LARVAL FEED ALTERNATIVES

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CONTENTS

INTRODUCTION

There are two major reasons why non-living produced feeds for rearing larvae of aquatic animals do not yet have an advantage over live food organism. These are: rapid deterioration of water quality due to disintegration of micropellets, which are usually fed in excess in order to achieve satisfactory growth and survival; high mortality rates, due to malnutrition and/or incomplete digestion of diet components. Cultivation of larval stages of various aquaculture species is still highly dependent on live food which is for herbivorous larvae, like molluscs and crustaceans, a fairly understood task. Many more difficulties have to be faced when live food animals are required, as is mainly the case in fish rearing, but holds true for latter stages of crustacean larvae as well. The reason why live food is so essential for larval growth has not yet been clearly defined. Enzymes present in phyto and zooplankton but not synthesized by the physiological system of a larvae are probably important. Also of importance are several essential biochemical compounds such as poly-unsaturated fatty acids, most of which have been defined as to species requirements. Primary producers of these fatty acids such as algae and bacteria form the base of the trophic pyramid, and as such constitute the largest link in the aquatic food chain. The large-scale, intensive production of microalgae and rotifers suffers from two major problems: it is expensive and often unreliable. Contributing to the problem is the fact that designs used for experimental and pilot scale units, which are the bulk of the published research, are usually inappropriate for larger system because of logistical problems, prohibitive cost of materials, or diminishing surface area to volume relationships which affect scale up performance. Scale up problems can arise in the bulk handling of materials such as animals, water and feeds which in a restricted laboratory situation are easily transported and held in small containers. Carrying out necessary life support functions can also become complicated, since daily work routines for large numbers of animals quickly becomes prohibitive. Routine maintenance and cleaning of culture units, while trivial in the laboratory becomes a major problem with increased scale. As hatchery managers try to stem the rising costs of production, the economic cultivation of live feeds or some alternative becomes ever more important. The sections that follow will attempt to

illuminate various options and potentials for larval penaeid shrimp feeds. The summary section will then rate the most cost effective choices for management consideration.

ALGAE

There has been widespread interest in the mass production of microalgae since the 1940's. Microalgae have been cultured as a source of oils, polysaccharides, fine chemicals and oxygen. They have been exploited for soil conditioning, eutrophication control, waste-water treatment, and consumption by humans, livestock and aquatic organisms. At one time, mass culture of algae was seen as a solution to the world protein shortage and numerous other global problems. In most cases, however, large scale microalgal culture has fallen short of economic expectations due, in large part, to the expense and difficulties associated with separation of the cells from the culture medium and processing. A clear exception to this is the cultivation of microalgae to feed aquatic organisms, i.e., its use as a feed as opposed to food. In general, the most important problems encountered in the large-scale production of microalgae as feed for bivalves, crustaceans and rotifers may be classified as either 1) economic in nature, or 2) related to the dependable output and consistent quality of large volume production. Algal production costs for aquatic animals may be determined indirectly by estimating the percentage of total hatchery costs used to grow algae. Very large volumes of select algae are required for the nursery culture of bivalve molluscs, while smaller amounts are used in the larviculture of commercially valuable molluscs and penaeid shrimp. From the data shown in Fig. 1, it is possible to see that penaeid shrimp hatcheries use more of the total hatchery costs to produce algae than do bivalve hatcheries.

Fig 1. Percent total hatchery costs used for algae production, ration of algae culture volume to target species culture volume.

	USA Washington (Thalassiosira pseudonana 3H clone; 1990 date)	**USA Hawaii** (Nanno-chloropsis oculata)	**USA Louisiana** (chlorella minutis-sima	**USA Maryland** (Unidentified sp.; projections based on 5,000 - 500,00 liter fermentors	**THAILAND** (Chaetoceros calcitrans)	**N. CHINA** (Isochrysis galbana)
% Total hatchery operating costs used for algae production	18 (algae production cost is approx. $50/kg dry weight)	1/6 labor force devoted to algal production	N/A	N/A (production cost: $2 - 25/kg dry weight)	Approx. 15%	1/3 labor force devoted to algal production
Ratio of algae culture volume: target species culture volume	1 : 3 (oyster larvae)	4 - 5 : 1 (rotifers) : 1 (fish larvae)	N/A	N/A	10 : 1 (Penaeus larvae) [calculated from the total volume of Chaetoceros water used in each batch]	4 : 1 (Argopecten broodstock) 1 : 2 (Argopecten larvae)
Ranking of major costs (% total algae production cost)	1. Labor (37%) 2. Overhead (30%) 3. Lab supplies & chemicals (19%) 4. Energy	1. Labor 2. Energy 3. Supplies & equipment	1. Supplies & chemicals (energy & capital not included)	1.Carbon source (15-45%) 2.Capital (18 - 30%) 3.Other chemicals and equipment (5 - 25%) 4.Energy (10 - 20%) 5.Labor (2 - 5%)	1.Chemicals and supplies (35%) 2.Labor (24%) 3.Matinenance (18%) 4.Energy (6%) 5.Starter cultures (4%)	1. Supplies and chemicals (70%) 2. Labor (20%) 3. Energy and misc. (10%)

	TAIWAN (skeletonema costatum)	S. KOREA NFRDA (Pavlova Lutheri)	S. KOREA NFUP (Nannochloris oculata)	SINGAPORE (Nannochloropsis oculata)	JAPAN SNFRI (Tetraselmis tetrathele)	JAPAN NRIA (Synecococcus sp.)
% Total hatchery operating costs used for algae production	2%	N/A	30 - 40%	25 - 30%	N/A	Small
Ratio of algae culture volume: target species culture volume	10 - 15 : 1 (penaeid larvae)	5 : 1 (1 mm spat)	3 - 4 : 1	1.5 - 2.0 : 1	4 : 1 (Penaeus japonicus larvae) 8 - 10 : 1 (Rotifers)	5 : 1
Ranking of major costs (% total algae production cost)	1. Facility (44%) 2. Labor (37%) 3. Stock (9%) 4. Supplies (6%) 5. Energy (4%)	1. Facility 2. Supplies 3. Energy	1. Facilities and supplies (50%) 2. Labor (30%) 3. Energy (10%) 4. Other (10%)	1. Labor (50%) 2. Facilities and equipment (30%) 3. Energy (10%) 4. Supplies (10%)	1. Supplies and chemicals (45 - 55%) 2. Energy (30-40%) 3. Equipment (10-20%)	1. Supplies and chemicals (60%) 2. Labor (20%) 3. Energy (20%)

Another estimate of resources devoted to algal production may be derived from the ratio of algae culture volume to target species volume. The highest algae : target species ratios are for penaeid shrimp, which indicates that these facilities probably devote a great deal more resources to producing algae than to growing shrimp larvae or rotifers. Why are the economics of shrimp hatchery algae production higher than those of bivalve hatcheries? The answer lies in the efficiency of the mass algae culture systems used at these facilities. Most bivalve hatcheries are located in temperate areas where outside temperatures never exceed high end limits for the species being cultured. Their problem is one of too little light in the winter months to maintain algal division rates at acceptable levels. Generally, they have to add light to outside tanks during the winter months. On the other hand, penaeid shrimp hatcheries are located in tropical and sub-tropical areas where temperatures are in excess of optimal algae species criteria for at least half of the average production season. Light is also in excess in these tropical areas for outside cultures. Rather than design facilities around these environmental parameters as have in the bivalve hatcheries, shrimp culturists adapt phytoplankton species to their system irregardless as to whether they are of optimal nutritional value to the shrimp larvae. As a first step towards designing better production facilities, shrimp hatchery biologists and engineers need to take a critical look at existing systems, their yields, operating costs, reliability, etc., and consider ways of improving performance through better design. Mass algal production systems should be designed around the most nutritionally sound algae species available for a particular culture organism. What most penaeid hatcheries do is simply rely on culturing whatever grows best in their system. In most cases these "weeds" are not the best food for penaeid larvae. Culture age/growth phase, light intensity, temperature, nutrient limitation and source and cell density can all affect the chemical composition of the algae. The problem of consistent nutritional quality is also especially pronounced in outdoor cultures. When during up-scaling, an algal culture is transferred outdoors, the cells often suffer from photic shock. This is due to not being adapted to light of such a high intensity, and they require a period of time to adapt. After this "lag phase' if the algae survives, the higher temperatures and higher incident light levels in outside cultures generally cause accelerated growth rates which are not as easily managed as indoor cultures. The pH of the culture is not maintained within desired ranges and algae is more often than not fed at a less than optimal nutritional level or past exponential growth rates. Algal cells in exponential phase may have a different biochemical composition to those in stationary phase. Changes in basic culture parameters can also change the fatty acid profile of the algae. It is well known that lipids are the most energy rich of the nutrient classes, providing approximately 9 Cal/g compared

to 4-5 Cal/g for carbohydrates. The principal components of most lipids are fatty acids. Many marine animals appear to have a limited ability to synthesize the poly-unsaturated fatty acids (PUFA) 20:5(n-3) or EPA and 22:6(n-3) or DHA from precursor fatty acids in the linolenic acid family. The growth and survival of penaeid shrimp has been shown to increase when foods rich in EPA and DHA are included in the diet. The fatty acid profiles of 10 algae species are shown in Fig. 2. These algaes were grown at 20 degrees C. with 79-80 uE/m2/s2 on 12:12 light:dark cycle with Guillard´s F2 medium. Three of the species shown were sampled 6 months later under similar conditions to test whether similar compositional data would be obtained. Noteworthy here are the 20:5(n-3) and 22:6(n-3) levels in the typical larval penaeid shrimp algaes *C.gracilis*, and *T. suecica* related to the bivalve larval feeds *C. calcitrans*, *T. isochrysis*, and *P. lutheri*. In a series of experiments conducted at the Hawaii Institute of Marine Biology, *Penaeus vannamei* and *Penaeus monodon* larvae were fed various phytoplankton species diets and to evaluate their performance against each other. Larvae fed *Thalassiosira weisflogii* had an 82% survival versus 76% survival for larvae fed *Chaetocerous gracilis*. More important was the fact the larvae fed *Chaetocerous gracilis* had a mean dry weight of 78.6 ug per postlarvae while those fed *Thalassiosira weisflogii* attained a mean dry weight of 132.7 ug per post larvae!!!. In separate experiments, mean dry weights of larvae fed *T. weisflogii*, exclusively, compared favorably to weights of animals fed phytoplankton and *Artemia*.

Fig 2. Percentage composition of fatty acids in diatoms and prymnesiophytes.

Percentage composition of fatty acids in green algae and Chroomonas

	Green algae	Green algae	Green algae	Cryptomonad	Cryptomonad	Cryptomonad
Saturates	DUN	NAN	TET no.1	TET no.2	CHRO no. 1	CHRO no. 2
12:0	TR	TR	0.1	0.7	0.1	TR
14:0	0.2	0.6	0.6	0.9	8.6	8.2
15.0	TR	0.1	0.3	0.3	0.3	0.2
16:0	14.7	20.1	20.3	24.0	15.1	12.9
17:0	0.1	TR	TR	0.3	0.4	0.2
18:0	0.4	1.1	0.9	0.6	0.9	0.7
20:0	TR	0.1	TR	-	TR	TR
22:0	TR	TR	0.2	TR	0.1	TR
24:0	TR	0.1	TR	-	TR	TR
SUM%	15.4	22.1	22.3	26.8	25.5	22.2
	Green algae	**Green algae**	**Green algae**	**Cryptomonad**	**Cryptomonad**	**Cryptomonad**
Monounsaturates	DUN	NAN	TET no.1	TET no.2	CHRO no. 1	CHRO no. 2
16:1(n-10)	-	0.5	-	-	0.2	-
16:1(n-9)	0.1	1.3	0.9	1.2	0.1	0.2
16:1(n-7)	0.1	0.6	0.3	0.3	0.5	0.6
16:1(n-5)	-	-	-	-	-	-
16:1(n-13)*t*	2.7	8.9	1.5	0.8	1.3	1.2
18:1(n-10)	-	-	-	-	0.1	0.4
18:1(n-9)	2.0	4.9	12.3	14.5	2.9	2.3
18:1(n-7)	0.3	0.4	0.4	1.1	3.5	3.2
20:1(n-9)	-	-	1.6	2.6	-	-
SUM%	**5.2**	**16.6**	**16.7**	**20.5**	**8.6**	**7.9**
	Green algae	**Green algae**	**Green algae**	**Cryptomonad**	**Cryptomonad**	**Cryptomonad**
Polyunsaturates	DUN	NAN	TET no.1	TET no.2	CHRO no. 1	CHRO no. 2
16:2(n-7)	-	-	-	-	-	-
16:2(n-6)	0.7	4.2	1.1	1.8	-	-
16:2(n-4)	-	-	-	-	-	-
16:3(n-6)	-	0.3	4.6	6.0	-	-
16:3(n-4)	-	-	-	-	-	-
16:3(n-3)	4.2	14.4	TR	0.5	-	-
16:4(n-3)	21.0	-	13.7	7.9	-	-
16:4(n-1)	-	-	-	-	-	-
18:2(n-9)	-	-	-	-	-	-
18:2(n-6)	4.8	10.3	13.8	13.9	11.6	10.5
18:3(n-6)	2.7	TR	0.7	2.7	3.0	2.6
18:3(n-3)	43.5	21.7	11.1	4.6	11.9	14.2
18:4(n-3)	1.0	2.7	8.4	4.8	19.8	21.3
18:5(n-3)	-	-	-	-	-	-
20:4(n-6)	-	0.5	1.5	2.1	1.0	0.9
20:4(n-3)	-	1.1	0.3	0.1	0.9	1.0
20:5(n-3)	-	3.2	4.3	5.3	10.9	11.9
22:5(n-3)	-	-	-	-	TR	0.3
22:6(n-3)	-	TR	TR	TR	5.7	5.2
SUM%	**77.9**	**59.0**	**59.5**	**49.7**	**64.8**	**67.9**
Others	***1.5***	***2.3***	***1.5***	***3.0***	***1.1***	***2.0***
TOTAL%	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**

DUN (*dunaliella tertiolecta*) / NAN (*Nannochloris atomus*) / TET (*Tetraselmis suecica*) /CHRO (*chroomonas salina*)

Fig 2. (CONTINUED)

Percentage composition of fatty acids in diatoms and prymnesiophytes.

Saturates	Diatoms C.CAL	Diatoms C.GRA no.1	Diatoms C.GRA no.2	Diatoms SKEL	Diatoms THAL	Prymnesiophytes T.ISO	Prymnesiophytes PAV
12:0	TR	TR	TR	TR	TR	TR	0.3
14:0	17.5	8.8	11.6	20.1	14.3	16.0	11.5
15.0	0.8	1.0	1.2	1.2	0.8	0.5	0.5
16:0	10.7	23.3	17.8	16.5	11.2	14.5	21.3
17:0	0.3	0.3	0.2	0.6	0.1	TR	0.2
18:0	0.8	4.1	3.1	0.8	0.7	0.2	1.3
20:0	TR	0.3	0.2	TR	0.1	0.3	0.3
22:0	TR	0.6	0.6	TR	TR	0.6	0.2
24:0	0.1	0.3	0.8	TR	TR	TR	0.2
SUM%	**30.2**	**38.7**	**35.5**	**39.2**	**27.2**	**32.2**	**35.9**

Monounsaturates	Diatoms C.CAL	Diatoms C.GRA no.1	Diatoms C.GRA no.2	Diatoms SKEL	Diatoms THAL	Prymnesiophytes T.ISO	Prymnesiophytes PAV
16:1(n-10)	-	-	-	-	-	-	-
16:1(n-9)	-	-	-	-	-	0.3	-
16:1(n-7)	30.0	33.4	26.8	28.6	18.0	4.2	16.8
16:1(n-5)	0.1	0.1	0.2	0.6	0.3	-	TR
16:1(n-13)*t*	0.7	1.2	1.6	1.3	0.4	-	-
18:1(n-10)	-	-	-	-	-	-	0.3
18:1(n-9)	2.8	3.6	6.0	1.4	0.5	20.1	1.7
18:1(n-7)	0.2	1.7	3.9	0.1	0.1	1.3	1.4
20:1(n-9)	-	-	TR	-	0.2	0.2	0.2
SUM%	**33.8**	**40.0**	**38.5**	**32.0**	**19.5**	**26.1**	**20.4**

Polyunsaturates	Diatoms C.CAL	Diatoms C.GRA no.1	Diatoms C.GRA no.2	Diatoms SKEL	Diatoms THAL	Prymnesiophytes T.ISO	Prymnesiophytes PAV
16:2(n-7)	3.5	2.9	2.4	3.3	2.7	0.5	0.2
16:2(n-6)	-	-	-	-	-	-	-
16:2(n-4)	1.6	1.7	0.7	3.5	4.5	0.7	0.2
16:3(n-6)	-	-	-	-	-	-	-
16:3(n-4)	8.0	2.3	2.2	3.7	12.7	1.4	0.4
16:3(n-3)	-	-	-	-	-	-	-
16:4(n-3)	-	-	-	-	-	-	-
16:4(n-1)	0.3	-	TR	2.6	2.3	-	-
18:2(n-9)	0.8	2.0	4.2	-	TR	-	0.4
18:2(n-6)	0.8	0.5	0.7	2.2	0.4	2.5	1.5
18:3(n-6)	0.4	0.8	1.1	0.3	0.2	2.4	0.4
18:3(n-3)	TR	-	-	0.3	0.1	3.6	1.8
18:4(n-3)	0.5	0.2	1.2	2.2	5.3	17.4	6.0
18:5(n-3)	-	-	-	-	-	2.5	-
20:4(n-6)	5.7	4.5	6.2	-	0.3	-	TR
20:4(n-3)	0.2	TR	TR	TR	0.3	-	-
20:5(n-3)	11.1	4.6	5.7	6.0	19.3	0.2	19.7
22:5(n-3)	-	-	-	-	-	1.8	2.0
22:6(n-3)	0.8	0.3	0.4	2.0	3.9	8.3	9.4
SUM%	**33.7**	**19.8**	**24.8**	**26.1**	**52.6**	**41.3**	**42.0**
Others	***2.3***	***1.5***	***1.2***	***2.7***	***1.3***	***0.5***	***1.8***
TOTAL%	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**

C.CAL (*Chaetoceros calcitrans*) / C.GRA (*Chaetoceros gracilis*) / SKEL (*Skeletonema costatum*) / THAL (*Thalassiosira pseudonana*) / T.ISO (*Isochrysis sp.*) / PAV (*Pavlova lutheri*)

ROTIFERS

Rotifers comprise a phylum of microscopic filter feeding metazoans (multi-cellular organisms). They are composed of approximately 1,000 cells and filter small particles out of the water column by means of a ciliated corona located on the anterior portion of the body. The corona may also be used for locomotion: however many species spend the majority of their lives attached to substrate. *Brachionus plicatilus* is one of the planktonic or unattached varieties which

has become the most popular for aquaculture. Geographic strains of *B. plicatilus* range in size from 125 to 300 um in length. The animal can reproduce either sexually or as is more common, asexually. A female rotifer reproducing asexually simply produces clones; genetically identical copies of herself. A change in the rotifers environment such as a sudden increase or decrease in salinity or temperature can trigger sexual reproduction. At this time males are produced via special resting eggs or cysts similar to *Artemia* cysts. Normally aquaculturists rely on only asexual reproduction because the rate of asexual reproduction is faster than sexual reproduction and males are inferior nutritionally due to lack of a functional digestive system. The onset of sexual production in a rotifer population usually signals an impending culture collapse. Culture of *B. plicatilus* has become an indispensable aspect of many marine finfish hatcheries. This organism is an excellent first feed for larval fish because of its small size and slow swimming speed and habit of staying suspended in the water column, and ability to be cultured at high densities due to a high reproductive rate. Rotifers can easily be enriched with fatty acids, antibiotics or probiotics and used to transfer these substances to larvae. As with microalgae, there are many recognized techniques for culturing rotifers. Production may be extensive in large 50 to 150m3 tanks or intensive in small 1.0 to 2.0 m3 **(See Fig. 3).**

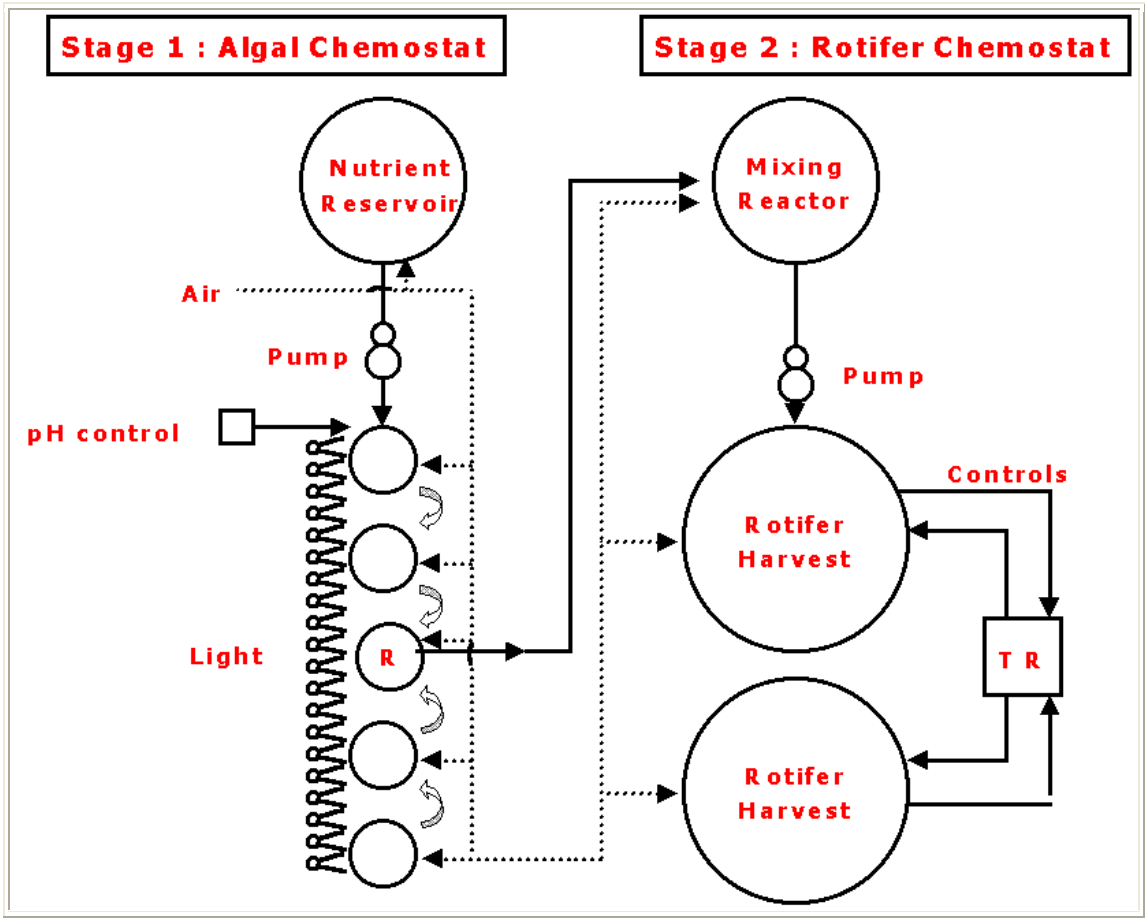
Figure 3: Culture methods and production of rotifers in Japan

Facility Name	Culture Method and B. plicatilis strain	Culture Tanks	Temp. (Celcius)	B. plicatilis inoculation density (N/ml)	Average Daily Production (10^8)	Amount of "Chloraila" used per 10^8 rotifers (m^3)	Amount of yeast used per 10^8 rotifers (kg)	B. plicatilis production per 1 ton of culture water
Miyazuki Prefecture Marine Testing Site	Batch S-type	4 tank 0.5 t panlite	27-32 (heated)	100-200	1.5	0.27	0.6	3.0
Hiroshima City Marine Promotion Association	Batch L- and S-type	7-8 tanks 26t concrete	6-29 (unheated)	80-150	62.6	0.009, concentrated Chlorella (0.28 m^3)	omega yeast (0.225)	2.4
Nagasaki Prefecture Fisheries Public Corp.	Batch S-type	4 tanks 80 t concrete	23-24 (heated)	80	25.7	0.69	0.64	0.6
Yamaguchi Prefecture Foreign Fish Hatchery Center	Semi-continuous L- and S- type	4 tanks 23 t and 21 t concrete	25-28 (heated)	100-130	61 (32)	0.75	0.28	1.7
Nagasaki Prefecture Marine Experiments Site of Culture Research Center	Semi-continuous L- and S- type	4 tanks 10 t concrete	27-32 (heated)	300	15-20	0.25	0.27	3.7
Nagasaki City Aquatic Center	Semi-continuous L-type	2 tanks 200 t concrete	18-25	31	17.4	1.26	0.65	0.96

In general though most culture methods are simply classified as either batch, semi-continuous, or continuous. Batch culture is generally an extensive method in which a culture is inoculated and allowed a growth period before the entire volume or a portion thereof is harvested. This approach has been the most reliable due to its technical simplicity but it is the least efficient. Continuous mass cultures are smaller than batch cultures and more intensively managed. A well managed large scale batch culture is used at the Hiroshima Prefecture Fish Farming Center. *B. plicatilis* are cultured outdoors in eight unheated 150m3 tanks. Each rotifer culture period lasts five days and the tanks are fed *Nannochloropsis oculata* and w-yeast. Tanks averaged 18.25 to 24.1 billion

rotifers production per day at a 300 to 500 rotifers/ml density at harvest. Semi-continuous culture done in large tanks yields less than the batch system per day but the amount of labor is also lower. The density of rotifers is maintained at 100 to 200/ml and cultures are harvested at a constant rate over a 20 to 60 day period. More rotifers can be produced from the same volume of culture water with semi-continuous and continuous rotifer culture systems. Semi-continuous culture systems suffer from a build-up of waste products in the form of feces and uneaten food, leading up to contamination problems with filamentous algae and fungal infections. It is imperative that they have some form of bio-filter and organic buildup removal system, but this adds to the cost of production. The use of Baker's or w-yeast to reduce algae feeding requirements for rotifers causes much of the water quality problems associated with semi-continuous culture systems. To grow a 500 liter culture of rotifers exclusively with algae would require approximately 5-10 times that volume of algae. For this reason rotifers are generally fed yeast at 0.4 mg/million rotifers/day and algae at 0.5-1.0 million cells/rotifer/day. Marine chlorella, *Nanochloropsis oculata* or similar species is the preferred algae for feeding rotifers since it is a small sized green algae, can attain densities of 50 million cells/ml in a short culture time and has a fair amount of poly-unsaturated fatty acids. This was the algae used by the Kuwait research team for their continuous rotifer production unit
(See Fig. 4).

Fig 4. Schematic diagram of the two stage rotifer chemostat system. A-Algal reservoir; TR-Temperature monitor and regulator.



The standard culture column of their rotifer system is 1m3. This tank is provided 20 million cells/ml/day from four 200 liter continuous *Nanochloropsis sp*. culture tanks which average 50 million cell/ml density. The algae is diluted in a mixing reactor before being fed to rotifers. In addition, Baker's yeast is provided at 0.3-0.4 g/million rotifers/day. A dilution rate of 0.5/day was used for the rotifers which allowed 500 l/day to be harvested from the 1.0 m3 chemostat each day. The average production from this system was 187 million rotifers per day run continuous over several months. This means that 5 or 6 m3 of this type of chemostat would be needed to produce one billion rotifers per day which is almost 100 fold less rotifer culture water than the best batch or semi-continuous culture system. An average days feeding for a 40 m3 shrimp larvae tank stocked with 100 nauplii/liter would be about one billion rotifers per day according to average penaeid larvae consumption of rotifers. It has been shown that protozoea 2 stage shrimp larvae consume and eat rotifers but that maximum feeding efficiency is P3 to M1 stage. At the P3-M1 stage both *P. kerathrus* and *P. indicus* larvae were shown to consume between 280 and 500 rotifers per day per larvae when fed a density of rotifers between 20-25 per ml. Reviewing the remainder of the work with rotifer ingestion by penaeid larvae we can see that about 250 rotifers per larvae per day is a good average to work from. Rotifer consumption drops dramatically after P2 stages as prey size decreases related to larval shrimp size. Feeding trials comparing *P. indicus* shrimp larvae fed *B. plicatilis* to *Artemia* showed some interesting energy consumption levels. In the *B.plicatilis* experiments a maximum consumption of 300 rotifers/larvae/day was obtained with P3-M1 larvae resulting in a Cal/larvae/day of 0.25. *P. indicus* P3-M1 larvae fed *Artemia* ingested 95 *Artemia*/larvae/day resulting in a Cal/larvae/day of 0.99. The investigators conclusion was that *Artemia* is a more efficient feed source and that rotifers could be dispensed within the hatchery. There was no cost comparison for *Artemia* vs. *B. plicatilis* use. This would be a very necessary study to come to grips with economic relationships and subsequent evaluations of the effects of "bioencapsulation" of high profile feeds, probiotics, etc.. Rotifers may yet become the most secure way to feed pure DHA and EPA to young shrimp larvae. The effect on larval survival and growth may be astounding enough to eliminate the need for economic comparison. What if a hatchery could produce larvae so consistently superior that farm growth and survival improved, say, at least 10% in each category every stocking?? Rotifers may not be the only ingredient for this futuristic scenario, but the science to improve performance is available to accomplish this and more with shrimp larval culture.

ARTEMIA NAUPLII BIOMASS

High density *Artemia* biomass culture systems were introduced in the 1970's with the development of batch culture techniques using micronized rice bran as a food source. The 5 kg/m3 wet weight biomass output of this system was too small to be commercially attractive, and led to the development of flow-through culture methods using phytoplankton as a food. The biomass output of these super-intensive systems improved to 25 kg/m3 wet weight but the cost-intensive automation and skilled personnel requirements still inhibited its commercial development. The newest systems have simplified the design and capital cost and achieve biomass production after 12 days of culture of 6.0 kg/m3 wet weight at a density of 8000 *Artemia* nauplii/liter and a dry cyst consumption of 40 g. This latest system uses micronized rice bran exclusively as a food source to achieve commercial feasibility. These *Artemia* biomass culture systems can be adapted for the controlled production of nauplii offering interesting prospects for aquaculture hatcheries. The controlled production of *Artemia* nauplii not only creates an independence from the international cyst market with its fluctuating prices and availability, but also gives a far better control on the quality of this live food product. Such an integrated production system would allow vertical integration of *Artemia* in the hatchery: the produced offspring can be used either directly as food source, or for stocking other *Artemia*

culture tanks which produce juvenile and reproducing brine shrimp to be fed to nursery and maturation stages of penaeid shrimp. The technique for controlled production of *Artemia* nauplii requires two essential modifications of the biomass production technique after stocking nauplii production tank with 14 day old adults from a biomass system:

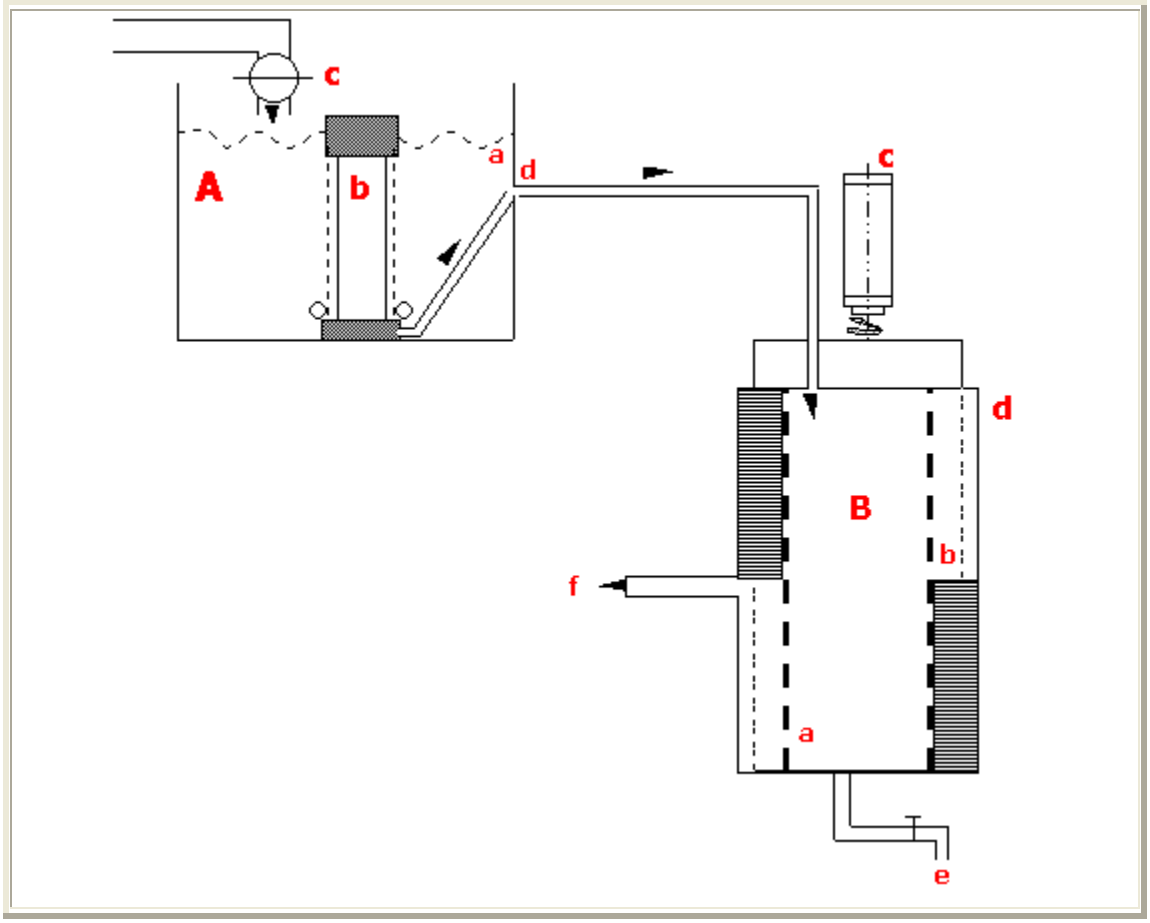
1. Specific diet and feeding strategy for reproducing adults.

In order to insure high survival rates and maximal reproductive activity, a much more complete diet needs to be offered to adults, including a mixture of rice and corn byproducts, single cell proteins and HUFA-enrichment. The fatty acid profile of the produced larvae is a reflection of the diet fed to the parental population. Feed requirements in dense cultures of adult brine shrimp cannot be dosed by transparency readings as with biomass culture because optimal food uptake in fully developed adults is already achieved at much lower concentrations. As a result of the decreased molting rates in adult *Artemia* the setae of the thoracopods become easily clogged with food at the higher particle densities. A daily feeding ratio of 10% dry weight feed to live weight biomass distributed on a semi-continuous basis will yield optimal production results.

2. Continuous nauplii harvesting technique.

The modifications to the biomass production systems consist of an inverted welded wedge screen cylinder of 150 um slit opening precisely fitted into a cylindrical PVC holding tank. The half submerged filter retains all produced nauplii and particles larger that 150 um from the culture effluent and drains water and smaller wastes via the holding tank back to the recirculating unit **(See Fig. 5)**. A mechanical cleaning system is required to avoid filter clogging. This consists of two brushes driven by an electrical rotor at 12 rpm to clean the outer edge of the welded-wedge screen continuously. The nauplii are harvested once or twice a day and separated from the waste materials by taking advantage of the photostactic behavior of the larvae. For this, a 100 liter cylindro-conical tank with a central overflow tube covered with a lid with hole cut over the overflow pipe with a light bulb installed. The nauplii suspension is poured into the separator-tank and a water flow is adjusted so as to let particles sediment and harvest only the nauplii attracted to the light with the overflowing water. Production trials with a 100 liter culture tank yielded 30 g wet weight nauplii/day. For a stocking density of 5,000 fourteen day old *Artemia* adults/liter this suggests a reproductive rate of 10 nauplii/female. If these numbers could be scaled up to full commercial scale they would translate to a daily production of 50 million *Artemia* nauplii/m3/day or the equivalent production of approximately 250 grams of purchased cysts, or roughly half a pound. There was no production cost analysis for this system but at a 10% dry weight baker's yeast feed to live weight *Artemia* biomass the cost is minimal including fish or squid oil enrichment. Baker's yeast consumption amounts to under 0.5 kilo per week/m3 culture. It is the fed algae amount which is difficult to evaluate since we do not know the ratio of algae to baker's yeast used in the reproductive adult culture tank trials. Biomass production of *Artemia* under super intensive systems using only algae as feed required 4,000 m3 of *Chaetocerous curvisetus* culture at a cell density of 45,000 cell/ml to harvest 25 kg wet weight *Artemia* biomass/m3.

Fig. 5. Schematic drawing of the *Artemia* nauplii production system. A. culture unit with 100 liter tank (a) *Artemia* retaining filter (b) and in/outflow (c,d). B. Nauplii recuperation filter with welded-wedge filter (a) cleaning brushes (b) driven by rotor (c) cylindrical holding tank (d) collector drain (e) and effluent drain to recirculation systems (f).



Ongrown *Artemia* Sizes for Feeding Shrimp Post-Larvae

A relatively unused option for feeding post-larval shrimp is to offer older and therefore larger, *Artemia* nauplii to post-larvae shrimp. It has been shown that when *Artemia* of progressively bigger sizes are fed, starting with newly hatched *Artemia* (0.6 mm) at PL-1 and ending with pre-adult brine shrimp (6.0 mm) at PL-20, satiation could be obtained by the volume of *Artemia* in the stomach rather than the number of organisms ingested. For instance, when the size of the available prey was 4 mm instead of 2 mm approximately half the number of *Artemia* were consumed. In terms of dry weight, ongrown *Artemia* of 4 mm weigh 8 times as much as *Artemia* of 2 mm. It is widely understood that *Artemia* must be enriched if fed to larvae more than a few hours after hatch to prevent nutritional deficiencies. Experiments have shown that feeding ongrown *Artemia* without enrichment resulted in lower shrimp body weights than shrimp receiving newly hatched *Artemia*. It is perhaps possible to view the use of ongrown *Artemia* as an extension of the current larvae culture practice of feeding 12 to 24 hour enriched *Artemia* to Mysis 3 and older postlarval shrimp. The following table (Fig. 6) shows a possible feeding routine using ongrown *Artemia* of progressively larger size.

Fig. 6. Ongrown Artemia feeding table for penaeid larvae.

PL age	Ongrown Artemia #/ml fed	Size of ongrown Artemia (mm)	Age of ongrown Artemia (days)	Dry weight Artemia (ug)
1	3-4	0.6	0.5	0.50
2	3-4	0.6	0.5	0.50
3	2-3	1.0	1.0	1.50
4	2-3	1.0	1.0	1.50
5	1-2	1.2	2.0	2.76
6	1-2	1.2	2.0	2.76
7	1-2	1.2	2.0	2.76
8	1	1.6	3.0	5.32
9	1	1.6	3.0	5.32
10	1	2.0	4.0	9.00
11	0-0.5	2.0	4.0	9.00
12	0-0.5	2.0	4.0	9.00
13	0-0.5	2.5	5.0	18.0
14	0-0.5	2.5	5.0	18.0
15	0-.25	3.1	6.0	36.0
16	0-.25	3.1	6.0	36.0
17	0-.10	4.0	7.0	72.0
18	0-.10	4.0	7.0	72.0

NEMATODES

Nematodes are close relatives to earthworms which may be either free-living in water or soil or parasitic on animals or plants. A particular species of free-living nematode, *Panagrellus redivivus* has been cultured since the mid 1930's by aquarists as a live food for a variety of fish species. Their small size and ease of culture has received renewed attention in recent years with rising costs and declining hatch out of brine shrimp eggs sold in the aquarium industry. In seeking an alternative to *Artemia* as an animal food source for aquatic larvae, it was noted that *Panagrellus redivivus* has as good if not better nutritional profile to that of *Artemia*. The nematode contains 48% protein, 21% lipids, 7% glycogen, 1% organic acids, and 1% nucleic acids. Approximately 70% of the lipids are fatty acids and the remainder are phospholipids. *Panagrellus redivivus* nematodes are about 0.5 to 2.0 mm in length and 0.05 mm in diameter with an average mean dry weight per individual of about 0.11 ug (as opposed to *Artemia* nauplii at about 2.69 ug each). Early experiments in feeding *Panagrellus redivivus* to *P. vannamei* larvae used this weight difference to suspect that feeding levels of 70 nematodes per ml would be the equivalent of 3 *Artemia* nauplii per ml. These feeding trials used Quaker Masa Harina mixed to the consistency of a thick paste with de-ionized water as a media for nematodes. Cultures were grown on 1.5 cm thick carpets on the bottom of plastic shoe boxes or cake pans. Cultures were incubated in semi-darkness at a temperature of 25-28 degrees C. Nematodes could be maintained for 4-6 weeks under this regimen if water was replaced on a daily basis to prevent dehydration. After a 5-6 day breeding period from an initial seeding of 10,000-20,000 nematodes, the cultures could be harvested every 2-3 days and yield a total of 40,000 to 50,000 nematodes per cm2 surface culture area. Although shrimp larvae could consume nematodes as early as P1 stage, early experiments found no significant growth parameter differences if nematodes were fed at P1, P2 or P3 stage, indicating that the critical larval nutritional needs were probably met by the diatoms being fed rather than nematodes. The results of early shrimp larvae experiments were somewhat mixed since the extent of actual utilization of nematodes by shrimp larvae was not determined. Results did conclude that nematodes could replace *Artemia* satisfactorily if fed from the P2 stage when *Artemia* was fed at the M1 stage. Feeding *Artemia* plus diatoms at P2 stage could not be matched by feeding nematodes plus diatoms at the same stage. The conclusions were that *Artemia* nauplii could not be completely eliminated from shrimp larval diets but that

they could satisfactorily be replaced by at least 50% with nematodes at no different production performance. A series of more recent tank trials have shown that with increased feeding levels of nematodes, the growth, survival and metamorphosis of larvae fed nematodes plus algae was equal to that of the control tank fed *Artemia* plus algae. All of the feeding trials with *Panagrellus redivivus* were done with organisms grown on the traditional media of wheat flour, or Masa Harina. The fatty acid profile of nematodes grown on this media show the animal to be lacking in essential fatty acids 20:5(n-3) (EPA) in relation to *Artemia* but with a higher level of 20:4(n-3) (ADA) than *Artemia*. Neither nematodes nor *Artemia* contain very high levels of 22:6(n-3) (DHA) which is thought to mastermind growth and survival in most marine species. More recent studies on enriched media for nematodes has shown encouraging results. Nematodes grown on wheat flour plus fish oil contained a higher percentage of n-3 HUFA (11.15% of total fatty acids) especially EPA (7.35%) and DHA (3.25%) **(See Fig. 7).** *Panagrellus redivivus* with this type of lipid profile offers interesting possibilities for *Artemia* replacement if water quality can be maintained and cost effective nematode production achieved.

Fig. 7 - Percentage of fatty acids (wt.) in total lipids extracted from nematodes cultured on wheat flour media with fish oil and two strains of *Artemia*.

Fatty Acid	Nematode (WFFO)	Artemia (GSL)[a]	Artemia (SFB)[a]
12:0	0.2	0.26	0.08
14:0	4.67	0.78	1.24
14:1(n-5)	1.52	0.98	0.36
16:0	12.89	14.12	11.11
16:1(n-7)	10.46	20.52	3.34
17:0	0.42	1.11	1.67
18:0	4.7	7.51	4.07
18:1(n-7)	11.28	8.78	7.70
18:2(n-6)	9.91	8.23	4.78
18:3(n-3)	9.28	28.19	3.76
20:0	0.23	0.63	0.6
20:1(n-9)	1.02	0.58	0.56
20:3(n-3)	0.44	0.27	0.34
20:4(n-6)	4.64	1.72	2.15
20:5(n-3)	7.35	1.19	9.32
22:0	0.47	0.27	0.04
22:1(n-9)	1.52	0.12	0.10
22:2(n-6)	0.78	0.36	0.12
22:4(n-6)	0.08	0.01	0.01
22:5(n-3)	0.11	-	-
22:6(n-3)	3.25	-	-
%(n-3)	20.41	13.44	29.68
% Saturates	**23.56**	**22.13**	**22.54**

CRUSTACEAN TISSUE SUSPENSION

A novel and inexpensive feeding system has been used for the mass rearing of penaeid shrimp larvae in India. The system is based on the exclusive use of a crustacean tissue suspension as a feed for all larval and early post-larval stages. It was developed out of the need to have an inexpensive feeding strategy which could be readily adopted by the owner-operator or rural farmer with limited resources. Its use has for the most part been restricted to the poorer countries where shrimp hatcheries lack highly trained labor, sophisticated live food production systems, and high capital investment for expensive procurement such as *Artemia* cysts. The feeding of tissue suspension consists of the following routine: The larval population is estimated by sampling every day so that a percent of biomass calculation for tissue suspension can be made

per larval stage. If a tank contains 350,000 Z1 larvae, their feed requirements per day are 175 grams (See Fig 8.). About 200 grams of raw material is taken out of the freezer, thawed in seawater and blended with sea water in an electric blender. It is then sieved through a 250 um screen and the material which passes is then boiled for about 10 minutes. This allows the blended tissue to solidify and the liquid to separate. After cooling, the entire contents are poured into a fine muslin cloth, allowing the clear liquid to filter through. The solid matter in the cloth is firmly squeezed to remove as much liquid as possible and then blended with seawater into a fine suspension. This is passed through a 50 um screen, and the volume made up to 600 ml for storage in a refrigerator to be dispersed at 5 hour intervals to the tank.

Fig 8. Fed ration, feed particle size and feeding procedure for rearing Penaeid shrimp larvae.

Larval stage	Feed raw material for 100,000 larvae per day (g.)	Particle size of feed suspension (microns)	Suspension dilution	Suspension ration for 100,000 larvae	
				per day (cc)	per feed (cc)
Z1	50	below 50	3 times	150	30
ZII	70	below 50	the bulk	210	42
ZIII	90	below 150	of raw	270	54
M1	110	below 250	material	330	66
MII	140	below 300		420	84
MIII	170	below 400		510	105
PLI	200	below 500		600	120

The crustacean species most commonly used for tissue suspension preparation are Metapenaeus afinis, M. dobsoni, Parapenaeopsis stylifera, Acetes indicus, Nematopalaemon tenipes, Mesopodopis sp. (Mysids) and the stomatopod Oratosquilla nepa. These shrimp have a very seasonal commercial value in the countries where they are found. These same feeder species were evaluated for performance nine penaeid species: P.monodon, P. merguiensis, P. indicus, P. semisulcatus, Metapenaeus affinis, M. brevicornis, M. dobsoni, and Parapenaeus stylifera. Mean survival of 43.8% from N6 to PL1 was obtained for P. indicus, 25.3% for P.monodon, 72.0% for P. semisulcatus, 32.9% for M. monoceros 62.5% for M. dobsoni, and 30.8% for P. stylifera. However, although the culturists claim that the shrimp larvae were fed exclusively on a non-living diet, the fact that algal diatom blooms occurred in the tanks indicates the presence of a live food organism. Some tanks in their experiments were dumped due to "excessive algal blooms" meaning perhaps overfeeding and deteriorated water quality. Even without a complete interpretation of the effects of diatoms in the tanks on tissue suspension feeding, the results were indeed very encouraging. Later experiments carried out by FAO investigators using more controlled systems compared various feeding options and found the survival of P.monodon as follows:

1. Dry Acetes feeding option 46.7%
2. Frozen Acetes option 29.4%
3. Cultured live food 24.7%
4. Fertilizers/dry Acetes 16.6%
5. Fertilizers/live food 16.0%
6. Fertilizer/frozen Acetes 16.0%

A major difference in this trial as opposed to earlier commercial runs in India is that the tissue was not boiled prior to being fed, and a dried tissue suspension was evaluated which obtained a significantly higher survival than other feed strategies. This might indicate some water quality problems affecting larval survival from feeding fresh material to the shrimp larvae, or some nutrient leaching caused by freezing the tissue prior to preparation. None the less, the system of larval rearing with crustacean tissue suspension as the exclusive feed has been demonstrated commercially feasible. If the system works in one place it should work in others and with other penaeid species. As a replacement for Artemia only, instead of for all early stage feeds, crustacean tissue suspensions may work very well for the culture of P. vannamei. Capture of suspended particles should be greater for Z3 larvae stages and larger, and assist in the maintenance of water quality. In addition, retaining the use of high quality algae in the larval tanks along with the crustacean tissue suspension may provide a total or partial replacement of Artemia.

SUMMARY

Reviewing our live feeds alternatives (rotifers, Artemia nauplii, and nematodes) we find two common denominators. One, there is a lack of complete experimental application with these feeds in the culture of larval penaeid shrimp, and two, there is insufficient economic breakdown of commercial production costs for these alternatives. Let's examine each possibility from an economic point of view.

Rotifers:

Rotifers are too small to be of much use after M2 larvae stages. Therefore, they might be able to replace a portion of Artemia usage between Z2 and M3 (assuming Artemia is fed at Z2 which it is usually not). Even at maximum feeding densities, rotifers do not have the energy of Artemia nauplii fed in equal proportions. Enriched rotifers would probably offset this and perhaps even surpass non-enriched Artemia nauplii, but this increases our cost of production. The Oceanic Institute manages fourteen 1,200 liter B. plicatilus tanks in batch system for fish larvae culture. Using a mixture of Baker's yeast and Tetraselmis chuii, their estimated cost of production was US$0.65/million rotifers produced. This is almost an identical rotifer production cost to the IFREMER sea bass hatchery in Brest, France. The only other available rotifer production costs are from other research institutes and are substantially higher. Commercial hatcheries should be able to reduce these costs dramatically since research scientists are far better paid than third world workers. For sake of example let's project a cost of half this of US$0.32/million rotifers. To feed a 40 MT larvae tank stocked at 120 shrimp larvae/liter at 250 rotifers/larvae/day we need to produce 1.2 x109 rotifers per tank per day. The cost of this at US$0.32/million would be US$384.00 per tank per day. For 5 days feeding from Z2-M2 this comes to US$1,920.00. Feeding Artemia to this same 40 MT tank between 3 and 6 nauplii/ml/day from Z3-M2 would require 7.2 x 108 Artemia nauplii. At a standard hatch of 2.0 x 105 nauplii/gram cyst we would need 3.6 x 103 grams of cysts, or 3.6 kilos of Artemia. To economically equate to the cost of feeding rotifers, the cost of the Artemia would have to be US$242.00 per pound. Obviously, there is a lot of room to play with this scenario. Using a cost of US$25.00 per lb for Artemia we can calculate that the equivalent cost for producing the required amount of rotifers has to be around US$0.033 per million. Even with a potential reduction in the amount of rotifers fed through enrichment practices to increase the energy levels, this cost difference is very hard to overcome.

Artemia nauplii:

On paper, the logistics of producing Artemia nauplii look workable if there is a reasonable cost factor for the algae which must be fed to maintain high productivity and nauplii quality. Assume for the moment, that the cost of Artemia cysts was US$25.00/lb. Also assume we could produce one half a pound per day of cyst equivalents in live nauplii from biomass per m2. Therefore, we have to produce 50 x 106 nauplii/day/m2 for US$12.50/day. But we don't start to get production from our adults until day 7 (21 days from hatch) after stocking in the nauplii production tank. In fact, the peak reproduction will not occur until day 21-35, so we must extrapolate the cost per day over an extra week period where we have too little production to utilize. Our productive life for adult Artemia can be several months in the wild but our survivals in culture will go below cost-effectiveness by day 35+. Let's assume we can produce 5.0 x 107 nauplii/day from days 21-35, survivals and all included. We would have produced 7.0 x 108 nauplii from our 14 day "productive time" total run or about 7.0 pounds Artemia cysts or about US$175.00. We need to feed a concentration of 5,000 adult Artemia/liter or 5.0 x 106/m2 a total of 140 grams dry weight enriched yeast, rice bran, or similar product, and algae per day. Let's say we fed 80% by-product and 20% algae. We then need 100 grams by-product enriched with 12 grams fish oil or equivalent, to give us 112 grams feed per day or 80%. We then need 28 grams dry weight algae or roughly 500 liters per day at a cell density of 1.0 x 106 cpm. For a complete production run of 21 days we would have to feed a 1.0m2 nauplii production tank 10.5 MT of live algae, 2.1 kilos by-product and 252 grams of fish oil, plus operation expenses. It would appear that the algae cost alone would surpass the breakeven point for Artemia cysts of US$175.00. Remember, too, that we would need roughly 2.3 x 108 Artemia nauplii per day average (maximum 4.0 x 108) for our 40 MT shrimp larvae tank. This means we would require about 8 m3 of Artemia nauplii production tanks per 40 MT shrimp larvae tank to replace Artemia cysts during our maximum demand. This may not be as conservative as some hatcheries operate but the scope of an Artemia nauplii production facility is similar to that of rotifers, in that it requires a fairly large infrastructure to start with to even begin to get these costs in line.

Nematodes:

An economic analysis of feeding media enriched nematodes to Z2-M3 stage shrimp larvae, begins with the assumption that we need to feed 100 nematodes per larvae/day. To feed our 40 MT tank stocked at 125 shrimp larvae/liter we need to produce 5.0x108 nematodes per day. Experiments with fish-oil enriched wheat flower showed a somewhat low productivity of 14,000 nematodes per cm2 culture area per week, or about 7,000 nematodes every 3.5 days. We can estimate conservatively that we would need three out-of-synchrony cultures of one cm2 each with one extra culture incubating to produce 7.0 x 103 nematodes/day. (4.0 cm2 nematode culture = 7.0 x 103 nematodes/day). To feed our 5.0 x 108 nematodes daily would require 2.8 x 105 cm2 media culture area, or approximately 2.8 m2. The cost of producing these nematodes in minimal. Masa harina costs about US$0.20 a kilo and we need 5 kilo/m2 plus 6 liters cod liver oil at US$4.00/liter, plus 1/2 kilo Baker's yeast at US$2.00/kg. Therefore, we need about US$26.00/m2 x 2.8m2 nematode culture per 40 MT tank = US$72.80. But the nematode culture will last 3-4 weeks for this cost or enough time to supply 4-6 larvae culture tanks. Artemia feeding between Z3 and M3 stages will average 3-7 nauplii/ml or total of 5.0 kilos or 11 lbs of Artemia cysts. At our reference price of US$25.00/lb Artemia cyst, this amounts to US$275.00 or about 3.8 times the cost of nematodes if labor costs are about equal, without dividing the

media cost of the nematodes among the 4-6 larvae tanks it should be able to feed. This is a big savings, and forces us to ask why enriched media nematodes have not gone into commercial shrimp hatchery systems especially during the frequent periods of hyper-inflated Artemia costs. One reason is that shrimp biologists are pretty much restricted to a small sphere of influence. They tend to only know what their neighbor is doing, and if he is doing it wrong, then chances are they are doing it wrong as well. On the other side, nematodes are less buoyant than nauplii, and will die and decay in saltwater within 72 hours after introduction unless eaten. Aeration and water exchange may have to be increased to support the use of nematodes on a commercial level, and this is probably too much work for a great number of existing hatcheries.

Algae:

Outwardly, it would seen that the cheapest method of providing higher levels of EPA and DHA for larval shrimp would be through the culture of those algal species rich in these fatty acids. In general, the literature states the costs for live algae production for aquaculture to be somewhere in the range of US$50.00 to $600.00/kg dry weight, with the majority falling in the $200.00/kg dry weight. Several commercial facilities are selling centrifuged algae paste for $80.00/kg, which would put the dry weight cost at closer to $200.00/kg. The upper end of this cost range is to be found in facilities which use artificial light to culture algae. Fluorescent tubes or other types of artificial lighting can account for about 98% of the cost of culturing algae indoors. It is safe to say that economics for the controlled culture of the high HUFA microalgae species, such as Thalassiosira weisflogii, Isochrysis galbana, or Rhodomonas balteca, will fall in the middle to upper spectrum of the surveyed production costs. A hatchery could afford these types of algae production costs only if the amount of the algae being produced were reduced to, say, smaller amounts for the critical larval stages, or to just the first or second larval feeding of a culture. This downsizing would require the use of suitable dry algae diets for a larger percentage of the larval requirements.

Crustacean tissue suspension:

This relatively simple technology is by far the cheapest Artemia substitute available. However, one of its chief reasons for success in certain areas of the world is the large populations of small, not commercially important, shrimp such as Metapenaeus sp., etc.. Not many western hatcheries are located close to these types of "trash" shrimp. Also, the effect on water quality in typically high density Western style hatcheries remains undocumented. Less problems relating to water quality are to expected from the use of dry tissue suspension as opposed to fresh or boiled tissue suspension from either shrimp or squid. However, while the drying process will cause the tissue suspension to behave more like a good microbound larval diet, the cost of the feed will also reflect this similarity.

RECOMMENDATIONS:

Our analysis of live feed alternatives for shrimp larvae culture has basically shown reason why most of these alternative have never been put into common commercial reality. Most remain in the research area of shrimp culture or in regions where there is a lack of availability of commercial feeds at economical prices. The infrastructure for culturing live feeds is perhaps the single most inhibitor of the development of these feeds. Most hatcheries are built around using

live algae and packaged artificial feeds. Building a new "mini" hatchery for the purpose of culturing alternative live feeds is not something most shrimp industries are willing to undertake. Especially since there is no historical commercially demonstrated facilities to lower the risk factor and learning curve. It is strongly felt that the use of the most economical dry diets (those containing the highest DHA-EPA concentrations for dollar value) coupled with reduced Artemia dependence is the best management strategy. Many hatcheries already realize the benefits of using enriched Artemia to promote increased larval survival and vigor. It is a simple extension to use ongrown Artemia of increasing age to decrease the amount of Artemia fed to larval shrimp. The infrastructure to accomplish this is minimal, the daily diets and available enrichments are available commercially, and the know-how for Artemia feeding are commonly understood worldwide.