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Lipid requirements of shrimp

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Abstract. — *A review of lipid requirements of shrimp and other crustacean species, when applicable, is presented. Qualitative requirements of crustaceans for cholesterol, fatty acids and phospholipids have been documented, but quantitative requirements generally remain underfined. Potential interrelationships between different classes of lipids that may influence requirements as well as differences due to age and species need to be investigated. A more accurate definition of lipid requirements can be achieved by supporting traditional evaluations based upon weight gain and survival with biochemical or histological evidence.*

INTRODUCTION

No absolute dietary lipid requirement for shrimp exists. Rather, provision of sufficient lipid is based upon the satisfaction of requirements for specific nutrients such as fatty acids, sterols and carotenoids and for energy. Requirements for these lipid classes may vary according to diet composition and the species under consideration. Research devoted to defining the best quantity and quality of dietary lipid has been principally confined to juvenile and subadult forms of shrimp. Most studies have used individuals having initial body weights below 10 g and generally within the range of 0.1 to 1.0 g. This review of the lipid requirements of shrimp also provides information from research results involving other crustaceans species whenever there is apparent applicability.

REQUIREMENTS

Dietary Lipid

Past nutritional studies with crustaceans indicate that the best survival and growth responses are achieved when the dietary level of one or a mixture of oils is between 5 and 8 % (Table 1). In most of these studies, however, the level observed to be best is ultimately influenced by the quality and quantity of dietary protein, the amount, type and availability

Tab. 1. — Summary of observations concerning EFA nutrition of shrimp.

Investigation	Species	OBSERVATIONS		
		Preferred dietary PUFA 18 : 3n-3 (*) vs 18 : 2n-6 (+)	Limited conversion of 18 : 3n-3 to C20, C22 HUFA (+)	Greater nutritive value of 20 : 5n-3, or 22 : 6n-3 vs 18 : 3n3 (+)
Bottino <i>et al.</i> (1980)	<i>P. setiferus</i> <i>P. aztecus</i> <i>P. duorarum</i>	--	+	--
Guary <i>et al.</i> (1976)	<i>P. japonicus</i>	*	--	+
Kanazawa <i>et al.</i> (1977a)	<i>P. japonicus</i>	--	--	+
Kanazawa <i>et al.</i> (1977b)	<i>P. japonicus</i>	*	--	
Kanazawa <i>et al.</i> (1978)	<i>P. japonicus</i>	--	--	+
Kanazawa <i>et al.</i> (1979c)	<i>P. japonicus</i>	*	--	--
Kanazawa <i>et al.</i> (1979d)	<i>P. japonicus</i>	--	--	+
Read (1981)	<i>P. indicus</i>	--	+	--
Colvin (1976)	<i>P. indicus</i>	--	+	-

of other energy sources and the source of oil. Nevertheless, high dietary levels of oils are usually associated with significant growth retardation. As levels increase, the amount in the hepatopancreas has been found to increase correspondingly. Exceedingly high levels may not be effectively processed. In addition, food intake is generally thought to be a function of energy supplement and excess dietary lipid may therefore inhibit appetite (Church and Pond, > 1982). A significant reduction in the growth rate of *Palaemon serratus* was observed when the level of corn or cod liver oil was increased from 7.5 % to 15 % (Forster and Beard, 1973). Deshimaru *et al.* (1979) found that when a dietary mixture of pollack liver oil and soybean oil was increased beyond 6 %, the growth rate of *Penaeus japonicus* was reduced. Castell and Covey (1976) used 1 %, 5 %, 10 % and 15 % levels of cod liver oil as a lipid source for adult *Homarus americanus* and found the 5 % dietary level to be most beneficial. Growth was not significantly improved when the cod liver oil was increased to 10 % and 15 % levels. Kanazawa *et al.* (1977a) observed a reduction in body weight gain of *P. japonicus* when a dietary level of 16 % was used. Davis and Robinson (1986) used menhaden fish oil as a lipid source (0 to 15 % in 3 % increments) for crayfish *Procambarus acutus acutus* and found reduced growth of crayfish fed diets containing 9 % or more lipid.

Growth of shrimp is superior when diets contain lipids (oils) of marine rather than vegetable origin (Guary *et al.* 1976; Kanazawa *et al.*, 1977a). Powdered pollack residual oil and short necked clam lipid were better dietary lipid sources than soybean oil for *Penaeus japonicus* (Kanazawa *et al.*, 1977a). A marine and vegetable oil mixture has generally achieved the best results (Read, 1981; Deshimaru *et al.*, 1979). Deshimaru *et al.* (1979) found that 6% of a mixture of pollack liver oil and soybean oil provided in a ratio between 3 : 1 and 1 : 1 was associated with high growth and feed efficiency of *P. japonicus*.

Fatty Acids

Fatty acids may be divided into four different groups as follows :

- (1) fatty acids that can be synthesized *de novo* from acetate. This group includes all even carbon number, straight chain, saturated fatty acids up to 20 or 22 carbons. The most abundant is 16 : 0 (palmitic acid). Shrimp, like other animals, apparently possess a delta-9-desaturase enzyme system which can convert these saturated fatty acids to monoenoic (monounsaturated) forms,
- (2) unusual fatty acid groups which include :
 - (a) odd carbon number fatty acids such as 13 : 0, 15 : 0, 17 : 0 and 19 : 0.
 - (b) non-methylene interrupted fatty acids which have two or more double bonds that are separated by more than three carbons;
 - (c) cyclopropanoic and cyclopropenoic fatty acids,
- (3) essential fatty acids (EFA) composed of two families of polyunsaturated fatty acids (PUFA). The linoleic (n-6) family has the first double bond at the sixth carbon from the methyl end of the molecule and has the greatest EFA value for homothermic animals. The linolenic (n-3) family has its first double bond between the third and fourth carbon from the methyl end. Neither of these two families of fatty acids are synthesized *de novo* by crustaceans ((Kayama *et al.* 1980) and the linolenic family has been observed to have the greatest EFA value for marine animals (Castell and Boghen, 1979),
- (4) essential fatty acids (EFA) composed of the linoleic and linolenic, n-3 and n-6, families of highly unsaturated fatty acids (HUFA).

Castell (1983) and Chanmugam *et al.* (1983) indicated that the body tissue of marine crustaceans generally tends to contain proportionately higher levels of fatty acids of the linoleic family of HUFA and PUFA than that of freshwater crustaceans. The freshwater species tend to have higher levels of acids of the linoleic family.

Kanazawa and Teshima (1977) injected juvenile *Penaeus japonicus* with acetate-¹⁴C and found activity almost exclusively associated with the saturated (16 : 0, 18 : 0) and monounsaturated fatty acids (16 : 1, 18 : 1 n-9, 20 : 1 n-9). Kanazawa *et al.* (1979a) also showed that *P. japonicus* has the ability to incorporate palmitic acid (16 : 0) into saturated and monosaturated fatty acids, but little is transformed into PUFA or HUFA,

Tab. 2. — Summary of investigations that evaluated response of crustaceans to quantity and quality of dietary lipid.

Investigations	Species	Lipid Sources	Lipid Level (%)	Protein Source and Amount	Observations
Castell & Covey (1976)	<i>Homarus Americanus</i>	cod liver oil	1, 5, 10, 15	50 % casein	best weight increase with 5 % CLO
Davis & Robinson (1985)	<i>Procambarus acutus acutus</i>	menhaden fish oil 12, 15	0, 3, 6, 9, 12, 15 + 4.5 % gelatin	28.1 % casein 0-6 %; growth reduction at 9 % or more lipid	no difference among
Deshimaru <i>et al.</i> (1979)	<i>Penaeus japonicus</i>	pollack liver oil (PLO) + soybean oil (SO) 1 : 11 : 1)	3, 6, 9, 12 egg albumin	60 % casein and at 6 % PLO : SO (3 : 1 or	best weight increase
Kanazawa <i>et al.</i> (1977a)	<i>P. japonicus</i>	soybean oil, PRO SNCO	8, 12, 16	50 % casein	best growth at 8 % SNCO; growth inhibition at 16 %

PRO : pollack residual oil
SNCO : short necked clam oil.

linoleic acid (18 : 2n6), linolenic acid (18 : 3n3), eicosapentaenoic acid (20 : 5n3) or docosahexaenoic acid (22 : 6n3). These early studies indicated that PUFA and HUFA might be essential for the growth of *P. japonicus*.

In a series of experiments (Kanazawa *et al.* 1977b, 1979b, 1979c), dietary additions of 1% levels of either 18 : 2n6 and 18 : 3n3 to diets containing either oleic acid, pollack (*Theragra chalcogramma*) residual liver oil, soybean oil, or short-necked clam (*Tapes philippinarum*) lipids improved weight gain of *P. japonicus*. Shewbart and Mies (1973) introduced levels of linolenic acid ranging from 0.5% to 5% into a commercially produced shrimp feed and observed the growth response of *P. aztecus* juveniles. A level between 1% and 2% appeared to achieved the best response. Read (1981) found that the addition of 18 : 2n-6 and 18 : 3n-3 fatty acids to diets for *P. indicus* improved growth and survival.

The nutritive value of linolenic acid was found to be greater than that of linoleic acid for the prawn, *P. japonicus* (Kanazawa *et al.* 1977b). Guary *et al.* (1976) also found that better growth of *P. japonicus* was achieved with diets containing high levels of 18 : 3n3 than with those containing comparative levels of 18 : 2n6. Fenucci *et al.* (1981) indicated that the ratio between 1.18 and 1.00 for n3 fatty acids to 1 of 18 : 2n6 should give the best growth response for the growth of *P. stylirostris*. Martin (1980) fed *Palaemon serratus* diets containing different ratios of 18 : 2n-6 and 18 : 3n-3 by varying the relative proportions of soybean oil and linseed oil of the dietary lipid mix. The best growth was achieved with a 18 : 2n-6/18 : 3n-3 ratio of 2.2. Kanazawa *et al.* (1978, 1979d) have observed that n-3 HUFA (20 : 5n-3 and 22 : 6n-3) possess higher activity as essential fatty acids than that of n-6 (18 : 2n-6) and n-3 (18 : 3n-3) PUFA families.

The 20C and 22C HUFA generally demonstrate higher nutritive value than 18C PUFA (Table 2). These results suggest that the often observed growth promoting response to the addition of dietary oils such as shrimp head oil (3% to *Macrobrachium rosenbergii*, Sandifer and Joseph, 1976), sardine or clam oil (4% to *Penaeus japonicus*, Guary *et al.*, 1976) and cod liver oil (4% to *Palaemon serratus*, Martin, 1980) is due to > - 20C n-3 HUFA. Sandifer and Joseph (1976) suggested that the functions of n3 fatty acids was for the biosynthesis of longer chain polyunsaturated fatty acids for tissue incorporation, whereas, the n6 fatty acids were utilized as energy sources. Kanazawa *et al.* (1977a) showed that the low nutritive value of soybean oil for prawns seems to be due to the small amount of HUFA. On the other hand, pollack residual oil containing large amounts of n3 HUFA improved prawn growth rate.

Shrimp appear to have little or no capacity to biosynthesize n-3 PUFA to n-3 HUFA. Kanazawa *et al.* (1979e) found that *Penaeus japonicus* had some ability to convert [1-14C] linolenic acid (18 : 3n-3) to 20 : 5n-3 and 22 : 6n-3. Kanazawa *et al.* (1978) concluded that growth of *P. japonicus* receiving either 18 : 2n6 or 18 : 3n3 dietary fatty acids was inferior to that achieved with 20 : 5n3 and 22 : 6n3. Although this species of prawn can elongate and desaturate 18 : 3n3 or 18 : 2n6 fatty acids, the requirements of the 20 and 22 C HUFA could not be optimally satisfied by dietary provisions of 18C PUFA.

Read (1981) indicated that juvenile *P. indicus* had a limited capacity to chain elongate and desaturate linoleic acid and linolenic acid to C20

and C22 HUFA. He also demonstrated that survival and weight gain associated with a dietary mixture of 18 : 2n6 and 18 : 3n3, 0.5% + 0.5%, respectively, was worse than 18 : 2n6 alone. Superior survival, feed conversion and weight gains were achieved when a combination of dietary 18 : 2n6 and 3% of oils rich in HUFA were provided. Colvin (1976) fed *P. indicus* diets containing sunflower, linseed, soybean or groundnut oil at a 5% level. No significant differences in growth were observed after a 35 days feeding trial. A comparison between the fatty acid profile of the experimental shrimp and that of wild caught individuals suggested a limited capacity for bioconversion of 18C n-6 or n-3 fatty acids to > - 20C n-6 or n-3 fatty acids.

Kanazawa et al. (1979f) investigated the nutritive value of shortnecked clam oil (*Tapes* oil) versus pollack liver oil for the growth of *P. japonicus*. They found that *P. japonicus* fed a diet supplemented with 1% *Tapes* lecithin grew best. The lecithin was the best phospholipid source and contained proportionately greater amounts of 18 : 2n6, 18 : 3n3, 20 : 5n3 and 22 : 6n3.

Bottino et al. (1980) stated that shrimp, *P. setiferus*, brown shrimp, *P. aztecus* and pink shrimp, *P. duorarum* have little ability for the conversion of C18 unsaturated fatty acids into C20 and C22 polyunsaturated fatty acids. Kanazawa et al. (1977a) evaluated pollack residual oil, soybean oil and short necked clam oil as lipid sources for *P. japonicus*. They found that the greatest growth was obtained when prawn received 8% short necked clam oil containing 1.1% of 20 : 5n3 and 1.1% of 22 : 6n3.

A quantitative requirement for any described essential fatty acid has yet to be established for any species of shrimp. Qualitative requirements for linolenic (18 : 3n3), linoleic (18 : 2n-6), eicosapentaenoic acid (20 : 5n-3) and docosahexaenoic (22 : 6n-3) acids have been documented. A specific requirement of the 18 C EFA when satisfactory levels of 20 : 5n-3 or 22 : 6n-3 are available has not been determined. Nevertheless, 20 : 5n-3 and 22 : 6n-3 are generally preferred to 18 : 3n-3. An exogenous source of the HUFA is generally necessary because biosynthetic conversion from 18 : 3n-3 is either absent or limited. Significant increases in weight (gain) growth have been achieved with relatively small additions of PUFA and HUFA. Levels as low as 0.075% of the diet may be sufficient to satisfy the requirements for particular essential fatty acids. Experimentation leading to the precise determination of fatty acid requirements should be directed toward the provision of pure fatty acids in a triglyceride or methyl ester form, rather than through the provision of oils rich in fatty acids of particular interest. The percent composition of other dietary fatty acids should be maintained constant. Qualitative and quantitative analysis of the fatty acids in the experimental diet should be conducted. Tissue analysis of shrimp should always complement feeding studies and should be conducted prior to the initiation of an experiment, after a preconditioning period that precedes the feeding of the experimental diets and at the termination of the experiment. Relating tissue and dietary concentration of suspected EFA should assist in identifying biosynthetic pathways and other fatty acids that may serve as possible indicators of an EFA deficiency.

Dietary polyunsaturated fatty acids also appear to be necessary for successful ovarian maturation (Middleditch et al., 1979, 1980). During induced ovarian maturation, through bilateral eyestalk ablation, the neutral lipids of the ovary of *P. setiferus* contained higher proportions of monoenes and 22 : 6n-3 and lower proportions of 20 : 4n-6 and 20 : 5n-3 than those of non-destalked prawns (Teshima et al., 1988). Nevertheless, no true relationship between dietary PUFA and HUFA and the achievement of ovarian maturity has been established.

Sterols

Most animals are capable of synthesizing sterol from acetate, but shrimp, like all other crustaceans, are incapable of *de novo* sterol synthesis from acetate (Teshima and Kanazawa, 1971). Several estimates of the cholesterol requirement of shrimp and other decapod crustaceans have been reported (Table 3). Kanazawa et al. (1971a) showed that the prawn *P. japonicus* fed with a sterol-free diet had poor growth and survival rates, but grew well on a diet containing 0.5 % cholesterol. Other workers also demonstrated the necessity of dietary cholesterol for good growth of *P. japonicus* (Deshimaru and Kuroki, 1974), juvenile lobster *Homarus americanus* (Castell et al., 1974; D'Abramo et al., 1984) and crayfish (*Pacifastacus leniusculus*) (D'Abramo et al., 1985a).

An optimum dietary level of 0.5 % cholesterol has been found for *P. japonicus* (Kanazawa et al., 1971a) and *H. americanus* juvenile (Castell et al., 1975). Other researchers have obtained the best growth of *P. japonicus* with a diet containing 0.2 % (Shudo et al., 1971) and 2 % (Deshimaru and Kuroki, 1974) cholesterol. D'Abramo et al. (1984) demonstrated that a level of dietary cholesterol as low as 0.12 % was satisfactory for normal growth of lobster and indicated that a cholesterol level ranging from 0.19 to 0.59 % did not improve growth rate. The recent work of Kean et al. (1985) also suggested that the dietary cholesterol requirement for lobster, *H. americanus*, is probably between 0.25-0.5 %. D'Abramo et al. (1985) found that a dietary sterol level of 0.4 % is associated with the best growth of juvenile crayfish, *Pacifastacus leniusculus*. Briggs et al. (1988) added 0.5 % and 1.0 % cholesterol to a semi-purified diet containing 0.12 % cholesterol and did not observe any effects on growth or survival of juvenile prawn, *Macrobrachium rosenbergii*. Teshima and Kanazawa (1983) have demonstrated that the absorption rate of dietary cholesterol is improved by adding lipids such as palmitic acid, tripalmitin or chicken-egg lecithin. The wide range of results concerning sterol requirements may primarily reside in differences in ingredient composition, particularly other lipid components of the test diets. The duration of experiments may also be a principal factor influencing the interpretation of results.

Teshima and Kanazawa (1973) have demonstrated that crustaceans can utilize dietary C28 and C29 sterols, such as *i*-sitosterol, ergosterol and stigmasterol, by bioconversion (dealkylation) of these sterols to cholesterol. The prawn has been shown to be capable of effectively absorbing both dietary cholesterol and phytosterols such as ergosterol and *i*-sitosterol (Teshima et al., 1974). However, *Penaeus japonicus* fed on diets containing either ergosterol, stigmasterol, or *i*-sitosterol have lower growth and

Tab. 3. — Summary of investigations that evaluated sterol nutrition of shrimp and other crustaceans.

Investigations	Species	Sterol (s)	Dietary level (s) (%)	Observations
Castell <i>et al.</i> (1975)	<i>Homarus americanus</i>	cholesterol	0.5, 1, 2	best growth achieved with 0.5 % level
D'Abramo <i>et al.</i> (1984)	<i>H. sp.</i>	cholesterol & phytosterol mix	0.12, 0.19, 0.39, 0.59	0.12 % dietary cholesterol satisfactory for normal growth. At least a portion and possibly all of the sterol requirement is specific for cholesterol.
D'Abramo <i>et al.</i> (1985)	<i>Pascifastacus leniusculus</i>	cholesterol & phytosterol mix	0.22, 0.47	at least 0.4 % total dietary sterol mixsterols required for survival optimum dietary level 0.5 % to 1 % Phytosterol mixture as effective as cholesterol in satisfying a portion of the sterol requirement.
Kanazawa <i>et al.</i> (1971a)	<i>Penaeus japonicus</i>	cholesterol, ergosterol, cholesterol, other sterols	0.05, 0.1, 0.5, 1, 5	best growth achieved with dietary level of 0.5 % cholesterol, cholesterol, other sterols other sterols were inferior to cholesterol
Kean <i>et al.</i>	<i>H. americanus</i>	cholesterol	0, 0.25, 0.5, 1.0	best growth at 0.5 % level
Teshima <i>et al.</i> (1983)	<i>P. japonicus</i>	cholesterol	0, 1, 5	best growth and survival at 1 % level.
Teshima and Kanazawa (1986)	<i>P. japonicus</i>	cholesterol β -sitosterol 24-methyl-22-dienol	0.5	cholesterol at highest r most effective feed conversion and protein.
Teshima <i>et al.</i> <i>P. japonicus</i> (1989)	cholesterol β -sitosterol		0.5, 1.0	No effective cholesterol best growth at 1.0 % level.

survival than those fed on diets containing cholesterol (Kanazawa et al., 1971b). Teshima et al. (1989) found sitosterol to be ineffective in replacing cholesterol at a level of 0.5% in diets for juvenile prawns *P. japonicus*. Partial substitution of cholesterol with sitosterol was associated with reduced weight gain, feed conversion efficiency and protein efficiency ratio. The rate of larval development of this species generally decreased as the amount of sitosterol proportionately increased as a substitute for dietary cholesterol. Neither a mixture (9:1 W/W) of i-sitosterol and cholesterol, nor 24-methylcholesta-5, 22-dienol served as a completely effective substitute cholesterol (Teshima and Kanazawa 1986). Teshima et al., (1983) indicated that when either ergosterol or 24-methylenecholesterol was used as sterol source for larval *P. japonicus*, each had a similar dietary value to that of cholesterol.

D'Abramo et al. (1984) observed poor growth and survival and large amounts of phytosterols in the body tissue of lobster when fed diets containing phytosterols with and without cholesterols. Phytosterols as exclusive dietary sources of sterols yielded poor growth and survival. The sterol requirement is not entirely satisfied by exclusive provision of a mixture of i-sitosterol or desmosterol. Most likely because of inefficient bioconversion. At least a portion or possibly all of the sterol requirement of juvenile lobsters is apparently specific for cholesterol. D'Abramo et al. (1985) found that a mixture of phytosterols could spare the cholesterol requirement and still maintain good growth of crayfish *Pacifastacus leniusculus*. Because of the nature of the diet, the possibility of a total satisfaction of the sterol requirement by dietary phytosterols could not be determined. The ability to effectively use dietary phytosterols as an exclusive or partial substitution for cholesterol appears to be possibly dependent upon substitution diet of the shrimp species. The recent investigations of Teshima and Kanazawa (1986, 1987) suggest that the inferior nutritional value of i-sitosterol for the prawn *Penaeus japonicus* may be attributed to greater turnover rates in the midgut gland and less residence time in the muscle.

Phosphoglycerides

There is evidence indicating that phospholipids are required by some crustaceans. Kanazawa et al. (1979f) found that lecithin (Phosphatidylcholine) and cephalin (phosphatidylethanolamine) fractions derived from the lipid of the short-necked clam, *Tapes philippinarum*, significantly improved the growth of *P. japonicus* when incorporated into artificial test diets at 1%. Teshima et al. (1986) found that weight gain and feed conversion efficiency of *P. japonicus*, increased significantly when 3% soybean lecithin was added to a diet. Similar beneficial effects of dietary phospholipid for *P. monodon* were observed by Pascual (1984). Conklin et al. (1980) found that inclusion of soy lecithin in a purified diet for culture of juvenile lobsters, *H. americanus*, was critical for survival. D'Abramo et al. (1981) later identified the active component of the soy lecithin to be phosphatidylcholine and demonstrated that phosphatidylcholine containing PUFA yielded the best survival. Lack of soy lecithin or the presence of alternative phosphoglyceride sources was associated

with reduced serum cholesterol levels and the rate of transport of this nutrient (D'Abramo *et al.*, 1982, 1985b). The reduction in serum cholesterol levels was later attributed to a decrease of lipoproteins that are believed to serve as vehicles for cholesterol transport and to have phosphatidylcholine as their principal lipid component. Teshima *et al.* (1986) found a lower retention of dietary lipids, especially cholesterol, in body tissue of *P. japonicus* when fed a diet lacking soy lecithin. Prawns receiving a diet with soybean lecithin had a higher concentration of phospholipids in their tissue than those fed a diet with no lecithin supplement. These data indicate that in some instances, phospholipid synthesis may be insufficient based upon observed increases in tissue levels corresponding to dietary increases. Insufficient dietary phospholipid also reduces the effective utilization of dietary lipids such as triglycerides and cholesterol.

Kean *et al.* (1985) demonstrated that juvenile *H. americanus* do not require dietary soy lecithin for survival when purified protein derived from rock crab, *Cancer irroratus*, replaced casein as the principal protein source in a purified diet. They also could not detect any significant growth enhancement when the lobsters were fed crab protein based diets supplemented with either soy lecithin or crab phospholipids at levels of 3.0 and 6.0% of the diet. Supplementation of lecithin to a diet fed to *Macrobrachium rosenbergii* did not increase survival nor promote growth (Hilton *et al.*, 1984; Briggs *et al.*, 1988).

A dietary source of phospholipids is an important factor in growth and survival of larvae of *P. japonicus* (Kanazawa, 1983; Kanazawa *et al.*, 1985). A 3.0% addition of soy lecithin to a larval diet was essential for growth and survival from the nauplius to the postlarval stage and was most effective when other dietary lipid sources such as 18:1n-9 and HUFA, or pollack liver oil were provided. The phospholipid requirement of larvae of *P. japonicus* was estimated to be between 0.5 and 1.0% of the diet. The most effective phospholipids were phosphatidylcholine and phosphatidylinositol, specifically those containing HUFA and PUFA in their ω and α positions.

There is no evidence of the production of bile acids by crustaceans, suggesting that the metabolic processes of emulsification, digestion and transport of lipids in crustaceans are unique. Lipid transport in shrimp is accomplished primarily by high density lipoproteins (HDL) (Teshima and Kanazawa, 1980a). Teshima and Kanazawa (1980b) found that high density and very high density lipoproteins in the serum of the prawn, *P. japonicus* contain substantial amounts (65-85%) of polar lipids. Polyenoic fatty acids, principally docosahexaenoic acid, make up almost 50% of the fatty acids of the lipids of the serum lipoproteins of this shrimp species. Efficient transport of lipids may reside in the provision of dietary phospholipids containing polyenoic fatty acids.

The need for dietary phospholipids appears to be related to the ingredient composition of the remainder of the diet. Lack of exogenous phospholipids has not been associated with mortality of any species of shrimp. Some growth enhancing effects associated with dietary phosphatidylcholine and phosphatidylinositol may be related to an efficient provision of choline and inositol which are at deficient levels in the diet.

Carotenoids

Pigmentation of shrimp is attributed to isoprenoid lipid compounds called carotenoids. Normal pigmentation of crustaceans can only be achieved through an exogenous source of these compounds. Carotenoids have been associated with enhanced growth, reproductive rates and fecundity in crustaceans. Astaxanthin is the carotenoid pigment most often associated with shell pigmentation. The degree of pigmentation appears to be related to the quantity and quality of dietary carotenoids (D'Abramo *et al.* 1983) and to other dietary nutrients. Otazu-Abrill *et al.* (1982) found that dietary methionine and isoleucine were associated with enhanced levels of pigmentation in *Palaemon serratus*. The suggested nutritive role of carotenoids needs to be empirically documented.

CONCLUSION

A thorough knowledge of the lipid requirements of shrimp is lacking. Future investigations need to be directed toward a greater understanding of quantitative requirements. Determinations should not be merely limited to measurements of weight gain and survival but rather should be complemented with well documented, diet dependent, biochemical and histological differences. Results must also be submitted to the appropriate statistical analysis to determine if observed differences are truly significant. Precise quantification can only be achieved through such approaches.

Lipid requirements of shrimp may be dependent upon other lipid or non-lipid constituents of the diet or age and such interactions need to be considered. Larval stages of species of crustaceans most likely require higher levels of dietary lipid relative to juveniles and adults. These forms consume food containing high levels of lipid, particularly if the species is carnivorous. Finally, care should be exercised in planning the duration of an experiment. If insufficient time is allowed for a response to occur, the erroneous conclusions can accordingly result.

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- Bottino N.R., J. Geanny, M.L. Lilly, E. Simmons and G. Finne, 1980.** Seasonal and nutritional effects of the fatty acids of three species of shrimp *Penaeus setiferus*, *P. aztecus* and *P. duorarum*. *Aquaculture*, **19** : 139-148.
- Briggs M.R.P., K. Jaucey and J.H. Brown, 1988.** The cholesterol and lecithin requirements of juvenile prawn (*Macrobrachium rosenbergii*) fed semi-purified diets. *Aquaculture*, **70** : 121-129.
- Castell J.D., 1983.** Fatty acid metabolism in crustaceans. *In* : Proceedings of the Second International Conference on Aquaculture Nutrition : Biochemical and Physiological Approaches to Shellfish Nutrition. Baton Rouge, Louisiana, pp. 124-145.
- Castell J.D., E.G. Mason and J.F. Covey, 1975.** Cholesterol requirements of juvenile American lobster (*Homarus americanus*). *J. Fish. Res. Bd. Can.*, **32** : 1432-1435.

- Castell J.D. and J.F. Covey, 1976. Dietary lipid requirements of adult lobster, *Homarus americanus* (M.E.). *J. Nutr.*, **106** : 1159-1165.
- Castell J.D. and A.D. Boghen, 1979. Fatty acid metabolism in juvenile lobsters (*Homarus americanus*) fed a diet low in methionine and histidine. *Proc. World Maricul. Soc.*, **10** : 720-727.
- Chanmugan P., J. Donovan, C.J. Wheeler and D.H. Hwang, 1983. Differences in the lipid composition of freshwater prawn (*Macrobrachium rosenbergii*) and marine shrimp. *J. Food Sci.*, **48** : 1440-1443.
- Church D.C. and W.G. Pond, 1982. Basic animal nutrition and feeding. John Wiley and Sons, New York, New York, U.S.A..
- Colvin P.M., 1976. The effect of selected seed oils on the fatty acid composition and growth of *Penaeus indicus*. *Aquaculture*, **8** : 81-89.
- Conklin D.E., L.R. D'Abramo, C.E. Bordner and N.A. Baum, 1980. A successful purified diet for the culture of juvenile lobster: the effect of lecithin. *Aquaculture*, **21** : 243-250.
- D'Abramo L.R., C.E. Bordner, D.E. Conklin and N.A. Baum, 1981. Essentiality of dietary phosphatidylcholine for the survival of juvenile lobsters, *Homarus*. *J. Nutr.*, **111** : 425-431.
- D'Abramo L.R., C.E. Bordner and D.E. Conklin, 1982. Relationship between dietary phosphatidylcholine and serum cholesterol in the lobster *Homarus* sp. *Mar. Biol.*, **67** : 231-235.
- D'Abramo L.R., N.A. Baum, C.E. Bordner and D.E. Conklin, 1983. Carotenoids as a source of pigmentation in juvenile lobsters fed a purified diet. *Canadian Journal of Fisheries and Aquatic Sciences*, **40** : 699-704.
- D'Abramo L.R., C.E. Bordner, D.E. Conklin and N.A. Baum, 1984. Sterol requirement of juvenile lobsters *Homarus* sp., *Aquaculture*, **42** : 13-25.
- D'Abramo L.R., J.S. Wright, K.H. Wright C.E. Bordner and De Carlin. 1985. Sterol requirement of cultured juvenile crayfish *Pacifastacus leniusculus*. *Aquaculture*, **49** : 245-255.
- D'Abramo L.R., N.A. Baum, C.E. Bordner, D.E. Conklin and E.S. Chang, 1985. Diet dependent sterol transport in the American lobster. *J. Exp. Mar. Biol. Ecol.*, **87** : 83-96.
- Davis D.A. and E.H. Robinson, 1986. Estimation of the dietary lipid requirement level of the white crayfish *Procambarus acutus*. *J. of the World Aquacul. Soc.*, **17** : 37-43.
- Deshimaru O. and K. Kuroki, 1974. Studies on a purified diet for prawn-II. Optimum contents of cholesterol and glucosamine in the diet. *Bull. of the Jpn. Soc. of Sci. Fish.*, **40** : 421-424.
- Deshimaru O. K. Kuroki and Y. Yone 1979. The composition and level of dietary lipid appropriate for the growth of prawn. *Bull. of the Jpn. Soc. of Sci. Fish.*, **45** : 591-594.
- Fenucci J.L., A.L. Lawrence and Z.P. Zein-Eldin, 1981. The effects of fatty acid and shrimp meal composition of prepared diets on growth of juvenile shrimp, *Penaeus stylirostris*. *J. World Maricul. Soc.*, **12** : 315-324.
- Forester J.R.M. and T.W. Beard, 1973. Growth experiments with the prawn *Palaemon serratus* (Pennant) fed with fresh and compounded diets. *Fish. Invest. Ser.*, **II 27** : 16 pp.
- Guary J.C., M. Kayama, Y. Murakami and H.J. Ceccaldi, 1976. The effects of a fat-free diet and compounded diets supplemented with various oils on molt, growth and fatty acid composition of prawn, *Penaeus japonicus* Bate. *Aquaculture*, **7** : 245-254.

- Hilton J.W., K.E. Harrison and S.J. Slinger, 1984. A semi-purified test diet for *Macrobrachium rosenbergii* and the lack of need for supplemental lecithin. *Aquaculture*, **37** : 209.
- Kanazawa A., 1983. Penaeid nutrition. In : proceedings of the Second International Conference on Aquaculture Nutrition : Biochemical and Physiological Approaches to Shellfish Nutrition. G.D. Pruder, D.E. Conklin and C. Langdon (Eds.), Louisiana State University, Division of Continuing Education, Baton Rouge, LA.
- Kanazawa A., N. Tanaka and S. Teshima, 1971a. Nutritional requirements of prawn-II. Requirement for sterols. *Bull. of the Jpn. Soc. Sci. Fish.*, **37** : 211-215.
- Kanazawa A., N. Tanaka, S. Teshima and K. Kasiwada, 1971b. Nutritional requirements of prawn-III. Utilization of the dietary sterols. *Bull. Jpn. Soc. Sci. Fish.*, **37** : 1015-1019.
- Kanazawa A., S. Teshima and S. Tokiwa, 1977a. Nutritional requirements of prawn-VII. Effect of dietary lipids on growth. *Bull. of the Jpn Soc. Fish.*, **43** : 894-856.
- Kanazawa A., S. Tokiwa, M. Kayama and M. Hirata, 1977b. Essential fatty acids in the diet of prawn-I. Effects of linoleic and linolenic acids on growth. *Bull. of the Jpn. Soc. Sci. Fish.*, **43** : 1111-1114.
- Kanazawa A., S. Teshima, M. Endo and M. Kayama, 1978. Effects of eicosapentaenoic acid on growth and fatty acid composition of the prawn, *Penaeus japonicus*. *Mem. Fac. Fish. Kagoshima Univ.*, **27** : 35-40.
- Kanazawa A., S. Teshima and S. Tokiwa, 1979a. Biosynthesis of fatty acids from palmitic acid in the prawn, *Penaeus japonicus*. *Mem. of the Fac. of Fish. Kagoshima Univ.*, **28** : 17-20.
- Kanazawa A., S. Teshima and M. Endo., 1979a. Requirements of prawn, *Penaeus japonicus* for essential fatty acids. *Mem. Fac. of Fish. Kagoshima Univ.*, **28** : 27-33.
- Kanazawa A., S. Teshima, S. Tokiwa, H.J. Ceccaldi, 1979c. Effects of dietary linoleic and linolenic acids on growth of prawn. *Oceanological Acta.*, **2** : 41-47.
- Kanazawa A., S. Teshima, S. Tokiwa, M. Kayama and M. Hirata, 1979d. Essential fatty acids in the diet of prawn-II. Effect of docosahexaenoic acid on growth. *Bull. of the Jpn. Soc. Sci. Fish.*, **45** : 1151-1153.
- Kanazawa A., S. Teshima and S. Ono, 1979e. Relationship between essential fatty acid requirements of aquatic animals and the capacity for bioconversion of linolenic acid to highly unsaturated fatty acids. *Comp. Biochem. Physiol.*, **63** : 295-298.
- Kanazawa A., S. Teshima, S. Tokiwa, M. Endo and F.A. Abdel Razek, 1979e. Effects of short-necked clam phospholipids on the growth of prawn. *Bull. of the Jpn. Soc. Sci. Fish.*, **45** : 961-965.
- Kanazawa A, S. Teshima and M. Sakamoto, 1985. Effects of dietary lipids, fatty acids and phospholipids on growth and survival of prawn (*Penaeus japonicus*) larvae. *Aquaculture*, **50** : 39-49.
- Kayama M., M. Hirata, A. Kanazawa, S. Tokiwa and M. Saito, 1980. Essential fatty acids in the diet of prawn-III. Lipid metabolism and fatty acid composition. *Bull. Jpn. Soc. Sci. Fish.*, **46** : 483-488.
- Kean J.C., J.D. Castell, A.G. Boghen, L.R. D'Abramo and D.E. Conklin, 1985. A re-evaluation of the lecithin and cholesterol requirements of juvenile lobster (*Homarus americanus*) using crab protein-based diets. *Aquaculture*, **47** : 143-149.

- Martin B.J., 1980.** Croissance et acides gras de la crevette *Palaemon serratus* (Crustacea, Decapoda) nourrie avec des aliments composés contenant différentes proportions d'acide linoléique et linoléique. *Aquaculture*, **19** : 325-337.
- Middleditch B.S., S.R. Missler, H.B. Hines, J.P. McVey and A. Brown, 1980.** Metabolic profiles of penaeid shrimp : dietary lipids and ovarian maturation. *J. Chromatography*, **195** : 359-368.
- Middleditch B.S., S.R. Missler, D.G. Ward, J.B. McVey, A. Brown, and A.L. Lawrence, 1979.** Maturation of penaeid shrimp : dietary fatty acids. *Proc. World Maricul. Soc.*, **10** : 472-476.
- Otazu-Abrill M., B.J. Martin and H.J. Ceccaldi, 1982.** Influence des acides aminés purifiés ajoutés dans les aliments composés, sur le métabolisme des pigments caroténoïdes, chez *Palaemon serratus* (Crustacea, Decapoda), *Aquaculture*, **28** : 303-310.
- Pascual F.P., 1984.** Lecithin requirement of *Penaeus monodon* juveniles. Abstract, First International Conference on the culture of Penaeid Prawns/shrimps, Iloilo, Phillipines.
- Read G.H.L., 1981.** The response of *Penaeus indicus* (Crustacean : Penaeid) to purified and compounded field of varying fatty acid composition. *Aquaculture*, **24** : 245-256.
- Sandifer P.A. and J.D. Joseph, 1976.** Growth responses and fatty acid composition of juvenile prawns (*Macrobrachium rosenbergii*) fed a prepared ration augmented with shrimp head oil. *Aquaculture*, **8** : 128-138.
- Shewbart K.L. and W.L. Mies, 1973.** Studies on nutritional requirements of brown shrimp- the effect of linolenic acid on growth of *Penaeus aztecus*. Proc. World Mari. Soc. 4th Annual Workshop 277-287.
- Shudo K., K. Nakamura, S. Ishikawa and K. Kitabayshi, 1971.** Studies on the growth-promoting effects of both squid liver oil and cholesterol. *Bull. of Tokai Regional Fisheries Research Laboratories*, **65** : 129-137.
- Teshima S. and A. Kanazawa, 1971.** Biosynthesis of sterols in the lobster, *Panulirus japonica*, the prawn, *Penaeus japonicus* and the crab, *Portunus trituberculatus*. *Comp. Biochem. Physiol.*, **38B** : 597-602.
- Teshima S. and A. Kanazawa, 1973.** Biosynthesis of sterols from demosterol in a mussel, *Mytilus edulis*. *Bull. of the Jpn. Soc. Sci. Fish.*, **39** : 1309-1314.
- Teshima S. and A. Kanazawa, 1980a.** Lipid transport in crustaceans. *Min. Rev. Data File Fish Res.*, **1** : 1-25.
- Teshima S. and A. Kanazawa, 1980b.** Lipid constituents of serum lipoproteins in the prawn. *Bull. Jpn. Soc. Sci. Fish.*, **46** : 57-62.
- Teshima S. and A. Kanazawa, 1983.** Digestibility of dietary lipids in the prawn. *Bull. of the Jpn. Soc. Sci. Fish.*, **49** : 963-966.
- Teshima S. and A. Kanazawa, 1986.** Nutritive value of sterols for the juvenile prawn. *Bull. Jpn. Soc. Sci. Fish.*, **52** : 1417-1422.
- Teshima S. and A. Kanazawa, 1987.** Turnover of dietary cholesterol and i-sitosterol in the prawn. *Nippon Suisan Gakkaishi*, **53** : 601-607.
- Teshima S., A. Kanazawa and H. Okamoto, 1974.** Absorption of sterols and cholesterol esters in a prawn, *Penaeus japonicus*. *Bull. of the Jpn. Soc. Sci.*, **40** : 1015-1019.
- Teshima S., A. Kanazawa and H. Sasada, 1983.** Nutritional value of dietary cholesterol and other sterols to larval prawn, *Penaeus japonicus* Bate. *Aquaculture*, **31** : 159-167.

- Teshima S., A. Kanazawa and Y. Kakuta, 1986.** Effects of dietary phospholipids on growth and body composition of the juvenile prawn. *Bull. Jpn. Soc. Sci. Fish.*, **52** : 155-158.
- Teshima S., A. Kanazawa, S. Koshio and K. Horinouchi, 1988.** Lipid metabolism in destalked prawn *Penaeus japonicus* : induced maturation and accumulation of lipids in the ovaries. *Nippon Suisan Gakkaishi*, **54** : 1115-1122.
- Teshima S., A. Kanazawa, S. Koshio and N. Kondo, 1989.** Nutritive value of sitosterol for the prawn *Penaeus japonicus*. *Nippon Suisan Gakkaishi*, **55** : 152-157.