (Show *et al.*, 1979). By evaluating each candidate species against a set of desired criteria, combined with empirical observations, seaweed species best suited for pilot scale trials were identified. The following criteria should be considered:

- Organic matter yield per unit area, annual
- Growth sensitivity to plant spacing
- Dependence on substrate and substrate depth, or free floating
- Susceptibility to disease, grazing and epiphytes
- Simplicity with which a species can be propagated
- Nutrient requirements

- Ability to take up and store nutrients for subsequent use
- Harvestable by part-cutting rather than removal of the whole plant.
- General robustness – tolerance to variable physical conditions
- Water and ash content
- Calorific content, and yield of methane on digestion
- Bound nitrogen (protein) concentration and extractability
- Concentration of other co and byproducts of value
- Variability in composition, e.g. with season
- Sulfur concentration (high S result in high H₂S in the digester gas, a major issue).

CHAPTER 6

ENVIRONMENTAL FACTORS THAT AFFECT OFFSHORE SEAWEED FARMING

Physical and chemical limitations to production

Seaweeds grow in the surface waters of the ocean, to depths where light penetrates, which can be from a few meters in coastal waters to 100 m in clear open oceans. The holdfast of plants such as kelp does not photosynthesize and may grow to depths below the sunlit layer. The plants are unaffected by wind, ocean waves and currents, except under severe storm conditions, when fronds may tear and separate from their holdfasts. In order to successfully grow seaweed offshore, a platform must be sufficiently robust to withstand severe oceanic conditions, while minimizing interference with sunlight reaching the plants, and allowing maximum water flow to the plants, to ensure delivery of dissolved nutrients and removal of plant waste products. Achieving all these requirements is a major challenge.

Sunlight at the surface of the ocean is often too bright for optimum plant growth, requiring that the seaweed to be held at some depth, often a meter or so. On the other hand, plants may be shaded by heavy growth of surrounding plants and the topmost layer of leaves will shade the deeper leaves. Ambient light is often too high for optimum growth of seaweeds, microalgae and terrestrial plants, particularly at low latitudes. As sunlight penetrates the ocean surface it is attenuated, with certain wavelengths absorbed first. Intense surface light, mutual shading, the inability of plants to make use of all incident sunlight, and light absorption by the water column all conspire to reduce the productivity that can be achieved in real production processes to well below that which is anticipated from theoretical projections, laboratory experiments or small-scale controlled plots. Different species of seaweeds have unique growth responses to light levels, which must be considered in the design a production system.

An even greater limitation to seaweed culture is the need for sufficient water flow across the blades of the plants. The plants require carbon, either in the form of CO₂ or bicarbonate, and this must be provided by seawater. Seawater contains about 2.3 meq of alkalinity, at pH near 8. This means that the plants cannot obtain more than about 0.5 mM of CO₂ from seawater, or 6 mg/l of C, without raising the pH to excessive levels (e.g. near 9.0) at which point photosynthesis would slow. A cubic meter of seawater would thus provide about 6 g of C, and since the plants would extend through several meters of depth, this would be sufficient to support the maximum

projected productivities, of about 10 to 20 g (ash free dry weight) biomass per m² of surface per day (g/m²-d).

However, a problem arises from the very slow diffusion of CO₂ (and bicarbonate) through the water, about 10,000 times lower than in air. This results in severe diffusion barriers, which need to be overcome by relatively fast water flow past the blades. Experimental work by Neushul, North, and many others, has demonstrated that only in rapidly flowing water, will the productivity of seaweeds be maximized. Natural meadows and plots of nearshore seaweed are generally anchored in areas where waves and currents create turbulence, providing adequate passage of concentrations of dissolved carbon and nutrients past the plants. Open ocean water, with few exceptions, moves with adequate speed to supply dissolved gases and nutrients to plants, even at high densities expected in offshore farms. Siting of farms in areas that provide adequate water flow, for example, areas of upwelling, ubiquitous current flow, and mild shear forces (over doldrums and slow-moving ocean gyres) is a prime consideration.

Due to the high capital cost of offshore or onshore containment systems, high levels of plant productivity must be achieved through growing plants at high densities. The placement of dense farms in the ocean will tend to slow movement of ocean waters, increasing demand for high water flow to ensure adequate diffusion of gases and dissolved substances like carbon dioxide and nutrients. This problem was not addressed by the Marine Biomass Program, due to the very small size of the pilot farms, and has not been addressed to any extent by the relatively few onshore seaweed production facilities.

Under natural conditions, a major limitation to seaweed growth is the concentration of nitrogen and other nutrients that can be obtained from seawater flowing past the growing plants. Temperate shallow ocean water typically has abundant nutrients in winter, but generally suffers from lack of nutrients in the warmer summer months. Shallow tropical waters are commonly very low in nutrients year-round. Deeper ocean water generally contains very high levels of dissolved nutrients. In addition there are coastal areas, typically on the western margin of the continents and some polar areas, where deep water upwells to the surface for much of the year, bringing with it abundant dissolved nutrients. Seaweed growth in large-scale cultivation facilities may be limited by nutrient availability in most open ocean areas, in particular tropical seas. Lack of adequate nutrients prompted Chinese seaweed farms and the Marine Biomass Program to develop processes for nutrient enrichment.

The concentration at which dissolved nitrogen and phosphorus become limiting to growth of *M. pyrifera* blade tissues are 1% and 0.2% of dry weight, respectively (Gerard, 1987). Based on this estimate, a minimum of 10 kg of N and 2 kg of P must be assimilated for each dry metric ton of biomass; maximum productivity will require approximately twice that amount.

There are many other trace chemicals found in seawater that are essential for the growth of seaweed. Most are found abundantly; however intensive cultivation of seaweed in enclosed areas would result in depletion of one or more of these chemicals, resulting in the need to augment the plants for optimum growth. However, this should not be a major problem for open ocean farms, provided adequate supplies of carbon, nitrogen and phosphorus are supplied.

Biological limitations – disease, predators, and epiphytes

Biological interactions are common with plants growing at sea, most notably disease, predation (grazing), and colonization of seaweed fronds by microalgae and smaller seaweeds (epiphytes). These interactions can affect the yield and survivability of plants under cultivation.

Intensive farming of seaweeds, like any domesticated crop, can encourage disease organisms to flourish. The greater the number of farms and concentration of plants, the greater number of diseases may be found. Disease has occasionally been widespread in Chinese *Laminaria* (kelp) farms, reducing yields in various regions of China (FAO, 1989). The major diseases, and their causes and effects, are shown in Table 8.

This list is not exhaustive and includes pathogenic and environmental effects on a single seaweed type. However, the implications are a clear warning that must apply to all cultivation operations. Poor environmental conditions lead to increased disease susceptibility, necessitating optimum selection of growing areas to reduce the risk of loss due to disease.

Numerous organisms such as sea urchins and herbivorous fish graze on seaweeds and are likely to create problems in small-scale cultures or for slow-growing seaweed species (North, 1987). For example, grazing by large half-moon perch destroyed the experimental kelp plants within a few days at one experimental Marine Biomass Program location in California (North, 1987). Larger farms are less likely to suffer widespread losses as overall plant productivity will greatly exceed grazing demand, and the damage from grazing will become negligible. For slower growing seaweed species ‘*Spinosum*’ and ‘*Cottonii*’, rabbitfish (*Siganus sp.*) have been seen to

Table 8. The most common diseases affecting *Laminaria* in Chinese farms (FAO, 1989)

Disease	Cause
<u>Environmental etiology</u>	
Green rot disease	Poor illumination
White rot disease	Change in transparency + insufficient nutrients
Blister disease	Freshwater mixing with seawater after heavy rainfalls
Twisted blade disease	Excessive illumination
<u>Pathogenic etiology</u>	
Malformation diseases	Hydrogen sulfide + sulfate reducing and saprophytic bacterial, e.g. <i>Macrococcus</i>
Sporeling detachment disease	Decomposing <i>Pseudomonas</i> bacteria
Twisted frond disease	Mycoplasma-like organisms

nip the growing tips of the seaweed thallus, reducing the plant growth for a week or more until the plant heals itself (Ask, 2006).

Seaweeds fronds provide an excellent substrate for epiphytes (small, usually unicellular algae) and encrusting organisms to grow. The epiphytes tend to shade the seaweed fronds from sunlight, thereby reducing the overall farm productivity (Lüning & Pang, 2003). Slow growing seaweeds, such as red algae, are particularly susceptible. Large farms that grow seaweeds in dense quantities are less likely to suffer severe productivity losses from epiphytes.

CHAPTER 7

ENVIRONMENTAL IMPACTS OF LARGE-SCALE OFFSHORE SEAWEED AQUACULTURE INSTALLATIONS

Conflicts between aquaculture operations and other coastal uses are common and generally revolve around competing use of space, and fears of environmental degradation resulting from construction and operation of facilities. Some fears of degradation are warranted, others are the result of perceptions created due to poor farming practices. Marine aquaculture operations have a history of impacting local environments and causing harm to native species and habitats. The public perception of these impacts is often far greater than the reality, but there are sufficient examples of damage and breakdowns in native systems to keep the public and decision-makers alert to potential impacts.

Current issues with nearshore marine aquaculture

Environmental impacts from marine aquaculture operations are largely associated with poorly sited and poorly maintained farms close to the coast. Typical concerns about coastal aquaculture operations, largely associated with nearshore animal aquaculture, include factors such as the following:

- Damage to coastal habitats due to physical presence of the farm;
- Contamination from excess feed and waste products from animals reared in farms;
- Introduction of antibiotics and other contaminants into the environment from farmed animal feed and waste;
- Spread of disease from farmed animals or plants to wild marine organisms;
- Introduction of invasive species from escapes;
- Release of reproductive materials;
- Hybridization of farmed animals or plants with native stocks; and
- Establishment of feral populations of farmed organisms through escape from culture facilities.

Additional concerns associated with nearshore aquaculture include aesthetic considerations such as the impact on views, and odors, noise and lights from operations. Competition for scarce

stretches of shoreline with ports, marinas, recreational facilities, and commercial water-dependent operations, can also create tensions for siting aquaculture operations.

Potential consequences of offshore culture of seaweeds

While locating installations in the open ocean avoids issues specifically pertaining to nearshore coastal locations, additional environmental impacts can arise in the open ocean:

- Interference with marine navigation, including shipping, commercial fishing, and recreational boating;
- Interruption of marine mammal migration routes and feeding activities; attraction and entanglement of migrating seabirds;
- Effects on native species from discharge of nutrients and chemicals used to treat seaweeds, including fertilizers. Changes in native species surrounding aquaculture operations include loss of species diversity, particularly sensitive genera like sea urchins, snails, and delicate sea stars, with an increase in more pollution-tolerant groups like tube worms;
- Threats to marine organisms due to loss of plastics, small amounts of rigging, and other materials from the farms (for example, sea turtles die in large numbers from ingesting plastic bags, mistaking them for jellies);
- Shading of native plants in the region of the farms, particularly phytoplankton. This change may affect organisms over a wide area as the base of the food web is changed, resulting in changes in species diversity, and abundance of larger organisms such as fish;
- Breakage of seaweed fronds that may wash ashore, smother native habitats and species, or become established (only a problem if non-native seaweeds are cultured);
- Loss of rigging, gear or platforms during storms that may cause damage to shipping, shore-side facilities, shoreline development or other property, human life, as well as native habitats and shorelines;
- Free-floating farms may spread reproductive products and plant material, causing the spread of species and disease over large areas of the ocean;
- Anchored farms may damage sensitive benthic habitats such as coral heads;

- Release of reproductive products from seaweed that may become established in the wild (only a problem if non-native species are cultured).

Potential decrease in ocean productivity due to offshore seaweed farming

Offshore seaweed culture, through photosynthesis and seaweed growth, will remove large quantities of dissolved forms of carbon, nitrogen, and phosphorus, as well as trace amounts of metals, chelated organics and other compounds that might otherwise support the marine food web. In order to assess potential ecological impacts of offshore seaweed culture on the productivity of the marine food web, and potential impacts on higher trophic levels such as marine fisheries, it is necessary to understand the magnitude of supply of each of these constituents.

Carbon dioxide is found in huge quantities in all surface ocean water, and is constantly replenished from exchange with the atmosphere. As global carbon dioxide levels continue to rise in conjunction with fossil fuel combustion and climate change, the supply of carbon to seawater will increase over time, unless the source is abated. Offshore farms would not be expected to reduce the supply of carbon to the marine food web. Any seaweed-related increase in demand would be expected to shift the net CO₂ flux to the oceans to one of greater absorption of atmospheric CO₂; estimates of the amount of carbon fixed by seaweed worldwide range up to 10⁹ ton of carbon per year (Smith, 1981). Use of seaweeds as a bioenergy feedstock represents, in principle, a carbon neutral contribution to the energy economy, releasing as much as is taken up. The operation of ocean seaweed farms and related infrastructure, however, can contribute to the production of greenhouse gases, and this should be considered in the overall carbon benefits of seaweed farming.

Dissolved nitrogen compounds, including nitrate, nitrite, ammonia, creatine, and urea, are commonly the limiting factor to growth in the surface waters of the ocean. Dissolved phosphorus, metals and other micronutrients remain in near-constant ratios with nitrogen levels throughout the world oceans.

Overall the world's oceans have an average dissolved nitrogen concentration of almost 500 g nitrogen per m³ in the deep ocean and, conservatively, 15 g nitrogen per m³ in the surface layer (upper 100m). Nitrogen fixation alone is not sufficient to maintain high productivities in the ocean surface waters, and replenishment of surface-utilized nitrogen must come from deep ocean

waters or from land-based sources. If offshore seaweed farms were to cover approximately 10,000 km² (1,000,000 hectares), with an expected yield of 1000 metric ton of seaweed per km² per year, and an average ash-free dry weight (AFDW) of 3% nitrogen, we would expect that the farms would fix and remove from the oceans approximately 300,000 metric ton of nitrogen per year. The surface oceans contain almost 2×10^{15} metric ton of nitrogen in a form usable by seaweed; the potential removal by offshore seaweed farms envisioned here is negligible in the world oceans. Localized nitrogen removal may be more significant; siting offshore seaweed farms in areas of active upwelling should minimize potential declines in nitrogen availability to the marine food web.

Optimum species selection and potential conflict with native/non-native and genetically modified organisms

There are production and economic issues to be taken into account when choosing one or more species for large-scale energy production. Agriculture has thrived throughout time by the introduction of non-native species. Such species have not co-evolved with the other native species in the habitats into which they are introduced and, in some cases, have a competitive advantage because they have no natural predators or competitors. The species adapted for agriculture are carefully bred and maintained to take advantage of pest resistance, to produce optimum yields, and to prevent inter-breeding with undesirable native species. With the advent of genetic modification, ever more specialized and successful agricultural systems are being developed.

While it may appear that similar gains could be realized by introducing non-native species of seaweed to obtain optimum yields and to resist disease and epiphyte growth, the potential harm to native species and habitats have led many countries to enact laws that prevent or regulate the introduction of potentially invasive marine species into their waters. Distinguishing which non-native species has the potential to become invasive is very difficult; often the answer is only discovered after an uncontrollable invasion has occurred. This has led to public perception and government response that cultivation of any non-native species not be allowed. It may prove very expensive to test each potential non-native seaweed for invasive potential. However, as in agriculture, non-native seaweed species, including genetically modified strains, may possess attractive attributes with regard to cultivability or increased production of biofuels or other

biobased products. In light of the growing need for new biomass to meet the needs of a variety of human activities, the relative merits of production potential and potential for undesirable ecological consequences will need thoughtful consideration.

In the case of open ocean farms, the definition of native species becomes more tenuous. There may be no native species occupying the niche of the farmed seaweeds in areas where these farms might be deployed. While the chance of cultured plants escaping and proliferating in such an environment may be small, storms and currents could allow a viable frond to travel to a far off shore and create an invasive opportunity.

CHAPTER 8

SEAWEED BIOTECHNOLOGY

Genetics and breeding seaweeds

The propagation of seaweeds for commercial purposes has several underlying considerations, the most fundamental of which is species selection based on application as food or other production possibility such as carrageenan, alginate, or biomass for energy. Strain selection within a species maximizes properties such as growth, disease resistance, and product quality. Strain selection and propagation in seaweeds has been dependent on taking control of life cycles of individual seaweed species or strain and focusing effort on manipulating critical stages in hatchery settings. Artificial selection pressure can be exerted by altering environmental conditions then selecting for robust individuals or by choosing individuals with the desirable phenotype for further propagation. Both sexual and asexual phases of seaweeds life cycles have been used as sources of propagules. All three divisions of macroalgae, i.e., brown, red, and green, have been the subject of such propagation, although of cultivated forms, the red and brown are dominant. Further details are provided below on *Laminaria* and *Porphyra*, two of the most important economically, as examples. Both are intensively farmed.

The culture of the kelp *Laminaria japonica* in China provides an interesting case history of the evolution of the seaweed industry from one dependent on imported dry product from Japan and northern Korea to one based on cultivation of domestic kelp. While it has been consumed in China for about 1000 years (Tseng, 1987), *L. japonica* is not native to China, having been accidentally introduced from Japan in 1927 to establish a so-called “wild stock” in Northern China (north of 36° N latitude) (Scoggan *et al.*, 1989). *L. japonica* is the only species of *Laminaria* in China. Prior to this introduction, the warm water of the Yellow Sea served as a physical barrier that restricted dispersal of this kelp to the northern Chinese coastline (Bruinkhuis *et al.*, 1987). Its range is naturally precluded in more southern latitudes due to its intolerance of elevated temperatures. The species now grows naturally or under cultivation in five northern provinces in China. Additionally, through transplantation and growth of artificial seed stock from northern populations, its range has been extended south to 25° N latitude in regions in which it will not reproduce. *L. japonica* is now considered one of the mainstays of the Chinese seaweed

cultivation industry (Wu & Lin, 1987), with production reported in 1999 at 4,500,000 ton wet weight (McHugh, 2003).

Up to the early 1950s, *Laminaria* cultivation was based on bottom culture using stones thrown into water of appropriate depth and relied on naturally seeding by nearby sporophytes (Bruinkhuis *et al.*, 1987). Subsequently, floating raft culture using artificially seeded hanging ropes was implemented in 1952 (Bruinkhuis *et al.*, 1987).

Commercial cultivation is based on the indoor cultivation of sporelings and the outdoor cultivation of the macroscopic form (Tseng, 1987) through control of the kelp life cycle (see life cycle in Figure 26). In indoor cultivation systems, sporelings are grown from microscopic haploid zoospores that are captured on ropes attached to frames and subsequently germinate, resulting in fertilization of oogonia that are attached to the female gametophytes. The haploid

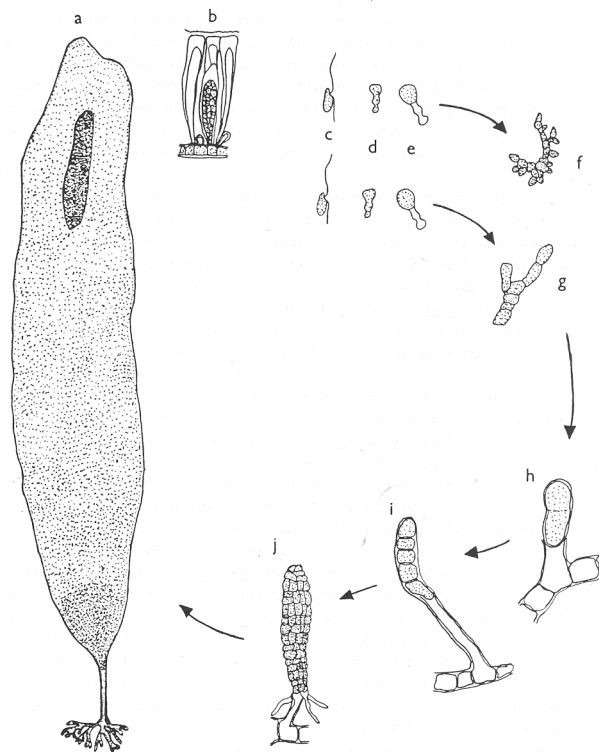


Figure 26. *Laminaria saccharina* life cycle. Mature thallus with sori patch (a) with sori containing haploid meiospores (b), which release “swarmers” or zoospores (c,d,e) that develop into haploid male (f) and female (g) gametophytes. Sperm from the male gametophyte fertilize oogonia attached to the female gametophyte. The diploid zygote (h) develops into the sporophyte (h,i,j as developing sporelings) (Tseng, 1987).

phase is of short duration, about two weeks. The sporelings attached to the ropes are subsequently transferred to floating racks in the field for growth to the macroscopic stage. “The kelp floating raft cultivation method” is the mainstay of the Chinese kelp industry (Tseng, 1987) and is based on an understanding of the *Laminaria* life cycle.

Starting in the late 1950s, Chinese geneticists developed two strains of *L. japonica* selected for improved growth and higher biomass at elevated temperatures and higher iodine content (Wu & Lin, 1987). The heterozygous nature of kelp growing in the wild facilitated the selection of existing genotypes conferring traits suitable for cultivation. These strains are widely adopted by the kelp cultivation industry in North China (Wu & Lin, 1987). In the late 1970s, male and female gametophytes were successfully cloned, enabling the production of hybrid sporophytes with improved traits. In this manner, genetic manipulation by crossing the haploid phase of the life cycle has also had application in seaweed cultivation in China.

Porphyra has been utilized in Japan and China for over a 1000 years, but commercial cultivation began just a few centuries ago: in Japan around 1600 through insertion of bamboo twigs into the bottom to allow settlement and growth of spores (Tamura, 1966), and in China, more than 200 years ago through clearing rocks to allow attachment and growth prior to the mass liberation of the spores from natural *Porphyra* beds. It was not until the mid-20th century that a science-based approach to the cultivation of this seaweed became possible. This followed the discovery that the conchocelis is a life history stage of this genera (Drew, 1954) and that the conchospore released from the conchocelis is the propagule that develops into the thallus (Tseng, 2001). Commercial cultivation methods reached modern standards in the 1960's with incorporation of the artificial collection of conchospores in the production cycle (Figure 27).

With the advent of the use of the conchocelis-spore culture technique, the predominant species for cultivation changed from *P. tenera* to *P. yezoensis* because the latter could withstand higher salinities (Wildman, 1971). The conchocelis is also preferred for use as brood stock (Tseng, 2001) *Porphyra* cultivation in Japan is a \$1.5 billion (USD) aquaculture industry with an average production of 400,000 metric ton wet weight per year (circa 1999) (McHugh, 2003). In

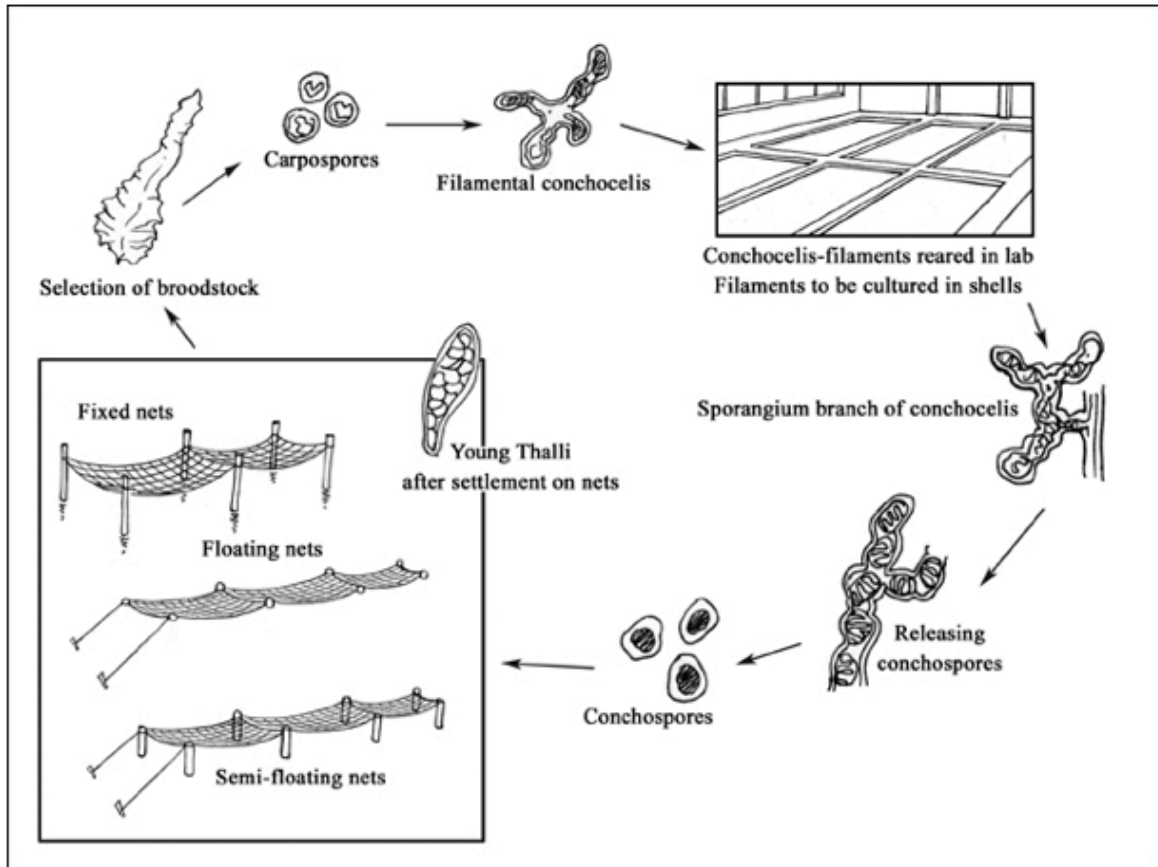


Figure 27. Production cycle for *Porphyra* culture. Conchocelis filaments are raised in the laboratory and allowed to attach to shells. Conchospores released by conchocelis are seeded onto cultivation nets, which, after suitable growth of young thalli, are placed in the environment for maturation of thalli.

China, the *Porphyra* industry is second to that of *Laminaria* with about 210,000 metric ton wet weight production; while in Korea 270,000 metric ton are produced. *Porphyra* has the highest value when compared to other cultivated seaweeds (McHugh, 2003). Kathleen Drew-Baker, the discoverer of the conchocelis, is honored by a statue and shrine erected by fishermen at Kumamoto, Japan.

Vegetative approaches to propagation

The forgoing account points out the importance of understanding the life cycle of macroalgae and manipulating genetics for successful cultivation. Life cycles can be complex,

and both sexual and asexual stages need to be carefully considered in developing breeding and selection strategies. The specific examples using *Laminaria* and *Porphyra* reflect approaches relevant to other types of seaweeds.

Alternate approaches to propagation exist in the exploitation of vegetative paths to regenerating seaweeds, taking advantage of the natural tendency of a plant to reproduce asexually. The progeny of asexual propagation are genetically identical to the parent plant. Asexual looping, described above, enables *Porphyra* to regenerate blades directly from spores produced by blades. Blade archeospores and conchocelis archeospores can germinate to become new blades or conchocelis, respectively, and endosporangia can produce spores that produce new blades (Nelson *et al.*, 1999; Nelson & Knight, 1995). Recent studies have shown that the haploid blade archeocytes, which are more resistant to antibiotics than the conchocelis, may be more suitable as seed stock (Choi *et al.*, 2002). Furthermore, tissue fragments and cultured tissues derived from blades can regenerate directly into blades and rhizoids, leading to the proposal that cultured blade tissues be considered as a seed stock for *Porphyra* (Notoya, 1999). The use of tissue fragments to seed seaweed beds is a common form of vegetative propagation used in the seaweed industry. In this section, focus is placed on use of tissue explants or fragments, the most basic method of vegetative propagation.

Dispersal by regeneration of thallus fragments is a natural form of population growth in brown, red, and green seaweeds (Hiraoka *et al.*, 2004; Rodriguez, 1996; Uchida, 1995). So, the use of thallus fragmentation in seaweed cultivation is the most basic method of vegetative propagation and is based on biological precedent. Fragments can grow faster than spores or other types of microscopic propagule (Meneses & Santelices, 1999), and strain selection is facilitated by this approach since the characteristics of the donor plant is replicated in progeny regenerated from fragments (Meneses & Santelices, 1999). The ability to regenerate from thallus fragments is characteristic of clonal seaweeds (Santelices, 2001) and include commercially important seaweeds such as *Gelidium* and *Gracilaria* (Santelices, 2001). Clonal seaweeds can produce multiple fronds on a single holdfast, while unitary seaweeds produce only one from a holdfast (Scrosati, 2006). Clonal forms grow and propagate by replicating genetically identical units, following natural or experimental fragmentation into pieces; unitary seaweeds lack this capacity (Santelices, 1999b). The kelps and *Porphyra* are classified as unitary and must be grown from spores, rather than from fragments (Santelices, 1999a), although the pluripotency of blades of

Porphyra and their ability to grow from fragments and dissociated cells is suggested from the work of Notoya (Notoya, 1999) and Polner-Fuller et al. (Polner-Fuller & Gibor, 1984).

Clonal seaweeds are amenable to one-step farming, while unitary species require two- or multi-step farming plus nursery facilities for collecting and germinating spores (Figure 28).

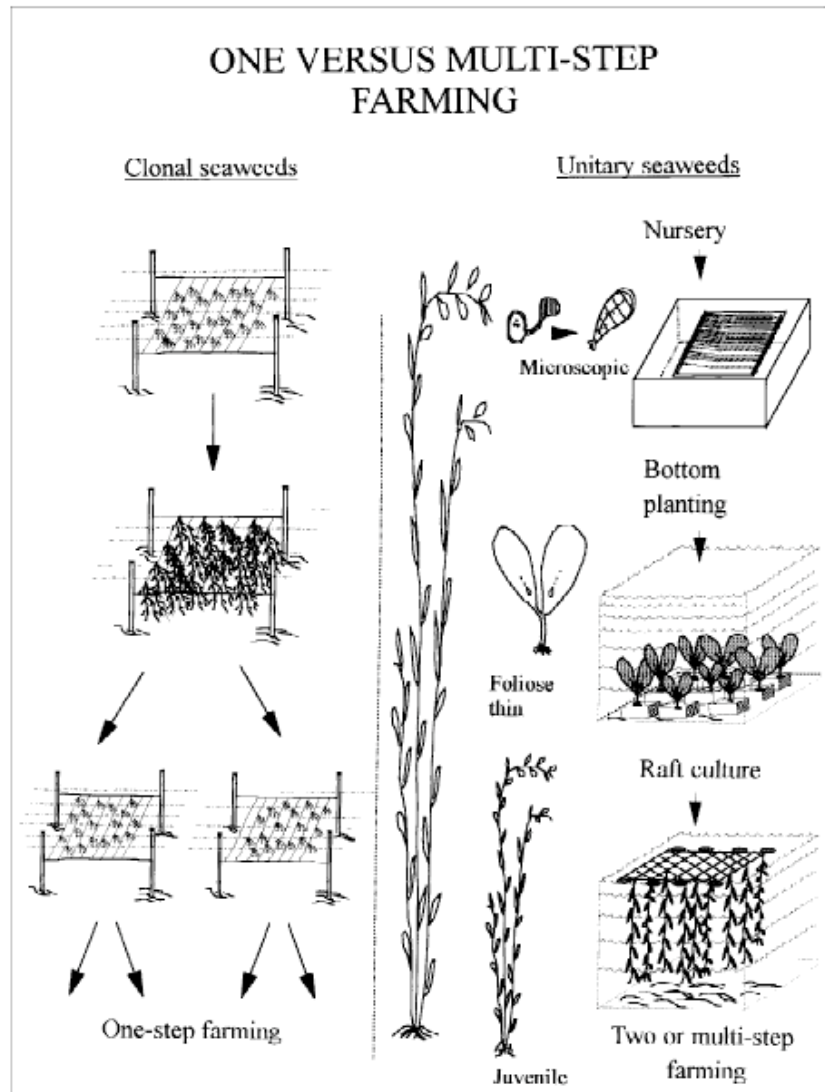


Figure 28. Depiction of one step versus multi-step farming for clonal and unitary seaweeds, respectively (Santelices, 1999a).

Thus, the biology of the species dictates farming practice. While the propagation of clonal seaweeds is, in principle, straightforward, species differences make some species difficult to farm. As a result, commercial harvests of *Gelidium*, the source of bacteriological grade agar and

agarose for the biomedical science community, is dependent on collection from fragile natural populations and the cyclic nature of natural *Gelidium* productivity (Titlyanov *et al.*, 2006). This has prompted attempts to develop artificial propagation techniques for this species (Titlyanov *et al.*, 2006), an approach yet to be commercially viable, and a turn to *Gracilaria*, a genus more receptive to vegetative propagation, as an alternative source of agar (Hansen, 1984). *Gracilaria* is harvested from both natural and cultivated sources (McHugh, 2003). Explants derived from thallus fragments are major seed source for the farming of *Gracilaria* (Santelices & Varela, 1995). Thallus fragments, explants, and juvenile blades also serve as a source of spores in various species that include *Laminaria*, *Porphyra*, and *Sargassum* (Choi *et al.*, 2002; Hwang *et al.*, 2006; Li *et al.*, 1999).

Advances in seaweed tissue and cell culture

Obtaining sufficient amounts of selected strains for commercial cultivation has been problematic (Renn, 1997) and points to a need for new approaches to seaweed propagation. Plant tissue culture is a more recent and advanced method routinely used for propagating higher plants. Modern tools developed in the larger plant breeding community are now available to seaweed biologists and culturalists to advance the vegetative propagation of seaweeds through cell and tissue culture techniques. The interest in seaweed calluses and protoplasts extends the potential of tissue fragments and explants from seaweed sources to the microscale.

Calluses are aggregates of undifferentiated plant cells that are formed at sites of wounds and also formed in tissue culture. Calluses can be induced to differentiate into plantlets. Protoplasts are plant cells whose outer cell wall has been removed by various artificial means. They can divide to become specific tissue types or germinate into plantlets. Protoplasts can also fuse with other protoplasts resulting in genetic recombination that creates desirable traits. Both calluses and protoplasts can be stored as germ stock and subsequently induced to differentiate into plantlets for growth into mature plants. The interest in calluses and protoplasts in seaweeds is several fold: 1) seed stock for cultivation of seaweeds, 2) germ plasm storage, 3) direct production of highly valued product such as phycocolloids and other substances, 4) genetic recombination through protoplast fusion, and 5) vehicle for transgenic seaweeds through transformation with novel genes. Although the field is still at an early stage of development, the micropropagation of plants is a concept adopted by seaweed biologists, and the number of

studies on the formation and regeneration of calluses and protoplasts in seaweeds is increasing (Garcia-Reina *et al.*, 1991).

For seaweeds, a consensus on structural characteristics of calluses has yet to be reached due to the application of term to “callus” to diverse structures displaying proliferative, disorganized growth (Garcia-Reina *et al.*, 1991; Robledo & Garciareina, 1993). True callus culture is considered growth derived from cells excised from an explant (Garcia-Reina *et al.*, 1991), a criterion not met in many studies. Moreover, a strict use of the definition may not be applicable when applied to seaweeds due to differences in tissue organization between higher plants and seaweeds (Aguirre-Lipperheide *et al.*, 1995). We use the term here in a broad sense. Thus, it was reported for *Porphyra* that cells originally prepared as protoplasts grew into callus-like clumps of cells that regenerated into plantlets (Polne-Fuller *et al.*, 1984). It was claimed, optimistically, that such procedures could bypass the need for conchocelis (Polne-Fuller *et al.*, 1984). In a subsequent study in which callus formation in representatives of red, brown, and green algae was tested (Polne-Fuller & Gibor, 1987), the frequency of callus formation was low, and several common generalizations could be made: most calluses developed on moist solid surfaces; the solidifying agent or media did not seem to make a difference; agar does not seem to contain an inducing substance; and auxins and cytokinins, which induce calluses in higher plants, do not induce calluses in seaweeds. Noting numerous unknown variables in successful induction of calluses, a productive short term future for application of calluses in seaweed culture was not foreseen in a study published in 1991 (Garcia-Reina *et al.*, 1991). This view is reinforced in a study questioning the reproducibility of callus production in seaweeds (Aguirre-Lipperheide *et al.*, 1995). Nevertheless, demonstrations of successful callus formation and plantlet regeneration are reported in commercially important seaweeds such as *Undaria* (Kawashima & Tokuda, 1993) and the phycocolloid producing seaweeds *Gracilaria*, *Hypnea*, *Sargassum*, *Turbinaria*, and *Gelidiella* (Collantes *et al.*, 2004; Kumar *et al.*, 2004; Kumar *et al.*, 2007) in studies spanning 1993 to 2007. The “calligenic potential” (i.e., percent of tissue explants that developed calluses) of selected seaweeds is shown in Table 9. In *Pterocladia capillacea*, a red seaweed, calluses were reported to produce polysaccharides with characteristics of agar (Liu *et al.*, 1990). The calluses were maintained for up to three years and produced single cells that grew into more calluses. With more effort directed to this and other species, the utility of callus production for propagation of seaweed may be realized, although the likelihood does not appear short term.

Table 9. Calligenic potential of selected seaweeds (Garcia-Reina *et al.*, 1991).

<i>Chlorophyta</i>	
88.0%	<i>Enteromorpha intestinalis</i>
86.0%	<i>Ulva augusta</i>
<i>Phaeophyta</i>	
70.0%	<i>Ecklonia cava</i>
29.0%	<i>Macrocystis pyrifera</i> (gametophyte)
20.0%	<i>Laminaria sinclairii</i> (gametophyte)
17.0%	<i>Sargassum muticum</i>
17.0%	<i>Pelvetia fastigiata</i>
10.0%	<i>Sargassum hystrix</i>
10.0%	<i>Cystoseira osmundacea</i>
9.2%	<i>Laminaria sinclairii</i> (sporophyte)
9.0%	<i>Sargassum fluitans</i>
7.8%	<i>Macrocystis pyrifera</i> (sporophyte)
<i>Rodophyta</i>	
87.0%	<i>Porphyra lanceolata</i>
84.0%	<i>Porphyra perforata</i>
81.0%	<i>Porphyra nereocystis</i>
75.0%	<i>Smithora naiadum</i>
33.0%	<i>Laurencia sp</i>
18.0%	<i>Phyllophora nervosa</i>
16.0%	<i>Furcellaria fastigiata</i>
15.0%	<i>Gelidium vagum</i>
10.0%	<i>Gracilaria ferox</i>
7.0%	<i>Euचेuma alvarezii</i>
4.0%	<i>Gracilaria verrucosa</i>
2.0%	<i>Ceramium kondoi</i>
1.0%	<i>Gigartina exasperata</i>
0.9%	<i>Euचेuma uncinatum</i>
0.6%	<i>Gelidium robustum</i>
0.5%	<i>Gracilaria papenfusii</i>
0.3%	<i>Gelidium versicolor</i>

The production of protoplasts from seaweed is well established, and the regeneration of calluses or plantlets from protoplasts in numerous species of red, brown, and green seaweeds is now reported (Aguirre-Lipperheide *et al.*, 1995). In comparison with calluses, new thalli are regenerated more easily from protoplasts, and protoplasts are more suitable for suspension culture (Aguirre-Lipperheide *et al.*, 1995). In recent years, emphasis in culture of protoplasts has shifted from production potential in different seaweeds to optimizing production protocols: e.g., improved enzymes for cell wall digestion (Reddy *et al.*, 2006), improved culture media for

protoplasts (Mussio & Rusig, 2006), and testing source material (Benet *et al.*, 1997), although some commercially important seaweeds such as *Gelidium* (Coury *et al.*, 1993) and *Laminaria* (Benet *et al.*, 1997) have been more recalcitrant to either production or regeneration. Seeding nylon filaments with protoplasts for regeneration and growth of plantlets is also feasible for some species. Growth of plantlets regenerated from protoplasts is possible in both the laboratory (Dipakkore *et al.*, 2005; Reddy *et al.*, 2006) and field (Dai *et al.*, 2004; Dai *et al.*, 1993). Recent studies have shown that *Porphyra*, in particular, appears especially promising for growing plants from protoplasts (Dai *et al.*, 2004; Dai *et al.*, 1993; Dipakkore *et al.*, 2005).

Somatic hybridization by protoplast fusion is a well-established technique in the plant sciences. Creating new traits through genetic recombination by protoplast fusion and by transformation with novel genes is particularly appropriate to protoplasts. These methods offer novel paths to creation of new strains of seaweeds. Protoplast fusion using genetically unmodified protoplasts from existing seaweed strains results in genetic recombination and does not alter the gene pool of the extant population, and the propagation of seaweeds from fused protoplasts is considered a “green” technology. It does not introduce genes modified by genetic engineering to the environment. Thus, it is, in principle, a viable approach to cultivation in both open and closed systems. In practice, protoplast fusion is at an early stage of development, as is the case for applications in cellular and molecular biosciences to seaweeds in general.

The first report of protoplast fusion in seaweeds was in 1987 for production of chimeric fronds following fusion of normal and green variants of *Porphyra* (Fujita & Saito, 1990). This and subsequent studies mainly demonstrate protoplast fusion potential, as success rates were generally low (Chen *et al.*, 1995; Fujita & Saito, 1990; Kito *et al.*, 1998; Mizukami *et al.*, 1993; Mizukami *et al.*, 1992; Mizukami *et al.*, 1995). The exception is a study of formation of a hybrid from a *Porphyra* and *Monostroma* fusion (Reddy *et al.*, 1992). Noting the lack of consistent success in prior fusion attempts with *Porphyra*, a recent patent (Cheney *et al.*, 2003) describes a novel approach based on fusion of *Porphyra* protoplasts derived from the conchocelis with those from the conchocelis and blades or thallus from other species. The supporting information shows that such fusions result in greater capacity to regenerate in comparison with fusions based solely on protoplasts from blades. Improved growth of fusion products using this patented approach, in comparison with use of existing strains for protoplast fusions, has been demonstrated in both laboratory and field conditions.

Advances in seaweed cell and molecular biology

Although at an early stage of scientific and technical development, advances in seaweed cell and molecular biology are applied in seaweed biotechnology. For example, restriction fragment length polymorphisms (RFLPs) and random amplified polymorphic DNA (RAPD) analysis, both extensively used in population genetics, are used in understanding seaweed populations (Alberto *et al.*, 1999; Bouza *et al.*, 2006; Dutcher & Kapraun, 1994; Ho *et al.*, 1995; Niwa *et al.*, 2005a) and in strain selection and characterization (Jin *et al.*, 1997; Meneses & Santelices, 1999; Niwa *et al.*, 2005b). Analysis of gene expression using gene specific probes (Jacobsen *et al.*, 2003; Moulin *et al.*, 1999; Roeder *et al.*, 2005), subtractive hybridization (Pearson *et al.*, 2001), differential display (Hong *et al.*, 1995), and expression profiling (Collen *et al.*, 2006a) is reported for representatives of red, brown, and green seaweeds. Transformation systems (Gan *et al.*, 2003; Huang *et al.*, 1996; Jiang *et al.*, 2002; Jiang *et al.*, 2003) and expressed sequence tag (EST) libraries (Collen *et al.*, 2006b; Crepineau *et al.*, 2000; Moulin *et al.*, 1999; Roeder *et al.*, 2005; Stanley *et al.*, 2005; Sun *et al.*, 2006; Teo *et al.*, 2007) are also developed for representatives of these three divisions of seaweeds.

Gene discovery in seaweeds is currently dependent on isolation and characterization of single genes and ESTs, which have now been developed for several species spanning the red, brown, and green seaweeds (Barbier *et al.*, 2005; Belanger *et al.*, 2003; Collen *et al.*, 2006b; Crepineau *et al.*, 2000; Lluisma & Ragan, 1997; Nikaido *et al.*, 2000; Roeder *et al.*, 2005; Stanley *et al.*, 2005; Teo *et al.*, 2007; Wong *et al.*, 2007), including commercially important species such as *Porphyra* (Nikaido *et al.*, 2000), *Laminaria* (Crepineau *et al.*, 2000), and *Gracilaria* (Lluisma & Ragan, 1997; Teo *et al.*, 2007). Pending elucidation of complete genome sequences for seaweeds, databases such as these serve as the basis for expression screening. *Porphyra* (Waaland *et al.*, 2004) and *Ectocarpus* (Peters *et al.*, 2004) are proposed for whole genome sequencing, and sequencing projects are currently underway for *P.purpurea* at the Joint Genome Institute (U.S. Department of Energy) and for *E. siliculosus* at Genoscope - Centre National de Séquençage (France). Such efforts will facilitate activities such as global genomic and proteomic profiling, constructing detailed pathways for secondary metabolite production, and metabolic engineering of seaweed genes to create valuable products.

It is also possible to genetically alter seaweed genomes using modern tools in the life sciences to obtain strains with new characteristics. Chemical mutagenesis of conchospores by treatment with the mutagen MNNG followed by production of protoplasts has resulted in pigmentation and high monospore producing mutant strains of *Porphyra yezoensis* (Yan *et al.*, 2000; Yan *et al.*, 2004). More significant has been the work developing transformation and expression systems for creating transgenic seaweeds. Electroporation of protoplasts with pBS and pQD plasmid vectors carrying the GUS gene for *E. coli* β -glucuronidase under control of the CaMV35S promoter has been successfully used in the transient expression of GUS in *P. yezoensis* (Liu *et al.*, 2003). In *Laminaria japonica* (Jiang *et al.*, 2003), bolistic transformation of dispersed gametophyte cells with the pSV- β -Galactosidase vector under control of the SV40 promoter resulted in expression of β -galactosidase in fronds of the sporophyte. Also under consideration is the use of viruses that affect algae as transformation vectors for transgenesis of seaweeds (Delaroque *et al.*, 2001; Henry & Meints, 1994). These studies represent the proof of principle demonstrations that novel genes can be expressed in seaweed using recombinant DNA technology, thus, paving the way for developing transgenic seaweeds with desired characteristics and marine bioreactors for valued products. *Laminaria* is currently a transformation model for seaweed biotechnology in China, and progress is reported in recent studies for photobioreactor cultivation of transgenic gametophytes of *Laminaria* expressing recombinant tissue-type plasminogen activator protein (Gao *et al.*, 2005a) and hepatitis B surface antigen (Gao *et al.*, 2006; Gao *et al.*, 2005b). These gene products have potential as reagents in the biomedical sciences.

Genetic modification of seaweeds

Commercial production of seaweeds for food is an important facet of the aquaculture industry. Moreover, a number of seaweed products have importance as food additives or in the pharmaceutical/bioscience industry. Seaweed derived polysaccharides, e.g., the agars, agaroses, algin, and carrageenans, are commonly used in food, pharmaceuticals, consumer products, and industrial processes (Renn, 1997). The demand for such products is currently being met through harvesting natural and farmed populations. While the potential for using advances in modern plant breeding techniques and biotechnology is recognized (Renn, 1997), the application of advanced approaches is at an early state of development. Consequently, this field lags behind the

mainstream of plant sciences and agriculture. We focus here on the advancements in seaweed biotechnology, which, in time, will likely advance seaweed culture and the improved biosynthesis of commercially important seaweed products. Furthermore, the release of genetically modified organisms to the natural environment, which will likely place constraints on future application of modern biotechnological approaches to genetic improvement of wild stocks, remains a topic that requires careful consideration. Introduced non-native species are considered a serious threat to marine biodiversity and marine resources, and seaweeds are considered by some to be significant contributors of such threats (Schaffelke *et al.*, 2006). For example, *Caulerpa taxifolia* is a particularly troublesome invasive species, not only for its ecological effects (Jousson *et al.*, 2000), but, in the context of seaweed biotechnology, especially troublesome because of its anecdotal origin as an aquarium strain that was genetically altered by exposure to ultraviolet light prior to inadvertent release to the Mediterranean Sea (Yip & Madl, 2005). It is highly unlikely that genetically engineered seaweeds will be allowed to be cultured in the natural environment in the foreseeable future.

On the other hand, the use of protoplasts and protoplast fusion to recombine existing genes for strain improvement is an approach amenable to producing plantlets for use in open systems where such genes already occur. Although yet to go to commercial application, the production of protoplasts and their regeneration is becoming a routine procedure for a number of seaweed species, including commercially important taxa such as *Porphyra*, *Laminaria*, *Undaria*, and *Gracilaria* (Reddy *et al.*, 2007). Pioneered for seaweeds in the laboratory of D.P. Cheney at Northeastern University, Boston, protoplast fusion adds a novel strategy for strain development, which, in principle, circumvents issues related to introduction of genes altered by genetic engineering or mutagenesis to the gene pool. While technical considerations still need to be addressed (Reddy *et al.*, 2007), it appears that this technology is most likely to have the greatest potential for use in natural environments.

Various stages of seaweed growth are amenable to culture in bioreactors, and interest in such applications has been growing since the mid-1990's. Suspension culture in bioreactors is effective with cell and tissue cultures of various brown, red, and green seaweeds (Rorrer & Cheney, 2004), early life history stages of *Porphyra* (Zhang *et al.*, 2006), and, most recently, transgenic gametophytes of the kelp *Laminaria* (Gao *et al.*, 2006; Gao *et al.*, 2005a; Gao *et al.*, 2005b; Qin *et al.*, 2005). Both protoplasts and somatic hybrids selected for strain improvement

are amenable to growth in bioreactors for plantlet production and later deployment in outdoor culture areas. The Chinese seaweed culturists, working on *Laminaria*, have been most active in applying and optimizing bioreactor technology to genetically engineered seaweeds. The technology of bioreactor based seaweed culture has moved beyond proof of principle to refinement and development for commercial application. Lagging is development of suitable source material of genetically altered seaweed life forms (protoplasts, calluses, plantlets, etc) that synthesize specific desired biochemical products.

More forward-looking is the notion that understanding the molecular mechanisms underlying biosynthetic pathways can result in procedures to construct seaweed gene networks in organisms such as bacteria or yeast, which are more amenable to bioprocessing. Using emerging tools in synthetic biology and metabolic engineering, it is not unrealistic to envision such organisms expressing seaweed genes that encode synthesis of polysaccharides, pharmaceuticals, and other seaweed-derived chemicals. In this scenario, seaweeds essentially become donors of genes in contexts outside of that of seaweed culture *per se*.

For offshore applications, genetic engineering immediately brings issues related to the release of genetically modified organisms to the environment. Given the rapidity at which advances in molecular biology and biotechnology can occur once concerted efforts are brought to bear, the ability to produce genetically modified seaweeds will quickly overtake society's willingness to allow their production in natural settings. Thus, the power of recombinant DNA technology in seaweed culture will likely be restricted to activities that can be conducted under confined conditions. These are also likely to be the first to move to application, because any proposal to use genetically engineered seaweeds in an open system such as those associated with offshore culture will likely be faced with major regulatory challenges. Appropriately contained land-based pond systems, mesocosms, and bioreactors are considered the likely venue for culturing genetically modified seaweeds tailored for production of specific valued commodities.

Potential of marine biotechnology products

Chemicals that have potential uses in biomedical and industrial applications have been isolated from macroalgae over the last few decades. Like most marine organisms, macroalgae produce a host of metabolites and enzymes that differ greatly from those produced by terrestrial organisms. These chemicals allow the marine organisms to deal with the harsh ocean

environment, and to compete for space and resources against other organisms in a type of chemical warfare. While only a small number of these marine products have found their way into the marketplace in a substantial way, many products are under investigation and the potential of future products of considerable value appears strong (Cordell 2000). The greatest efforts are going into finding products with pharmaceutical value, with many lines of inquiry demonstrating that macroalgal metabolites have strong antiviral properties for human pathogens including *Herpes simplex* virus, HIV, and a variety of respiratory viruses, as well as possessing antibiotic properties in humans and animals (Smit 2004). Further biomedical uses include macroalgal metabolites that assist in blood and fluid coagulation and other cellular level processes (Rogers and Hori 1993). Additional lines of biomedical inquiry include the use of macroalgal products as contraceptives, anti-inflammatories, and anti-cancer agents (Smit 2004). Macroalgae, like numerous microalgal species, produce neurotoxins that affect higher organisms including mammals. Many chemical weapon systems are based on marine-derived neurotoxins (Dixit et al 2005); these chemicals are useful in medical and veterinary fields when used in very small doses, analogous to the use of the botulism toxin as the cosmetic application Botox. Industrial uses of macroalgal-derived products include a variety of vermicides, anti-UV sunscreens, and cellular tags for tracking constituents through industrial chemical processes (Smit 2004). Although of great interest and potential, such products would not be directly relevant to the present study, because they would be independently derived, rather than being produced through production paths associated with bioenergy or coproduct production.

CHAPTER 9

TECHNO-ECONOMIC FEASIBILITY OF OFFSHORE SEAWEED PRODUCTION

We present in this chapter an assessment of technology associated with offshore seaweed farming and market perspectives from seaweed production for both biofuels and other valuable coproducts. The economic assessments for the Marine Biomass Program, described above, remain the most comprehensive analyses conducted to date for offshore seaweed farming. Additional progress in offshore seaweed farming has been nominal, and, because sufficient data are lacking for a detailed update, we recommend in our roadmap that such an analysis be conducted in concert with future R&D activities. We focus below on the economic and public policy perspectives of offshore seaweed farms, the notion of seaweed biofuels, seaweed production potential compared to other biomass resources, and potential co- and by-products from seaweed digestion.

Offshore seaweed farms

The offshore seaweed cultivation concept

Advances in marine engineering of offshore platforms have provided new designs and improvements on existing designs. The failures of early offshore seaweed farms, specifically of the Marine Biomass Program, were due to equipment failures, which occurred because the rigging, platforms, and anchoring systems were not sufficiently robust to withstand the rigorous conditions presented by unsheltered ocean waters. Even small storms wiped out entire operations and pilot projects (It should be pointed out, however, that the loss of the last offshore platform was clearly due to a hit-and-run ship collision, not an act of nature, but humans). Newer designs, including firmly anchored, “floating” but lightly tethered, and wholly floating systems, have all benefited from advances in marine engineering, mainly from lessons learned with oil and gas rigs. More recently, the construction of offshore wind farms, and now wave and tidal power generators, have provided additional advances in marine engineering

New lightweight but very strong materials, such as metal carbides and strengthened steel extrusions, are being used to build such offshore structures strong enough to withstand ocean conditions. Lighter weight materials decrease installation and transportation costs and allow for less massive infrastructure, allowing, in principle, for greater surface area for seaweed growth

and spread. Of course, for the specific applications envisioned here, most such materials are much too costly to be applicable, at least in the foreseeable future. Still their availability suggests that they can be used in at least the experimental and pilot phases of such projects.

New concepts and designs of open ocean seaweed farms draw on the availability of new materials and ocean expertise. One such design is a very large (400 km², 100,000 acres) unmoored structure towed slowly by ships and barges, which was discussed at the 1990 Marine Biomass Workshop (Chynoweth, 2002). The concept would be to maintain such structures in areas of natural upwelling, where nutrients are supplied, water movement is sufficient to break down diffusion barriers, and sunlight and temperature are optimal.

Economic aspects

Determining the market value for large volumes of seaweed biomass is difficult as there is little published information about the costs and values of farmed seaweed from land-based and nearshore operations, much less offshore farms. What information is available is for dried seaweed, including dried kelp raised for food in China [about \$0.60 to \$0.80 per kg – (Chen personal communication)]; and dried kelp for mannitol, algin and iodine production (\$0.30 to \$0.40 per kg). These prices allow a crude calculation of costs on the order of \$0.50 per kg dry weight (\$0.075 per kg wet weight). By comparison the price of corn for ethanol production is presently about \$0.16 per kg dry weight.

In the 1980s, Marine Biomass Program economists calculated that, in order to economically grow seaweed for energy biomass in offshore seaweed farms those farms would need to be up to 2,600 km² in area, and produce additional revenue in the form of animal feedstocks at \$23 to \$72 per dry ash free metric ton in 1987 dollars (Bird, 1987). Assuming that seaweeds average 85% water and that ash typically makes up about 5% of the wet weight (Show 1979), this corresponds to a cost range of \$2.30 to \$7.20 per metric ton wet weight, or 0.23 to 0.72 cents per kg wet weight in 1987 dollars. An update of these figures, as well as the changing economic context of biofuels in today's market is presented in Chapter 3.

Most operational offshore farms raise fish and shellfish, are much smaller than those envisioned for seaweed biomass growth, and yield higher values per unit area. Offshore fish and shellfish farms are usually no more than five hectares in area, may involve fixed capital

investments of \$3 million or more (Kama *et al.*, 2003), and may generate annual revenues in excess of \$3 million (Posadas, 2004) with unit process of \$3 per kg or more.

Public policy perspective

Public perceptions of marine aquaculture operations differ around the world. In Asia, nearshore operations are viewed as a sign of prosperity. In North America they are often viewed with suspicion and hostility. Overfishing, shoreline development, contamination and global climate change have affected capture fisheries all over the globe, and attitudes towards farming of marine products have been changing. In North America and Europe, the public is demanding safeguards for native species and habitats, but the willingness to consider aquaculture farms is increasing. For example, by proposing to group different aquaculture species together in culture operations and to co-locating aquaculture facilities with wind farms and other infrastructure, decision-makers in Europe are beginning to support such operations (Reith, 2005).

Public acceptance of marine culture operations will probably always be higher in Asia, South America, and the Pacific Islands than in North America and Europe. In 2004 the National Oceanic and Atmospheric Administration introduced the Offshore Aquaculture Act that promotes research and technology development to assist in siting and operating aquaculture operations offshore.

Biofuels from seaweeds

With the energy crisis starting in the early 1970's, a need to develop new energy sources resulted in efforts to develop biofuels such as ethanol from lignocellulosic biomass, oils from microalgae cultivated in land-based ponds, and methane from seaweeds growing in the open oceans. This interest was due to the perception that these approaches could avoid land and water resource limitations of crop plants and led to the Marine Biomass Program, which is described in more detail in a previous chapter. Currently there is again a great deal of interest in 'next generation' biofuels, which include novel and alternative feedstock resources for producing lignocellulosic ethanol, "green" biodiesel and other biofuels, beyond those based on corn, sugar or vegetable oil crops.

The original ideas for large offshore seaweed farms that led to the Marine Biomass Program was an integrated multi-product concept leading to production of food, feed, fertilizer, and

energy (Benson & Bird, 1987). Of these general product categories, energy was originally considered the least important economically in the open ocean seaweed farm concept. Moreover, the production of the first three items supported established industries, whereas the production of energy had yet to develop into an industrial activity, as is still the case today. Nevertheless, the potential of producing energy from seaweeds was considered to be extremely high and inspired considerable past expenditure of energy and funds for an initial proof of principle test of large scale offshore seaweed culture facilities, i.e., the Marine Biomass Program. Currently, optimism regarding the untapped potential in seaweeds as a biomass source for energy production is renewed in some circles and is leading countries like Japan (Aizawa *et al.*, 2007), Denmark (Denmark, 2007), and the United Kingdom (Kelly, 2006) to begin contemporary trials in growing seaweeds for anaerobic digestion to produce methane, or for fermentation to produce alcohols. Additionally, recent advances in offshore technology are being applied to the offshore culture of seaweeds. Successful efforts at growing *Laminaria* in the North Sea, for example, have been encouraging (Buck & Buchholz, 2004).

An extensive technological and economic analysis (Chynoweth *et al.*, 2001) showed that the energy potential of seaweeds compares very favorably to a variety of terrestrial biomass and waste sources, exceeding the latter by over three-fold using exajoules per year as a basis of comparison. This study concluded that it would be economically viable to convert seaweeds to methane through anaerobic digestion, provided an adequate supply of seaweed biomass was available. High yields and conversion rates can be expected with use of appropriate salt tolerant microbial consortia as inocula for the anaerobic digestion (Chynoweth *et al.*, 2001).

In the context of developing new processes for the production of alcohol-based biofuels from seaweed, it is worth noting that seaweed could be fermented, as was done during World War I (see Chapter 4), to n-butanol. Butanol is currently considered a second generation biofuel because it is particularly well suited as a replacement for gasoline and jet fuel. It is superior to ethanol because of its higher energy content, lower corrosiveness, better miscibility, greater octane-improving power, and other positive qualities (Zerlov *et al.*, 2006). Indeed, DuPont and British Petroleum joined in 2006 to manufacture butanol biofuel via acetone-butanol (AB) fermentation of sugar beets, with the goal of producing 9 million gallons of bio-butanol per year (Chase, 2006).

The acetone-butanol (AB) fermentation is carried out by anaerobic heterotrophic bacteria of the genus *Clostridium*, most commonly the species *Clostridium acetobutylicum*. Pilot-scale and industrial AB fermentations have been successfully carried out in a number of countries, such as the United States, the Soviet Union, South Africa, Austria, and France (Nimcevic & Gapes, 2000). Butanol production ceased in the United States in the 1960s when the main agricultural feedstock (i.e., molasses) needed for the fermentation process was diverted to animal feed and cheaper butanol from inexpensive petroleum sources became available (Zerlov et al., 2006). However, given the high crude oil prices today, fermentative butanol (Zerlov et al., 2006) production could very well become economically competitive if an abundant and cheap feedstock could be identified (Gapes, 2000; Zerlov et al., 2006). Following the earlier successes of AB fermentations in the Soviet Union, it is suggested that lignocellulosic and agricultural wastes could be converted in a butanol biorefinery to a number of useful products, including bio-butanol (Zerlov et al., 2006).

Seaweed biomass, because of its large resource potential (see Chapter 10), could serve as an inexpensive and abundant feedstock for such a butanol biorefinery. However, given that there are no published reports on AB-fermentations using seaweed other than the historical description of World War I acetone-butanol production facility in Southern California (Chapter 4), further research is required to test the feasibility of this concept. Specifically, it is necessary to determine whether seaweed needs to be treated by acid hydrolysis or enzyme digestion prior to AB fermentation by *Clostridia*. It may also be possible to identify specific species that can ferment seaweed biomass directly without pretreatment. In addition, butanol production rates and yields need to be determined by testing different seaweed feedstocks and fermentation conditions.

In the offshore farm concept noted above, the aim was to split the source material so that a portion would be used for energy conversion and the remainder for production of other valued products. However, more efficient production scenarios can be envisioned in which the same seaweed feedstock is used to produce both energy and valuable products. Such alternative schemes are explored below.

Seaweed production potential compared to other biomass resources.

Perhaps the best example, for comparative purposes, is the production of microalgae biofuels. Like the Marine Biomass Program, the Aquatic Species Program for terrestrial microalgae production was a major U.S. Department of Energy R&D effort (See Sheehan et al., 1998, for a review). At present, there are still as many unresolved technological issues with microalgae biomass production on land as with open ocean seaweed farming offshore. In both cases it is possible to point to prototype commercial systems that provide guidance on how this technology could advance in this future towards the goal of cost-competitive biofuels production. There are microalgae pond systems used for treatment of wastewater and for production of food supplements (in separate ponds). Seaweeds are grown in near-shore farms for foods, colloids and other relatively high value products.

An objective comparison of these two technologies would have to conclude that seaweed production is the more advanced: global production from near-shore seaweed farms is in excess of a million ton (ash-free dry weight) versus barely 10,000 ton for microalgae biomass. And costs are much lower: microalgae sell for, on average, close to \$10,000 per ton versus under \$1,000 per ton for cultivated seaweed. Although neither technology is close to achieving the required production cost for biofuels, near-shore seaweed culture is certainly closer to the goal than microalgae cultivation.

The strong argument for open ocean seaweed culture is based on the potential resource base, something that microalgae enthusiasts also claim, but with rather weaker arguments. The stronger argument for microalgae biofuels is that the technology, and the requirements to meet the goals for biofuels production, are better understood and perhaps more achievable, compared to the large uncertainties and high risks in offshore seaweed culture.

A billion ton of biomass is optimistically projected as the potential future lignocellulosic biomass resource for the entire landmass of the United States, including all forest and agricultural resources that could possibly be marshaled to the cause of biofuels (Perlack, 2006). However, it is doubtful that more than about half of the projected amount of biomass would ever become available for conversion to biofuels due to limitations to extensive biomass production. Furthermore biofuels compete with food, feed, and fiber, not to mention lumber, land, water, fertilizers, etc. No biofuels approach can promise to provide a major alternative to fossil fuels, which would require several billion ton of biomass.

Further, the economics of biofuels is far from certain or assured. For example, although the U.S. Government has mandated the production of over 20 billion gallons lignocellulosic ethanol within 15 years under the 2007 energy bill, the process has yet to advance to the pilot plant in the U.S. Nevertheless, as the availability of fossil fuels, a finite resource, declines in the future, biofuels should be regarded as one of a suite of energy sources available to society. The integrated multi-product ocean farm concept, which combines production of biofuels and other valued products from cultivation of seaweeds, merits further consideration.

Potential co- and byproducts from seaweed digestion

Opportunities to produce and sell co- or byproducts from farmed seaweed depend on the way it is processed. If methane is a priority and if the only way this can be produced is by anaerobic digestion, then most other products of any value from seaweed will have to be extracted before digestion because, with the exception of minerals, most other materials will be degraded during the digestion process. It is possible that residues left after digestion may have value as an animal feed ingredient but this has not yet been demonstrated.

Development of large scale offshore seaweed farming can therefore follow one of two paths: (1) focus on bioenergy like the Marine Biomass Program, which, in turn, would mean a major new research effort to develop very large scale methods for offshore seaweed farming; or (2) try to develop offshore seaweed farming on the scale needed to establish profitable businesses and, in doing so, explore numerous options for products, processes and location, which may lead to improved technologies for farming seaweeds for energy.

To determine if the second path makes sense it is necessary to understand more about the nature of the market for seaweed products in general. The information that exists in the public domain is limited and inconsistent. Surialink.com provides data from a 1991 study by Indergaard and Jensen (Table 10) and 1996 data from Perez (Table 11), excluding data of *Maerl* because they would distort understanding of the market that is of interest in the present study.

McHugh (2003) provides FAO data (Table 12), which is most likely up to date to 2001 based on the way FAO collects and reports statistics.

Recently, FAO reported that global production of all farmed seaweeds in 2004 was 13.9 million metric ton valued at \$6.8 billion and data kindly provided by Subasinghe (personal communication) shows how it has grown to this level since 1990 (Figure 29).

**Table 10. World seaweed market in 1991 after Indergaard and Jensen
(www.surialink.com).**

Product	Value (\$ million USD)	Metric ton dry weight /yr	Metric ton wet weight /yr)
Alginates	230	27,000	500,000
Agar	160	11,000	180,000
Carrageenans	100	15,5000	250,000
Kelp meal	5	10,000	50,000
Liquid extracts	5	1,000	10,000
Nori	1,800	40,000	400,000
Wakame	600	20,000	300,000
Kombu	600	300,000	1,3000
Total	3,500	424,500	2,990,000

**Table 11. World seaweed market segments after Perez 1996
(www.surialink.com).**

Market Segment	Metric ton wet wt / yr	Share %
Food staple	3,600,000	51
Food gums	3,200,000	46
Others	200,000	3
Total	7,000,000	100

Table 12. Worldwide seaweed production (McHugh, 2003).

Total world seaweed production (farmed and wild)	7.5 to 8.0 million metric ton
Value	\$5.5 to \$6.0 billion
Food products for human consumption about \$5 billion	83%
Hydrocolloids \$600,000 from wild harvested seaweed	10%
<u>Remainder, including farmed seaweed used for hydrocolloids</u>	<u>7%</u>

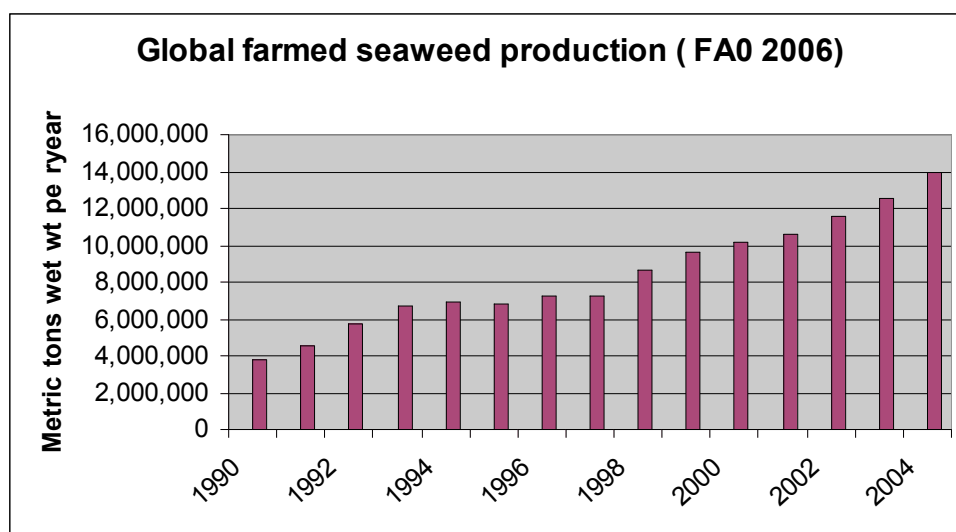


Figure 29. Global farmed seaweed production from 1990 to 2004.

Key numbers on worldwide seaweed taken from each of these sources are summarized in Table 13 to produce a global perspective on seaweed production. There appears to have been an enormous increase in production from 2001 to 2004, without a comparable increase in value.

Table 13. Summary of worldwide seaweed production

Date	Source	Production metric ton/year	Value \$ million
1991	Surialink.com	2,990,000	3,500
1991	FAO	4,550,000	Not available
2001	McHugh (FAO)	8,000,000	6,000
2004	FAO	13,900,000	6,800

The data also suggest that much of the growth in production has been for consumption as human food versus other uses such as hydrocolloid production. In 1996, the ratio was 51% to 46% respectively, compared to 83% to 10% in 2001. Part of the explanation for this is that the 1996 figures include wild harvested seaweed most of which is used for hydrocolloid production. Even so, there seems to have been a shift towards the use of seaweed for food, driven almost entirely by markets in China, Korea and Japan. Whether this trend continues is uncertain. Chen (personal communication), for example, indicates that about half Chinese *Laminaria* production is now used as food and half is use for industrial purposes including production of iodine, mannitol and algin. This is supported by Pawiro (2006) who reported that the alginate industry is facing strong competition from Chinese producers who sell cheaper alginate made from *Laminaria*. Further, there are reports that demand for seaweed for food in Asia has now reached a plateau and is stagnant (Pawiro, 2006).

New technologies that may add value to co-products and byproducts of algal growth are under development. For example, fermented seaweed could be used to replace microalgal cultures in marine fish and shellfish hatchery feeds (Uchida, 2003). And, most recently the same group of researchers, working in collaboration with Nippon Suisan Kaisha, announced that a seaweed species of the genus *Ecklonia*, when fermented with *Lactobacillus*, was effective in controlling red sea bream iridovirus. They have now begun work to test mass fermentation of seaweeds (Anonymous, 2006).

The nutrients in seaweed are bound in such a way that makes them relatively indigestible to monogastric animals; research into alternative extraction methods are needed to address the extraction of nutrients from whole seaweed tissue. While extraction and sale of various complex carbohydrates such as alginates is already well established, there may be opportunities to develop proprietary process technologies for other seaweed derivatives. In particular, research on extraction of seaweed derivatives can be addressed towards the animals feed market, as well as the human food market. The human market already accounts for the largest proportion of seaweed produced in the world, although the demand is almost exclusively in Asia. It is questionable if consumption of raw seaweed allows for extraction of the maximum nutritional value. In fact, its primary demonstrated nutritional benefit is the provision of iodine (Teas, 2006). Many potentially nutritious components of seaweed are bound to phenols and polysaccharides so that humans or animals cannot readily digest them. If such compounds could be ‘unlocked’

through processing and then incorporated into products that are appealing to western tastes, the market could be created through promoting potential health and ecological benefits.

If large scale methane production by anaerobic digestion of seaweeds is feasible (currently thought to be unlikely in the near term due to the lack of sufficient seaweed feedstock), further research will be needed to characterize the constituents and nutritional value of digester residues. It is also important to examine alternative extraction processes that might be used instead of anaerobic digestion or prior to digestion, in order to separate coproducts before they are degraded.

A comprehensive private study on seaweed markets by CPL Consultants dated some time after 2001 identified possible partners or acquisition targets by reviewing seaweed business in 52 countries (<http://www.cplsis.com/index.php?reportid=195>).

Other market sectors for seaweed products and services

We can identify eight market segments that merit further discussion: human food; polysaccharide gels (hydrocolloids); other polysaccharides & biologically active materials; minerals; soil conditioners and supplements; animal feeds; cosmetics; and seaweed for bioremediation. Some key points about each of them are noted below.

Human Food

- This market consumes at least half of all world seaweed production.
- The four main product forms are nori (*Porphyra*), kombu (*Laminaria*), hijiki (*Hizikia*) and wakame (*Undaria*).
- These are mostly eaten and traded in China, Republic of Korea and Japan but demand in these countries now is reported to be stagnant.
- Seaweed protein is often relatively indigestible; the main nutritional value from seaweeds is from micro constituents including minerals and biologically active polysaccharides that may inhibit viruses and cancers.
- There is an active, though relatively small market in seaweed supplements, nutraceuticals and health foods. This is based on the presumption of health benefits as much as hard evidence.

- Some seaweed is eaten in Western countries, especially in ethnic communities and with sushi, and it seems probable that volumes are increasing based on healthy eating trends in some sectors. Attempts have been made to develop seaweed preparations and dishes that appeal more strongly to Western tastes, notably in France, but with no obvious success so far.
- Increasing public preoccupation with health suggests that it might only be a matter of timing and development of the right products for considerable growth in these non-traditional markets to occur.

Polysaccharide gels

- These are Alginate, Agar and Carrageenan
- Agar production is almost 15,000 metric ton valued at \$114.2 million and about 90% is used in the food industry where demand is not expected to expand, whereas the use of agarose in biotechnology will expand (Pawiro, 2006).
- Carrageenan production is about 49,000 metric ton valued at \$340 million. About 90% is used in the food industry where prospects for growth are better than for agar (Pawiro, 2006).
- Alginate production is between 32,000 to 39,000 metric ton, about 67% of which is technical grade used in industrial applications and 33% used in food and pharmaceutical industries (Pawiro, 2006).
- Production and marketing of all three hydrocolloids is well established and is dominated by international companies such as CP Kelco, Danisco, Degussa, FMC Biopolymer and ISP.
- Raw material for Agar and Carrageenan comes mostly from farmed, red tropical seaweeds.
- Raw material for alginates comes mostly from brown, wild-harvested, cold water seaweeds.
- Gels from terrestrial plant sources and synthetic or microbially produced gums are competitive in some cases.

For several reasons the market for these polysaccharide products does not appear particularly attractive for a new offshore, seaweed farming business, because established players

already dominate the market and the demand for alginates and agar is forecast to increase only modestly. Additionally seaweeds from which agar and carrageenan are extracted are slow growing red seaweeds and it seems unlikely that these species would be suitable candidates for large scale offshore seaweed farming, and there is competition from non-seaweed sources.

Other polysaccharides and biologically active materials

- These include mannitol, fucoidan and other sulfated polysaccharides such as fucans.
- Mannitol is a white crystalline solid that is odorless and has a sweet taste. Its primary uses are as a low calorie sweetener and various medical applications.
- Mannitol can be extracted or made from various sources such as fructose derived from corn starch.
- A recent US patent application proposes a biobased method of making it that involves feeding fructose to *Lactobacillus sintermedius* and indicates the possibility of substantial cost reduction.
- Most mannitol that is made from seaweed is produced in China, probably reflecting the low cost of raw material there.
- Fucoidan is one of several ‘sulfated polysaccharides’ found in seaweeds and seems to occur at quite high levels (4-10% dry weight) in some (Horn, 2000). This group of organic chemicals is found only in seaweeds.
- Fucoidan especially has been studied for its antiviral properties and is sold as a nutraceutical and in creams and ointments for protection against HIV.
- An Australian company, Marinova, bills itself as the world’s leading fucoidan company and uses seaweeds harvested from the wild in Tasmania.
- There is a worldwide research effort and various claims, some of doubtful value, on other biologically active materials that might be extracted from seaweeds.

While interesting and helpful in that the discovery and use of these substances enhances the image of seaweeds as being useful to humans, it is doubtful that their production and sale would drive the development of large scale offshore seaweed farming. In most cases the quantities of material are quite small so, if they have a role to play it would be supplementary to other products that would drive the business.

Consideration of potential byproducts in the Marine Biomass Program put quite a high value on what was termed “L-Fraction”. As best as it can be understood, this is refractory phenolic material bound with carbohydrates and/or protein and it was thought that they might have value in industries such as plastics. A web search for the term and present day possible applications reveals no obvious uses or any volume sales. Therefore it seems that its value as a possible byproduct may have been overstated in earlier work.

Minerals

The primary minerals of interest in seaweed are iodine and potash, both of which were itemized as potential byproducts of digestion in the Marine Biomass Program. Insofar as these materials would survive the digestion process, they could be legitimate byproducts from an energy focused offshore seaweed farming program. However, there is already a plentiful supply of both minerals from terrestrial sedimentary deposits. Potash is the most interesting of the two because of the large world demand for potassium in fertilizers estimated at 25.8 million metric ton. But natural deposits are vast with 8.4 billion metric ton of deposits being considered commercially exploitable. The leading producers are in Canada. Similarly, the main source of industrial iodine is from terrestrial deposits with the leading producer in Chile, which produces 50% of the world’s supply. Neither of these minerals, therefore, appears attractive as a product that would drive or even materially help to drive the development of an offshore seaweed farming industry.

Soil conditioners and supplements

There is worldwide use of seaweed in agriculture and it is used in two ways. First, compounds known as cytokinins can be extracted from some seaweeds and these have been found to promote growth in some plants, probably by mimicking hormone function. Second, whole seaweed or seaweed compost is used as a soil conditioner where the polysaccharides in it serve to increase moisture retention.

Cytokinin extracts are made by several companies including a company known as Kelpak in S. Africa that makes it from wild harvested seaweed, and several Chinese companies who advertise on the Internet and presumably use farmed sources of seaweed. Though this market seems quite small presently, being within the 7% of ‘other’ products mentioned by McHugh

(2003), it seems of interest because of the potential to grow substantially if seaweed growth promoters were to become more widely accepted and used more broadly in agriculture. To accomplish that will require more research to document the benefits followed by a major promotional and marketing effort.

Seaweed composts are made from storm tossed seaweeds collected from beaches in Europe and from some residues from various polysaccharide extraction processes. Whether residues from anaerobic digestion could be composted is not clear, as much of the beneficial carbohydrate in seaweeds would have been degraded by digestion. Composting might remove the resulting high levels of ammonia.

Animal feed

The potential for animal feed production from digester residues and/or raw seaweed prior to digestion merits serious consideration. The following from a report on a presentation in 2005 by Roger Gilbert, General Secretary of the Feed Industry Federation illustrates why this matter is more important today than it was at the time of the Marine Biomass Program.

“The demand for animal products will outstrip production if we do not take into account population and economic trends in our calculations. Demand for protein and energy sources for animal feed will dominate our industry. In the next 45 years, the world will need to produce three times more meat, milk and eggs than it does now. The world currently produces some 600 million tonnes of compound animal feed, with the United States at the top of the list with 145 million, the EU producing 140 million tonnes followed by China and Brazil, with 90 and 44 million tonnes respectively. In my view China will help Asia overtake North America within the next five to 10 years.” (Reuters News <http://www.planetark.com/dailynewsstory.cfm/newsid/31257/story.htm>)

It should be noted that these numbers are for dry ton. Assuming equivalent moisture content to most seaweeds of about 85%, 600 million ton dry weight of animal feed equals 4 billion ton of raw seaweed.

The nutritional value in seaweeds suggests an alternative use of digester residues or even an alternative bioprocessing pathway whereby products such as proteins are extracted prior to anaerobic digestion (Horn, 2000). This would avoid destruction of valuable products prior to

conversion to energy. The brown algae *Ascophyllum nodosum*, for example, has been used in animal feeds in Norway for many years. Indergaard and Minsaas (1991) showed in feeding trials that the nutritional value of meal from this species was less than 30% of the feed value of grains. Primarily, this seemed to be due to poor digestibility of the protein, which is likely due to its being bound to phenols to form insoluble compounds. These authors noted that if a simple method for removing phenols from brown algae could be found, it would make brown algae meal much more attractive as a feed supplement.

Cosmetics

Though highly marketed and often highly priced, these products almost certainly represent a relatively small share of the 7% of remaining uses for seaweed described by McHugh (2003). The life cycle of products in the cosmetics industry is short. No medically established ingredients with clearly unique cosmetic properties have been identified. It seems unlikely that this market would represent a significant opportunity for an offshore seaweed farming business.

Bioremediation

There is an apparent beneficial effect of large-scale seaweed farming on the quality of Chinese coastal waters. There are also a number projects and reports of seaweeds being grown to polish effluents or to remove nutrients from seawater including nutrients from fish farms. The concept of integrated marine aquaculture includes the use of seaweeds to recycle animal waste products. Where such seaweeds can be sold for at least the cost of operating a separate activity to grow them, this would seem to make good sense. The Seapura project in Europe contemplates the steps shown in Figure 30 as part of its program. However, in the context of the present study this prompts further questions: Are there any opportunities for large scale remediation of coastal waters that in turn would require the development of large scale seaweed farms, prior to testing offshore farms? Would use of seaweeds in remediation add monetary value to seaweed production?

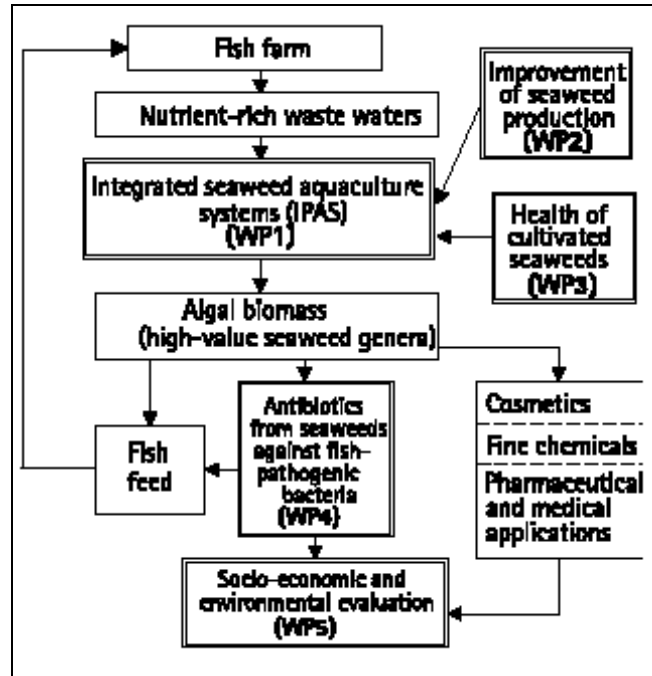


Figure 30. Function flow sheet of the SEAPURA project in Europe (Wadden Sea News, 2001).

CHAPTER 10

SCIENCE AND TECHNOLOGY ROADMAP

The oceans account for over 70% of the earth's surface, yet contribute less than 2% of our food, feeds, and biomaterials. Over five billion metric ton of biomass are removed annually for human use from the land, over 90% of which is plant biomass, harvested from over 24% of global land area (Millennium-Ecosystem-Assessment, 2005; Waggoner, 1995). By comparison, the world's oceans currently yield only 13.9 million metric ton of wet seaweeds annually, or somewhat over 1 million dry ton of biomass, the large majority of which comes from seaweed crops cultivated in Asia (Table 1).

This enormous dichotomy illustrates the current limitations and potential opportunity for the cultivation and harvest of ocean resources, particularly marine plants. For example, if methods were perfected to farm seaweeds on 1% of the world's 361 million km² of ocean surface area (Sverdrup *et al.*, 1942), at production levels already achieved in large-scale coastal seaweed farms in China [average yield approaching 10 metric ton of dry biomass per hectare per year, (Chen, 2006), 3.5 billion metric ton dry weight of new biomass derived from seaweeds would be produced annually. This could be processed into biofuels, animal feeds, industrial polysaccharides, fertilizers and other co-products and would exceed by several-fold what is projected to be available for biofuels from terrestrial biomass resources. The future cost of production of energy from seaweeds is projected to be equivalent to that of energy crops like sorghum and poplar (Chynoweth *et al.*, 2001). Thus, overall, the potential for seaweeds compares very favorably with terrestrial sources. Although current technology cannot deliver more than a small fraction of this potential, the rapidly changing economics of biofuels and other land-based biomass production, as well as advances in marine engineering and biotechnology generally, suggests that there is significant potential for large increases in seaweed production. However, this will require open-ocean seaweed farming, as it is the only way to increase the worldwide marine biomass harvest without damaging already crowded and fragile coastal environments.

Open ocean seaweed farming could also reduce pressures on converting pristine terrestrial ecosystems into agriculture, which is now reported to be a major source of greenhouse gases as an unintended consequence of recent expansion of first generation biofuels production

(Searchinger *et al.*, 2008). Siting offshore farms in natural upwelling areas, as proposed in this report, would avoid the need to provide fertilizer to grow the biomass, reducing costs and the potential impacts of artificial upwelling, which could actually increase greenhouse gas emissions due to additional CO₂ releases. Seaweeds grown on offshore farms in natural upwelling areas would be a carbon neutral renewable energy resource, and have other benefits, such as not requiring a new source of fresh water for cultivation and the recovery of nutrients for production of fertilizers.

Cultivation of seaweed offshore poses enormous technical challenges, and even if these were overcome, the economic viability of such a technology is far from secure. The U.S. Department of Energy Marine Biomass Program remains the singular effort that attempted offshore seaweed farming on a large scale. Notable were its failures related to the lack of structural viability of the offshore structures and attached seaweeds in a dynamic marine environment, but its successes were also noteworthy, as they relate to the *a priori* economic analyses, the increased understanding of the importance of structural integrity of the plant support systems, and the greatly increased understanding of seaweed biology. Despite the initial failures, and the large remaining challenges, the ultimate potential, in terms of biomass production, is so high that continued investigation is warranted.

In this report we propose locating farms only in natural upwelling areas to overcome the nutrient (fertilizer) supply issues related to the initial ocean farms concept, which required artificial upwelling of nutrients. We believe that this approach, together with a more conservative assumption of productivity and advances in marine materials sciences and engineering, can overcome previous limitations. We must point out that use of natural upwelling areas was considered in the Marine Biomass Program, but dismissed at the time as impractical, because the upwelling zones were neither stationary nor predictable enough in U.S. waters (Ashare *et al.*, 1978). This was in part due to the fixed position of the moored structures then investigated, and the emphasis on the U.S. Exclusive Economic Zone.

Deployment of offshore seaweed farms is a long-term objective, and we envision a phased approach, with the incremental steps starting with demonstration of the fundamental principles of the process using experimental land-based, growth chambers and mesocosms to optimize species selection and growth conditions. This would be followed by nearshore testing

of growth apparatus (Figure 31). In addition a continuing engineering design and economic

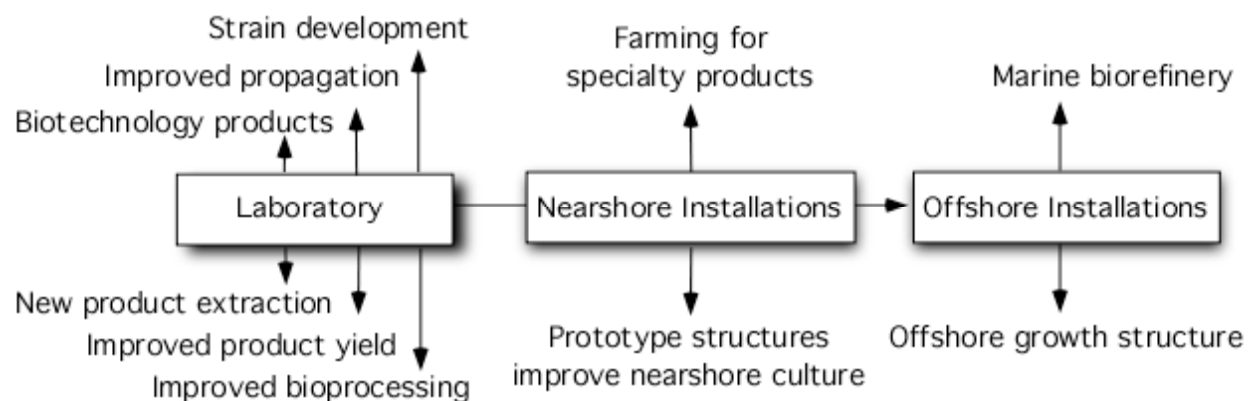


Figure 31. Examples of secondary targets of interest to industry on the path to realization of the Offshore Seaweed Farm.

feasibility analysis would be required to develop the concept of the offshore systems. Success will be measured in terms of developing the long-term vision, while achieving incremental goals. Success in laboratory- and near-shore based activities, for example, may lead to improved processing techniques for second-generation biofuels such as butanol and production of higher value seaweed products of commercial interest. This would encourage near term participation by industry, and provide a pathway for the longer-term advance to truly open ocean systems. Advancing the technology for producing high value products from seaweeds is integral to the path leading to the realization of large-scale offshore farms.

Conceptual system for the Offshore Seaweed Farm

The structures used in the Marine Biomass Program were designed around the concept of tethered systems with attachment sites provided for seaweeds and upwelling pipes to bring nutrient rich waters from depth. Both the tethering of the structures (which limits deployment to relatively shallow areas) and the mechanical nutrient upwelling systems were major design limitations of this initial concept. Indeed, their costs required the farms to achieve biomass productivities that, at least in hindsight, were unrealistic. Recently, in Japan, the concept of floating seaweed pens, with mechanisms for controlling drift, has been introduced. Although

lacking in detail, these concepts are for very large areas, tens of square kilometers, enclosed by simple nets, containing floating seaweeds. However, it is not readily conceivable how floating plants would achieve even distribution in such systems. Furthermore, the supply of nutrients has not been clarified. We believe that one reasonable approach would be to combine the concept of a free-floating farm with dynamic positioning (both horizontal and also vertical, to avoid storm events), with a support system for the plants, similar to the designs of the Marine Biomass Program. We suggest floating platforms of roughly 1 km² (100 hectares) as life support systems for attached seaweeds and term this concept the Offshore Seaweed Farm. We will focus on such a system in a preliminary financial cost-benefit assessment below. We will continue to evaluate other systems, such as the tethered German offshore-ring, which has been successfully tested in nearshore open water environments, before a recommended system is identified.

Taking advantage of upwelled nutrients avoids the issues of nitrogen and phosphate fertilizer supplies, distribution, and utilization efficiency. What becomes important, instead, is availability of the nutrients to all parts of the seaweed life support system. The upwelled nutrients would also be eventually recovered as nitrogen, phosphate, and potassium fertilizer from the seaweed biomass processing facility. It should be noted that this concept depends on the ability to place offshore farms in desirable locations around the world, where nutrient upwelling combines with favorable climates, and other requirements.

An actual resource assessment for such locations, their extent and potential availability, remains to be carried out. However, a very preliminary assessment suggests that there is a potential of a billion ton of biomass that could be produced by such systems. This is based on an estimate of using about 0.3% of the ocean surface, or 1 million such 100 hectare floating ocean farms, producing 10 metric ton of dry biomass per hectare annually (about ten-times less than the projections for the Marine Biomass Program). Of course, this is an order of magnitude projection, and only a more detailed techno-economic analysis will allow a better estimation of the likely resource limits to the proposed Offshore Seaweed Farm technology.

Such an analysis will need to address plausible advances in technologies that can be applied in developing such offshore farms, including marine engineering, material sciences, robotics, and improved seaweed culture and processing. The environmental benefits and drawbacks, as well as social and political implications of the development of such a technology will need to be addressed. Most important, it will be necessary to provide a vision of the

potential costs and benefits of such a technology, within a future that is pointing to a need for renewable and sustainable energy and resources. These benefits and costs are addressed below.

The Marine Biorefinery as part of the Offshore Seaweed Farm

In addition to the Offshore Seaweed Farm, we also propose the Marine Biorefinery, located onshore; to process the seaweed biomass produced offshore, converting it into biofuels and other high value products. The concept of the Marine Biorefinery is adapted from the "sugar" and the "thermochemical" biorefinery "platforms" championed by the U.S. DOE Biomass Program. Moreover, marine biorefineries are part of a biorefinery concept put forth by the Dutch Biorefinery Network for processing micro- and macroalgae.

We propose to add a "seaweed platform", which would include producing both fuels and valuable coproducts. This is a key component of the Technology Roadmap outlined below. It envisions developing improved bioprocessing of seaweed biomass as a function of the Marine Biorefinery, even before the concept of the open ocean Offshore Seaweed Farm is fully developed. This would encourage private companies to take an early commercial interest in this R&D effort and become a driver in the long-term path towards the development of the Offshore Seaweed Farm. The Marine Biorefinery concept is applicable to near-shore seaweed resources and could later be expanded to the scale envisioned for the open ocean Offshore Seaweed Farm. Thus, development of the Marine Biorefinery, through private sector participation, would reduce the technology risk by combining product development with early-stage onshore (laboratory and mesocosm-based) and nearshore project activities.

Our target for the long-term Offshore Seaweed Farm program is about 1 billion dry ton of seaweeds grown on a very small fraction of the world's oceans. This compares favorably with a maximum biomass potential from all forest and agricultural lands of the U.S. of 1.3 billion dry ton annually, with the potential to deliver roughly one-third of the current liquid transportation fuels (Perlack *et al.*, 2005). A billion dry ton of lignocellulosic biomass feedstock is also a production target to supply the biobased industry by 2030 (Cushman *et al.*, 2003). Although the potential marine biomass supply is more uncertain due to the need for offshore technological research and development, the resource potential suggests that marine biomass could have significant impacts on the global supply of biofuels.

Preliminary Cost Estimate

The large potential resource driver must be reinforced by plausible economics of such systems. Essentially, costs must be roughly in line with the delivered (to the biorefinery plant) cost of terrestrial biomass resources, if processing of seaweeds to produce large amounts of biofuels, in addition to higher value products, is to merit serious consideration. It is possible to set some boundaries on the economics of such a process and compare them with prior efforts in this field. We base our initial estimates on construction of the dynamically positioned floating Offshore Seaweed Farm. We assume here a value of about 10 metric ton dry weight per hectare per year for seaweed production already achieved in near-shore systems. We further assume a production unit size of 1 km² (=100 hectares) producing 1000 metric ton per year. Current terrestrial lignocellulosic biomass (from forests, residues, or farmed biomass) can be estimated to have a delivered (to the biorefinery) cost of about \$75/metric ton (dry weight) ranging from \$50 to 100/metric ton. Corn is somewhat above \$4/bushel now, or about \$250/metric ton. Seaweed biomass, due to its high content of more readily fermentable carbohydrates, proteins and higher value co-products, can be assumed to have a similar value to that of corn, rather than lignocellulosic biomass, or about \$250 per metric ton of organic biomass delivered to the biorefinery plant gate.

This sets the limits on the possible cost of the Offshore Seaweed Farm. Assuming that 40% of the production costs would be for operations (harvesting, plant management, transportation to shore, etc.), and an annualized cost of capital (for depreciation and maintenance, about 15%, and a moderate return on investment of only 5%) of 20% per annum, this would allow an annual investment of \$750/metric ton-year production capacity. A 1 km² farm producing 1,000 metric ton of seaweed biomass annually would thus need to be established for a capital investment of no more than \$750,000, or \$7,500 per hectare. This would include support systems, such as harvesting and transport ships, seaweed nurseries, robots for planting, etc., which would be common to many such size farms, but would likely have a capital cost of at least half of that of the farm itself, which thus could likely not exceed \$2,000 per acre. Of this, a good part would be taken up by the positioning systems, limiting the cost of the grids and holdfast to which the seaweeds would be anchored to no more than \$2,500 to \$3,750/hectare. Clearly, this is a restrictive budget for even a minimum of grid lines to be strung out over this area. Although that may argue in favor of the very large open pen designs described in recent Japanese proposals,

these systems based on floating (i.e., not attached) seaweeds would have the fundamental problem of bunching of the plants which would not allow their efficient cultivation. A more detailed analysis is of course required, but it can be noted that the capital investments considered here, after adjusting for the lack of upwelling pipes, is not greatly different than those projected for the Marine Biomass Program and earlier studies. To offset these costs is the estimated revenues, which we set arbitrarily as \$100/barrel of oil, or about \$2.50/gallon of crude oil or \$3.00/gallon for a fuel ready to use (e.g. butanol, green diesel, etc.). Renewable fuels that avoid greenhouse gas emissions can be expected to have a premium of \$1/gallon equivalent, which is what biodiesel substitutes and next generation biofuels would receive in the U.S. under current incentive programs. A maximum projected yield of about 100 gallons of gasoline equivalent fuel from a ton of seaweed biomass can be projected, or about \$400/metric ton of biomass. We anticipate that fertilizers, animal feeds, and higher value co-products would add at least another \$100/ton, for total revenues of about \$500/ton of seaweed biomass. With the biomass costs delivered to the Marine Biorefinery set at \$250/metric ton, the biomass conversion costs would account for about half the total product value.

Environmental considerations

Allowing for the need for spacing between farms, to provide room for shipping and maneuvering of the farms themselves, the area of ocean needed for a billion ton of seaweeds would be significant. Upwelling zones are restricted to limited ocean areas, encompassing several hundred thousands of square miles. In the foreseeable future, this would not be a major space limitation. The total amount and local concentrations of upwelled nutrients is also not likely a limiting factor, as the total amount would be only about 30 million ton of nitrogen and about three million ton phosphate, only a small fraction of naturally occurring ocean nutrients. If recovered and contributed to the fertilizer industry, this amount would be a significant fraction of the 100 or so million ton of synthetic nitrogen fertilizer currently produced by industrial processes.

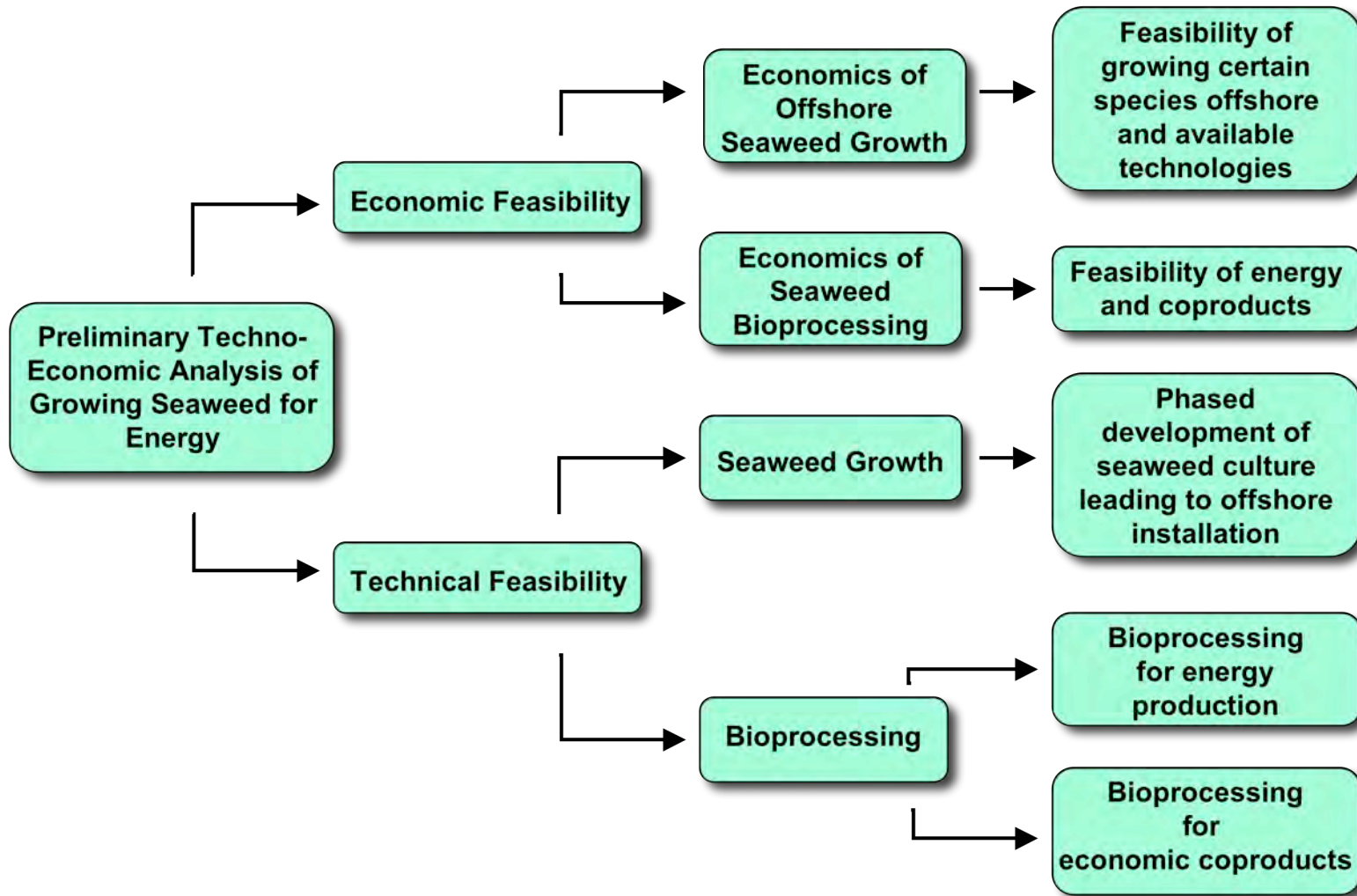
The environmental impact of offshore farms, as discussed in Chapter 7, must be weighed against the benefits of seaweeds as a renewable energy source. The oceans are absorbing one million ton of CO₂ per hour. Seaweed farming can recycle some of this back to beneficial uses by man. Although this must be viewed together with the energy required to sustain and operate

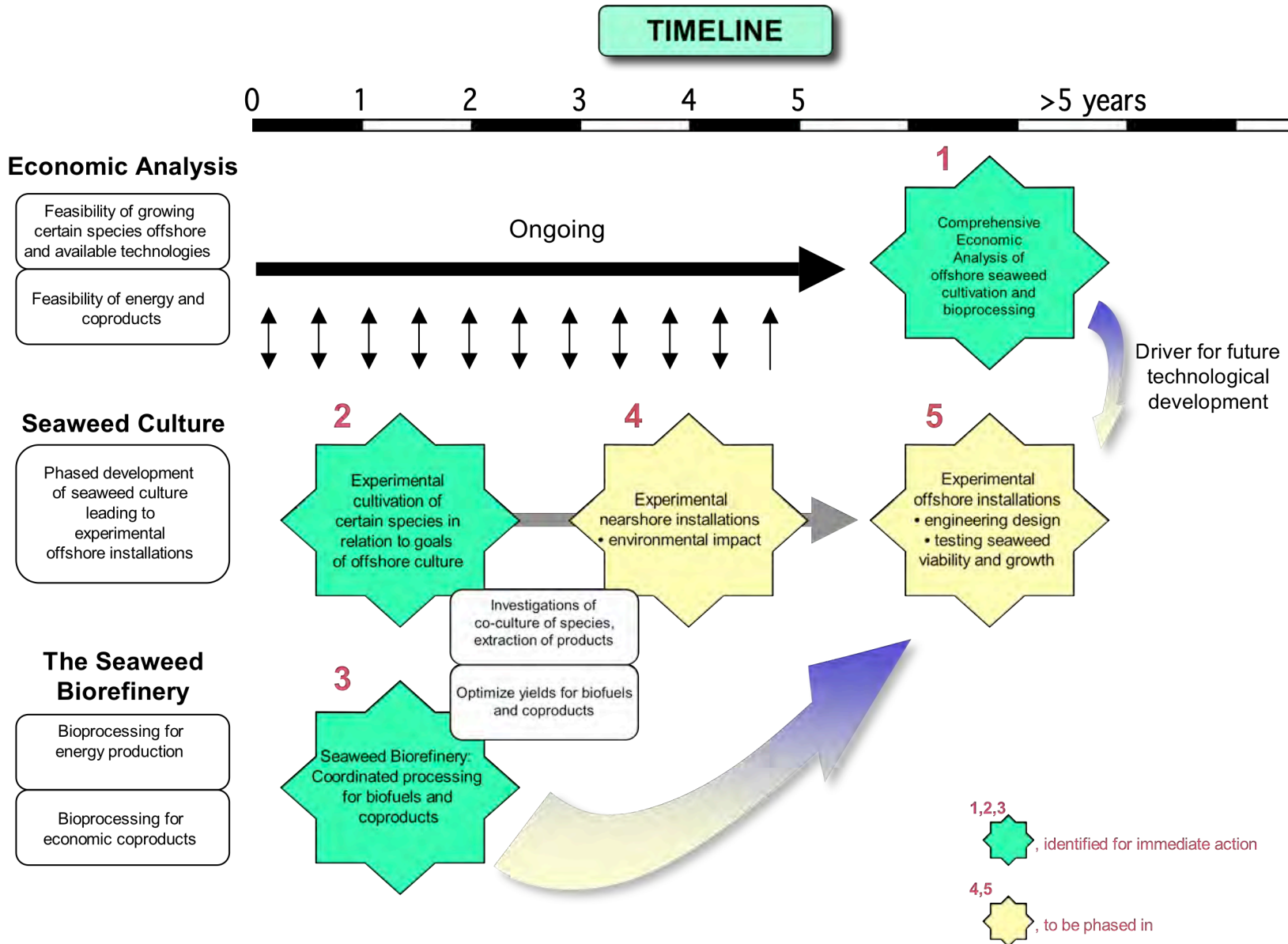
the farms, use of the oceans avoids problems currently being uncovered for diversion of terrestrial biomass for energy production (Searchinger *et al.*, 2008). Open ocean seaweed farming has, in principle, inherent advantages over terrestrial biofuels production systems, where the availability resources – land, nutrients, water – are much more limited for such large-scale biomass production.

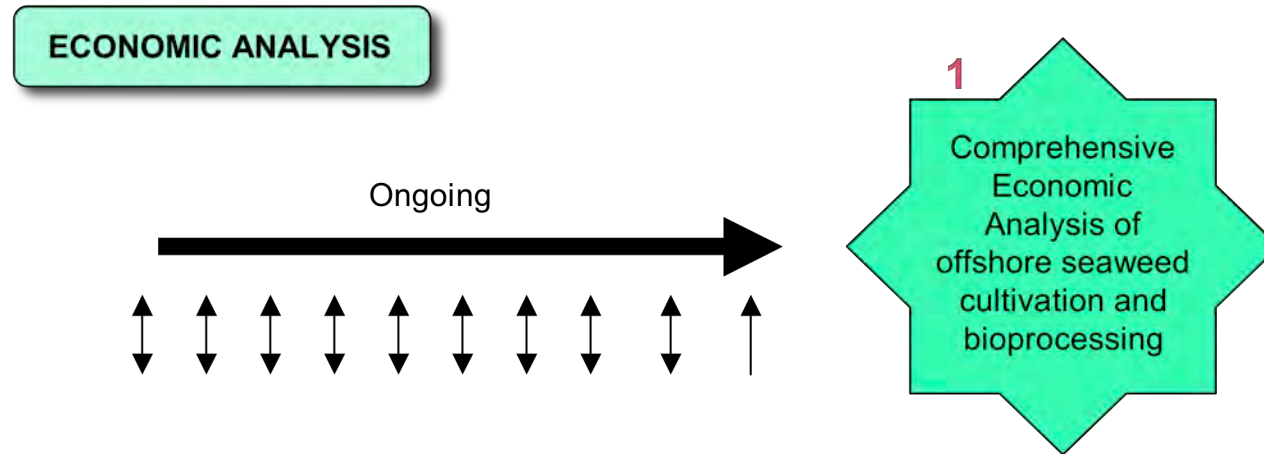
Visual Roadmap

A visual roadmap with activities targeted for immediate action is presented in the following pages. The Offshore Seaweed Farm is regarded as fulfillment of a long-range vision, with the intermediate activities serving as the initial priorities. Shown in the following panels is a recommended path of activity, with the initial focus on land-based laboratory and mesocosm research leading to nearshore deployment of seaweed life support systems. The structure used for nearshore studies can be viewed as prototypes to be tested for feasibility in later offshore, open ocean activities. In going to offshore seaweed farming, consideration of engineering design and stability of seaweeds in a dynamic physical is paramount. The intermediate activities focus on making advances in culturing methodology and improved bioprocessing for biobased products. We view these as having the potential for short-term commercial application. Ongoing economic analysis is also recommended to accompany the technical progress expected to be made throughout the program. Identification of suitable offshore sites will also need to be made. The actual time-frame can be expanded or contracted depending on the rate of progress and available financial resources. Paramount are establishing an understanding of the biological and engineering needs of growing seaweeds offshore, prior to making the venture into dynamic oceanic marine environments, and securing the capital investments needed to make it possible.

Roadmap for Growth of Seaweed for Energy and Coproducts







- The economic analysis is an ongoing exercise leading to assessment of the feasibility of offshore seaweed farming.
- The economic analyses associated with the inception of the Marine Biomass program in the 1970s represent the most recent analysis of offshore seaweed farming. Thus, a current analysis will need to assess those studies and their relevance to current conditions.
- An analysis of both growing seaweed offshore and the processing of seaweed into fuels and other valued products should be examined in a coordinated fashion.
- Construct an economic model reflecting current advances, which can be modified by new information generated from future activities.
- Perform sensitivity analysis to determine which factors of offshore seaweed farming need optimization or improvement through further research and development.

TECHNICAL R&D (NEAR-TERM)

2

Experimental cultivation
of certain species in
relation to goals of
offshore culture

- Match species selection with following
 - Highest yields for energy and coproducts
 - Habitat: geographic location, upwelling areas, storm tracks
 - Factors affecting viability on growth structures
- Propagation and seeding growth structures
- Laboratory simulations of offshore systems to estimate productivity

3

Seaweed Biorefinery:
Coordinated
processing for biofuels
and coproducts

- Biofuel production from seaweeds
 - Methane and ethanol are currently preferred biofuels from seaweeds
 - Second generation biofuels
 - Are seaweeds an economically viable feedstock for **butanol** production?
 - What species, apart from *Macrocystis*, can be suitable feedstock?
 - Optimize fermentation conditions to maximize butanol yield.
- Determine best procedure for maximizing production of both biofuels and coproducts.
 - Identify coproducts whose value is competitive with biofuels.
 - Sequential processing to obtain value-added product from seaweed feedstock, prior to conversion to biofuels.
 - Parallel processing of common feedstock source to obtain biofuels and “so-called” coproducts.
 - Conversion of any waste product to methane to anaerobic digestion.

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