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INITIAL PALATABILITY AND GROWTH TRIALS ON PELLETED DIETS FOR CEPHALOPODS

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ABSTRACT

The palatability of frozen shrimp, live shrimp, live fish, fish fillets, surimi, raw chicken meat, pureed shrimp and chicken, turkey hot dogs and pellets formulated with penaeid shrimp, mysid shrimp and chicken, was tested on octopuses and cuttlefishes. The results of the octopus and cuttlefish palatability trials demonstrated that cuttlefishes took longer to grab all foods except live shrimp and fish than did the octopuses and neither ingested the surimi diets. The feeding responses of octopuses for pellets appeared to be most closely related to the moisture content of the pellets while cuttlefishes seemed to be less influenced by texture. Octopuses fed upon the penaeid and chicken pellets at the same rates (2.7–5.2% body weight·day^{-1}, % bw·d^{-1}) as live shrimp (4.9% bw·d^{-1}) and raw chicken (3.5% bw·d^{-1}), but only the latter diets promoted growth. Adult *S. officinalis* (124–465 g) survived and grew on penaeid pellets over 60 days, although there was substantial mortality by day 40 in group-reared cuttlefishes due to cannibalism. Feeding rates increased steadily from 1.0 to 3.5% bw·d^{-1}, although growth rates were about one-third to one-half that of cuttlefishes fed live shrimp. Juvenile cuttlefishes (14 g) ingested pellets routinely after 4 days and the feeding rates ranged 6.4–8.1% bw·d^{-1}. The cuttlefishes doubled their weight in 57 days although 7 of 20 were cannibalized. The moist, pelleted diet appeared to be palatable to both octopuses and cuttlefishes but the reduced growth rates for cuttlefishes and no growth exhibited by the octopuses fed the pellets suggest that nutritional deficiencies or imbalances exist. The results demonstrate that pelleted diets can be used to maintain cephalopods and modest growth can be expected at this stage of diet development. The disadvantages of lower growth rates are offset by the cost savings compared to expensive natural diets that require high labor costs for collection and maintenance.

The mariculture potential of cephalopod molluscs has been recognized for many years (reviewed by Hanlon, 1987) because of their rapid growth rates, short life cycles and high food conversion efficiencies (Forsythe and Van Heukelem, 1987). However, the inability to grow any cephalopod species on a storable, inexpensive diet has inhibited commercial cephalopod mariculture (O'Dor and Wells, 1987). Recently, the range of natural foods on which cephalopods can be maintained or grown in the laboratory has been expanded (Boletzky and Hanlon, 1983; Toll and Strain, 1988; DeRusha et al., 1989).

This research project has four interrelated objectives: (1) analyses of the influence of behavioral conditioning on feeding behavior; (2) identification of chemical attractants and feeding stimulants (Lee, in press); (3) evaluation of diet palatability (reported herein) and digestibility of the component nutrients (studies in progress) and (4) growth trials to evaluate the nutritional quality of diets (reported herein). The effect of palatability on food ingestion and growth has been documented previously for fish, crustaceans and molluscs (Ghittino, 1979; Carefoot, 1982; Jobling, 1986; Mearns et al., 1987). The form of the diet (shape, size and moisture content) has been related directly to ingestion and growth in fishes (Wankowski and Thorpe, 1979; Stradmeyer et al., 1988). The integration of the chemotactile senses of octopuses is well described in the literature (Wells, 1978); hence the importance of these senses must be assessed carefully in the design of an artificial diet. Our experimental hypothesis was that differences in feeding response to a pelleted diet were related not only to the total nutrient composition, but equally to the textural and chemical (taste) attributes.

Table 1. Feeding responses of five *Octopus bimaculoides* to various foods

Food	Mean latency-to-grab (s)*	Ingestion
Live shrimp (LS)	75 ± 159	yes
Raw chicken (RC)	9 ± 13	yes
Chicken puree (CPE)	152 ± 258	yes
Shrimp puree (SPE)	20 ± 0	yes
Penaeid pellet (PP)	49 ± 79	maybe (broken up)
Chicken pellet (CP)	74 ± 59	maybe (broken up)
Artificial surimi (SA)	5 ± 4	no (broken up)
Mysid pellet (MP)	27 ± 34	no (broken up)
Natural surimi (SN)	19 ± 20	no
Turkey hot dog (TH)	32 ± 46	no

* Mean and standard deviation.

Palatable diets can have the correct chemical and physical characteristics for ingestion, yet they may still not support normal growth. The ultimate test of any diet is the biological performance (survival, growth and reproduction) of the animal to which it is fed. Cephalopods exhibit high rates of growth (Forsythe and Van Heukelem, 1987) on natural live diets. This paper describes the first results obtained in growing cephalopod molluscs solely on a pelleted diet for up to 2 months, which represent approximately a fifth of their laboratory life cycle.

Materials

Animals.—*Octopus bimaculoides* were caught by trap near Long Beach, California by a commercial collector, shipped to Galveston and acclimated to laboratory conditions for 1–2 weeks before being used in trials. The European cuttlefishes *Sepia officinalis* were all laboratory cultured from hatching. One population (P) was cultured from eggs collected on the Atlantic coast of France, brought to Galveston and hatched in the laboratory. The second group of cuttlefishes was the F_4 generation from an initial brood of eggs collected in England, sent to the Monterey Bay Aquarium (CA) and brought to Galveston (June 1985). Initial and final weights were determined by netting the animal, draining the water retained in the mantle cavity and placing them in a wet-tared weighing tray or beaker; the entire procedure required less than 30 sec.

Culture Systems and Methods.—All culture tanks were closed, recirculating seawater systems ranging in size from 1,500–9,500 liters and were similar in design to those described in detail by Hanlon and Forsythe (1985). These systems used a combination of artificial seawater and highly filtered bay water maintained at 35‰ and pH 7.8–8.1. The culture methodology for cuttlefishes was described in detail by Forsythe et al. (1991).

Diets.—The control diet in all palatability and growth experiments was live and/or thawed, frozen penaeid shrimp (live—LS, frozen—FS) and various common fishes (live—LF, frozen—FF) that had been captured from Galveston Bay, TX. Pellets were formulated using marine animal and terrestrial plant meals, vitamin and mineral premixes and binders (sodium alginate and gelatin). The mysid shrimp-based pellet (mysid pellet, MP) was formulated with mysid shrimp meal, fish protein hydrolysate, fish meal, casein, egg white, soybean meal, torula yeast and raw squid. The moist penaeid shrimp-based pellet (penaeid pellet, PP) contained raw penaeid shrimp, casein, soybean meal, mysid meal and raw squid. The moist raw chicken-based pellet (chicken pellet, CP) contained raw chicken, egg white, casein, soybean meal, torula yeast, feather meal and raw squid. The pellets were extruded through a meat grinder with two sizes of dies, 10 mm and 3 mm in diameter. The larger die was used to prepare all pellets for octopuses (approximately 3–4 cm in length). The cuttlefishes in Palatability Trial 1 and Growth Trial 1 were fed the same pellets as the octopuses while Growth Trial 2 used the smaller diameter pellets (2 cm in length) for the first part of the experiment and later the larger diameter pellets (3–4 cm in length) were fed. The mysid pellet (MP) was dried in an oven until the pellets contained approximately 10% moisture. The estimated proximate composition of the mysid (MP), penaeid (PP) and chicken (CP) pellets were 63, 33, and 54% protein; 7, 3, and 4% lipid; 5, 7, and 6% carbohydrate; 17.8, 3.5 and 7.7% ash; and 9, 43 and 27% moisture, respectively. All the pellets and purees were kept frozen or refrigerated until just before feeding. After weighing, the pellets were allowed to hydrate for 2 min in seawater so that they would sink quickly.

Various diets were used for the palatability studies (Table 1). Only the penaeid shrimp-based diet and the control diet were fed in the cuttlefish growth trials. The raw fresh and frozen diets (shrimp, FS and chicken, RC) provided foods that were natural (i.e., texture and taste) but non-motile. Purees of shrimp abdomen (SPE) and chicken breast (CPE) were made by blending until smooth the raw, cleaned meat in a commercial blender. Thus, the purees contained the same nutrients as the raw diets but the texture of flesh was destroyed. Both surimi products were purchased at markets and had been prepared from pollack and enhanced with either natural crab flavoring (SN) or artificial crab flavoring (SA). The surimi should have simulated the flesh of crabs, one of *Octopus*' preferred prey.

Methods and Results

Octopus bimaculoides

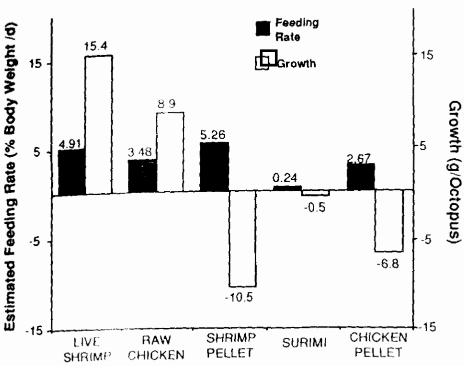
Palatability Trial 1, Methods.—Five *O. bimaculoides* (125–200 g) were isolated individually in 7.5 × 7.5 × 7.5 cm opaque chambers with clear bottoms. The chambers were situated above a VHS video camera attached to a remote-controlled servomotor. Each octopus was presented with each type of food (Table 1) over a period of 6 days (maximum of two trials$\cdot d^{-1}$ and two foods$\cdot d^{-1}$). The octopuses were given 15 min to react to the food and then it was removed. For each trial, the amount of food offered was calculated to be 2% of body weight/feeding (2.5–2.7 g) and each octopus was tested once or twice per day. Initial feeding responses to the various foods were graded by the timed Latency-to-Grab (the interval between introduction of a food and when the octopus first grabbed it with its arms) and responses were evaluated by Analysis of Variance (ANOVA; Sokal and Rohlf, 1969). Finally it was noted whether the octopus eventually ingested some or all the food.

Results.—The Mean Latency-to-Grab and degree of ingestion are listed in Table 1. No statistical differences ($P > 0.05$) were detected in Mean Latency-to-Grab because of the high variance among the five octopuses. Octopuses frequently moved the food immediately to the buccal region where it disappeared or they manipulated the food for varying periods, eventually bringing it to the mouth or dropping it. Live shrimp (LS), raw chicken (RC) and both purees (SPE and CPE) were usually drawn directly to the mouth and ingested. The pellets (PP, CP and MP) were grabbed and drawn to the mouth but frequently pieces would be lost due to an excurrent blast of water from the siphon. These pieces might be grabbed again later or ignored. The surimi (SA and SN) was grabbed, moved to the mouth and discarded usually after a single bite; octopuses would sometimes touch the pieces again without ingesting. Turkey hot dogs elicited the weakest responses; they were never brought to the mouth.

Growth Trial 1, Methods.—Twenty five octopuses (124.5 ± 37.6 g) were divided into five equal groups. They were weighed, starved for 24 h, and fed one of five diets for 10 days: live shrimp (LS), raw chicken (RC), surimi (SN), penaeid pellet (PP) or chicken pellet (CP). All octopuses received a ration of 5.4% body weight per day (% bw$\cdot d^{-1}$). Octopuses were fed ½ of their ration at approximately 0830 and 1630 and 3 h later the uneaten food was removed and weighed. Food conversion ratios (FCR) were calculated as follows:

$$(\text{Food in} - \text{Food out}) \div \text{weight gain} = \text{FCR}$$

where "Food in" is the total amount of food fed to five octopuses over 10 days. "Food out" represents the total amount of food not eaten, retrieved and weighed over the 10 days. Food out was multiplied by an appropriate correction factor



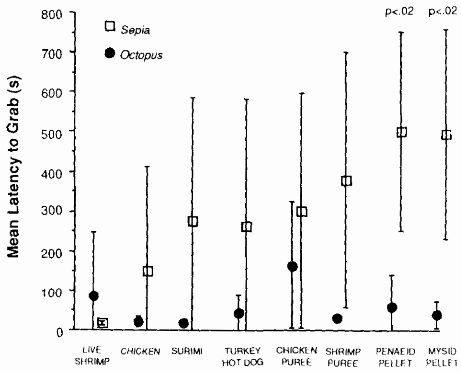


Figure 1 (left). Growth and estimated daily feeding rate of *Octopus bimaculoides* receiving five different diets. The estimated daily feeding rate is given as percent of body weight eaten per day (% bw·d^{-1}).

Figure 2 (right). Comparison between the Mean Latency-to-Grab for *Octopus bimaculoides* and *Sepia officinalis*. Symbols represent the mean of five animals and the error bars are the standard deviation. Significant differences are indicated by probability value.

based upon the amount of water adsorbed by each food in 3 h. An ANOVA was used to evaluate differences in the ingestion of food based upon diet type (five diets) and time of day (AM vs. PM).

Results.—Figure 1 displays the estimated feeding rates and growth for each diet. Growth was achieved on the live shrimp (LS) and raw chicken (RC) diets, resulting in the only positive FCR's (1.00 and 1.22, respectively). The pelleted diets and surimi produced no growth. However, there were no statistical differences when comparing weight gain by food group (Student's t-test, $P > 0.05$) except between the final weights of the live shrimp (LS) group versus the penaeid pellet (PP) group ($P < 0.029$). In terms of the feeding rate, all octopuses ingested more at the morning feeding ($P < 0.0003$). The effects of diet were significant ($P < 0.0001$): a Duncan's Mean Separation test indicated that the estimated feeding rates for the penaeid pellet (PP) and live shrimp (LS) were similar and greater than those for the other diets. The feeding rate for surimi (SA) was the lowest. These results agree with the qualitative results of the palatability trial since all the foods were grabbed quickly but the live shrimp (LS) was always ingested, the penaeid pellet (PP) was partially ingested and the surimi (SA) was never ingested.

Sepia officinalis

Palatability Trial 1, Methods.—Five *Sepia officinalis* (20–35 g) were isolated in the same chambers used in the octopus palatability trial and the same methods were used. Additional data included whether or not the cuttlefish ingested half or more of a given food (Table 2). Each cuttlefish was offered each food four times (N = 20 for each food tested). Differences in the frequency of ingestion were examined (Chi Square), along with comparisons of the Mean Latency-to-Grab between *O. bimaculoides* and *S. officinalis* (ANOVA and Paired t-test).

Results.—The mean Latency-to-Grab (N = 5; the first presentation to each cuttlefish) and the number of ingestions where at least half the food was ingested are listed in Table 2. The live (LF) and frozen fish (FS) were grabbed and ingested at the highest rate while the surimi (SA) and mysid pellets (MP) were never eaten.

Table 2. Feeding responses of *Sepia officinalis* to various foods (N = 20 each)

Food	Mean latency-to-grab (s)*	Ingestion >50% (N/20)
Live fish (LF)	5 ± 2	20
Live shrimp (LS)	6 ± 4	17
Frozen shrimp (FS)	135 ± 260	17
Fish fillets (FF)	138 ± 258	17
Shrimp puree (SPE)	365 ± 32	19
Turkey hot dogs (TH)	250 ± 319	6
Chicken puree (DPE)	288 ± 296	4
Penaeid pellet (PP)	487 ± 251	3
Raw chicken (RC)	137 ± 260	3
Artificial surimi (SA)	263 ± 308	0
Mysid pellet (MP)	482 ± 264	0

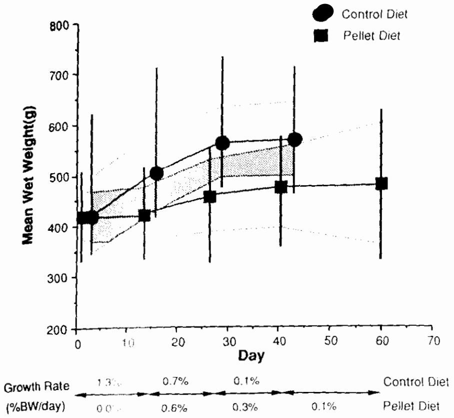
* Mean and standard deviation.

A Chi-Square test on ingestion frequency confirmed the differences ($P < 0.01$). Individual differences in response were also noted since the frequency of grabbing varied from 35% to 76% for the five cuttlefishes during the trials, a statistically significant difference ($P < 0.05$). An ANOVA comparing Mean Latency-to-Grab between *O. bimaculoides* and *S. officinalis* revealed a significant difference between the timed responses (Mean Latency-to-Grab) related to species but no differences in the responses for individual foods. Paired *t*-tests (Fig. 2) comparing the responses for similar foods between the two species revealed that octopuses grabbed the penaeid and mysid pellets in significantly less time ($P < 0.05$) than the cuttlefishes.

Growth Trial 1, Methods.—A population (N = 56) of six-month-old (P generation) cuttlefishes was divided into four groups. The 20 largest cuttlefishes (420 ± 75 g) were isolated into two individual culture systems (chambers 30 × 120 × 15 cm), with 10 receiving the penaeid pellet (PP) and 10 the control diet (LS). Two groups of 18 cuttlefishes each were established in two different culture systems. The first group (243 ± 73 g) was held in a 1.8 m diameter tank with 40 cm water depth in a 1,500 liter culture system and were fed the penaeid pellet (PP). The second group (309 ± 82) was held in a 4.0 × 2.0 × 0.4 m tank on a 5,000 liter culture system and was fed the control diet (LS).

Initial ration levels were set at 5.0% $bw \cdot d^{-1}$ and adjusted based on ingestion, allowing a slight excess of food. The daily ration was presented in two equal feedings at 0900 and 1600. Uneaten food remains from the group pellet population were collected and weighed periodically to estimate ingestion. Visual estimates of food ingestion (0, 25, 50, 75 or 100% ingested) were recorded daily for the 10 isolated cuttlefishes fed the pellet to approximate feeding rate. Estimated Daily Feeding Rate (% $bw \cdot d^{-1}$) was calculated for each cuttlefish. Daily weight of each cuttlefish was estimated from a linear regression connecting the individual cuttlefish's initial and final weights. The estimated amount of food ingested on a given day was then divided by the cuttlefish's estimated weight and multiplied by 100 for the Estimated Daily Feeding Rate.

Results.—The trial lasted 60 days but after day 41 the surviving isolated cuttlefishes were combined into groups to allow mating and spawning. At day 60, the cuttlefishes fed the penaeid pellet (PP) were weighed because the control diet (LS, LF and FS) population was in the midst of mating and egg-laying. Among the isolated cuttlefishes, there was no statistically significant difference in survival between the pellet-fed (PP) and control cuttlefishes through day 41. Among the



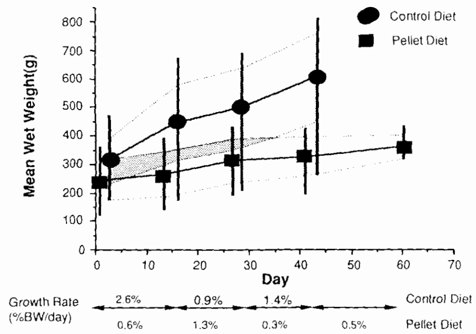


Figure 3 (left). Growth of isolated *Sepia officinalis* fed the penaeid pellet versus the control diet. The data are offset equally at each measurement day for clarity. The plot symbol represents the mean weight, the black lines denote the range and the shaded areas encompass the standard deviation above and below the mean. Mean growth rates from measurement to measurement are included below the graph for comparison.

Figure 4 (right). Growth of group-reared *Sepia officinalis* fed the penaeid pellet versus the control diet. The data are offset equally at each measurement day for clarity. The plot symbol represents the mean weight, the black lines denote the range of measurements and the shaded areas encompass the standard deviation above and below the mean. Mean growth rates from measurement to measurement are included below the graph for comparison.

group-reared cuttlefishes, survival was lower on the pelleted diet (PP) versus control diet (day 41, 50% vs. 94%; day 60, 28% vs. 94%). Most of the mortalities in the pellet-fed (PP) group had been cannibalized but no attacks were ever observed so the cuttlefishes could have been cannibalized before or after death. During the first 40 days of the trial, seven of the 10 isolated cuttlefishes fed pellets gained weight (average weight gain, 20% bw and mean instantaneous growth rate, MIGR, was 0.44% bw·d^{-1}), one lost weight (11% bw) and two died after 24 days (Fig. 3). However, one mortality was due to loss of water flow to the chamber. The maximal individual growth rate observed was 1.3% bw·d^{-1} (two cuttlefishes over different 2-week periods). Four cuttlefishes survived to day 60. By comparison 8 of the 10 isolated cuttlefishes fed the control diet (LS, LF and FS) grew an average of 42% bw during the first 41 days at 0.86% bw·d^{-1} MIGR, one cuttlefish lost weight (13% bw·d^{-1}) and one died. The maximal individual growth rate was 6.0% bw·d^{-1} MIGR during the first 2-week interval. The mean weight on day 41 (Fig. 3) and the MIGR for day 1 to 41 of the pellet bed and control groups were significantly different ($P < 0.04$, respectively). All isolated cuttlefishes showed a trend of decreasing MIGR. Feeding rates of isolated cuttlefishes fed the penaeid pellet (PP) gradually increased from near 1% bw·d^{-1} to nearly 3% bw·d^{-1}. The highest individual feeding rates were in the 4–5% bw·d^{-1} range. There was no significant difference ($P > 0.05$) in feeding rates between the AM and PM feedings.

Growth of group-reared cuttlefishes fed the penaeid pellets, PP, (Fig. 4) was higher than for the isolated cuttlefishes. They grew an average of 34% of bw and an average instantaneous growth rate (AIGR) if 0.74% bw·d^{-1}. The highest AIGR occurred in the second 2-week period (1.3% bw·d^{-1}). Five cuttlefishes survived to day 60 and showed a significant increase ($P < 0.025$) in mean weights compared to the mean weights on day 41. Seven cuttlefishes were cannibalized partially during the trial but the influence of this food source on growth should have been minor. The cuttlefishes that fed immediately and vigorously on pellets when pellets were presented the first few times used tentacular strikes to grasp the falling pellets. During group culture, these tentacular strikes were common initially but grasping pellets using the arms became progressively more common.

The group-reared cuttlefishes that were fed the control diet (LS, LF and FS) grew an average of 98% bw at an AIGR of 1.63% bw·d^{-1}. The highest AIGR was 2.6% bw·d^{-1} during the first 14 days. The lone mortality in this group was a cannibalism on day 38. The group-reared cuttlefishes fed the control diet ingested an estimated daily ration of 4.0% bw·d^{-1}. By contrast, the group-reared cuttlefishes fed the penaeid pellets (PP) ingested an average of 1.8% bw·d^{-1}, starting at 1% bw·d^{-1} the first 2 weeks and increasing to 3.5% bw·d^{-1} the last 2 weeks.

Growth Trial 2, Methods.—A group of 20 F_4 generation cuttlefishes (13.9 ± 2.6 g) was weighed, placed in a 1.8 m diameter tank and fed dead shrimp (*Palaemonetes* spp.) for 3 days then penaeid pellets (PP) for 57 days. A ration level of 7–8% bw·d^{-1} was divided among 3–5 feedings·d^{-1} in an effort to increase daily feeding rate. Uneaten food remains were collected after each feeding, placed in a drying oven (45°C) for 24 h and weighed. All cuttlefishes were weighed on days 1, 26 and 57 and linear regressions between these mean population weights were used to estimate daily mean animal weight for calculations of estimated daily feeding rates. Since there were six cannibalisms between days 26 and 57 and the smallest cuttlefishes are the most likely victims, the regression for that period was calculated using the 13 largest cuttlefishes on day 26.

Results.—The small cuttlefishes required 3 days of conditioning with dead shrimp before they grabbed non-living food readily. Pellets (PP) were taken on the fourth day and eaten actively. There was one cannibalism during the conditioning phase but the remaining 19 cuttlefishes survived to day 35 after which cannibalisms occurred every 3–5 days, resulting in the survival of 13 cuttlefishes on day 57. Over the first 26 days, the cuttlefishes ingested an estimated average of 6.4% bw·d^{-1} (or 82% of the food offered) and grew from 13.9–16.2 g for an AIGR of 0.61% bw·d^{-1}. The 13 largest cuttlefishes on day 26 weighed an average of 17.7 g compared to 28.0 g for the 13 survivors on day 57 (an AIGR of 1.5% bw·d^{-1}). Feeding rates increased over the second growth period to 8.1% bw·d^{-1} (93% of food offered).

Discussion

Cephalopods will grab and ingest non-living, non-motile foods but behaviorally there is considerable intra- and interspecific variability in the intensity of feeding response to pelleted diets.

Octopuses.—Wild-caught adult *O. bimaculoides* grabbed any potential food offered including pellets (Table 1). The octopuses ingested shrimp and chicken, whether live (LS), raw (FS and RC) or pureed (SPE and CPE). However, they were less likely to ingest pelleted diets (PP, CP and MP). Texture may be a major factor affecting ingestion of pellets since the dry mysid pellet (MP) was never ingested while the penaeid (PP) and chicken pellets (CP) were ingested partially (Table 1). Texture did not appear to be as important for the raw diets since the purees (SPE and CPE) were ingested by the octopuses as readily as the live shrimp (LS) and raw chicken (RC) even though the Mean Latency-to-Grab was somewhat lower. The moisture content of the purees was similar to that of the live or raw diets, whereas moisture differed significantly for the dry (10% moisture content) versus moist pellets (40% moisture content). As a result, moisture content may be the most important textural quality affecting ingestion and the moist pellets were chosen for use in the growth trials. A surprising result was the rejection of both the surimis, SN and SA. The octopuses' quick responses to grab the surimi were expected since surimi should approximate the same texture and taste of crab meat and crabs are a preferred prey for octopuses. Surimi may contain chemical attractants that encourage grabbing but these same chemicals or other chemicals may serve as feeding suppressants or deterrents once the surimi is touched (Lindstedt, 1971).

The *O. bimaculoides* did not grow on the pelleted diets despite an estimated feeding rate of 5.26% bw·d^{-1} for the penaeid pellet. These octopuses were spawning adults and some may even have been senescent, explaining the lower growth rates and low survival (67%) over the 10 day experiment. During a 2-week preliminary trial, laboratory cultured *O. maya* ate a pelleted diet (PP) but they did not grow either. We have maintained a single *O. vulgaris* (>250 g) for 60 days on the penaeid pellet (PP) but no growth resulted. Therefore, the penaeid pellet (PP) appears to be relatively palatable to several species of octopuses but the absence of measurable growth suggests that the pellet is nutritionally deficient or imbalanced.

Cuttlefishes.—Laboratory cultured *S. officinalis* appear well suited behaviorally to studies aimed at developing a pelleted diet for cephalopods. Live prey (LS and LF) were preferred to raw (FS and RC) or pelleted diets (PP, CP and MP) and the surimi (SA) and mysid pellet (MP) were never ingested (Table 2). In contrast to octopuses, the initial responses (grabbing) by cuttlefishes seemed to be elicited by movement of the food since live shrimps (LS) and fishes (LF) were grabbed quickly while the raw or pelleted diets were grabbed more slowly (Fig. 2). However, once a food was grabbed the cuttlefishes were more likely to ingest at least half the food (Table 2) than were the octopuses and moisture (% water) did not appear to be related to ingestion as directly although the dry mysid pellet (MP) was rejected repeatedly. As a result, the foods can be divided qualitatively into three groups: (1) natural foods—live or raw shrimp and fish; (2) prepared foods with intermediate palatability—raw and pureed chicken, pureed shrimp, penaeid pellet and turkey hot dog and (3) unpalatable prepared foods—the surimi and dry mysid pellet. The fact that some cuttlefishes (30% of the trials) ate the turkey hot dogs (an artificially prepared food product composed of turkey and artificial pork flavoring), yet would not eat the surimi (composed of pollock with artificial crab flavoring) is especially interesting. Current studies are focusing on the exact nature of their rejection (texture, taste or both).

Individually reared and group-reared cuttlefishes grew when fed the penaeid pellet (PP) although growth rates were one-third to one-half of those on laboratory control diets (LS, LF and FS). Feeding was intensified in group culture because of competition. There are currently no published data on growth rates for cuttlefishes in the size range used in Growth Trial 1 (>200 g). Instantaneous growth rates projected from data reported by Pasqual (1978) and DeRusha et al. (1989) are 2–4% bw·d^{-1} for cuttlefishes approaching 200 g. The MIGR ranging from 1.0 to 2.6% bw·d^{-1} for the group-reared control diet (300–600 g) seem consistent with the normal gradual decline in instantaneous growth rate with age in cephalopods (Forsythe and Van Heukelem, 1987). Using the group-reared control diet cuttlefishes as a reference, it was clear that growth on the pelleted diet (PP) was approximately one-third of that on natural diets. As a maintenance diet this is satisfactory growth provided survival is high. Survival was high among the isolated cuttlefishes (80% at 6 weeks) but lower for the group-reared cuttlefishes (50% at

6 weeks) due mostly to cannibalism. The data for the isolated cuttlefishes agreed with the group-reared data considering that growth rates were skewed downward, presumably due to the stress of isolation. Growth Trial 2 demonstrated that smaller cuttlefishes (20 g) grow faster and ingest proportionately more food than larger cuttlefishes when fed a pelleted diet. Growth rates were again a third or less of comparably sized cuttlefishes fed natural or live food diets (DeRusha et al., 1989).

Although growth rates were depressed, feeding rates on the penaeid pellet (PP) were comparable to the control diet (Growth Trial 1, 3.5% vs. 4.0%; Growth Trial 2, 2.8% vs. 7.9–8.5% reported by DeRusha et al., 1989). The frequency of feedings (Growth Trial 1, 2, times$\cdot d^{-1}$; Growth Trial 2, 3–5 times$\cdot d^{-1}$) may have contributed to the higher feeding rate of the smaller cuttlefishes, as well as the higher metabolic rate typical of small animals. The fact that the cuttlefishes grew on the pellets while the octopuses did not grow despite similar feeding rates (3.5–8.0% vs. 5.3%) indicates that there may be significant differences in their nutritional requirements.

Palatability and Growth on Pelleted Diets.—The influence of palatability on the ingestion rate of a diet and the resultant growth of the cultured organisms cannot be over-stated. Previous research with fishes and aquatic invertebrates has demonstrated that size, shape, moisture content and biochemical composition all influence the ingestion rate of an aquatic diet (Ghittino, 1979; Halver, 1989). Stradmeyer et al. (1988) reported that young cultured salmon ingested long thin pellets at twice the rate of short fat pellets and four times the rate of round pellets while moist pellets were ingested at twice the rate of dry pellets. Cuttlefishes and octopuses also ingested moist pellets (both penaeid, PP and chicken, CP) more readily than the dry pellets (mysid pellet, MP). These pellets had different chemical compositions but the most significant overall difference was the moisture content and firmness of the pellet. The moist pellets appear to be ingested more quickly and with less loss of small particles than the dry pellet. Another aspect of palatability is the animal's response to the chemical composition and balance of the food once it has been touched and tasted. The interference or suppressant effect on ingestion by certain nutrients (i.e., amino acids and vitamins) has been described by Carefoot (1982) for the molluscs, *Aplysia kurodai* and *A. dactylomela.* He found that certain combinations of nutrients caused decreased ingestion of chemically impregnated agar wafers. The feeding rates of the cuttlefishes increased over the course of the growth trials, indicating that palatability was not inhibited but an inhibitory effect for the penaeid (PP) and chicken (CP) pellets cannot be ruled out completely for the octopuses because of the short duration of the experiment, 10 days.

Two other problems frequently associated with feeding dry pellets versus moist pellets are: (1) the increased energy content of a dry pellet causes lower feed ingestion and (2) the decreased digestibility of the dry pellet due to overloading the gastric capacity (Halver, 1989). The former results because the animal acquires the energy needed for maintenance and growth, satisfying its appetite before the overall nutritional requirements are met (NRC, 1983; Lindberg and Doroshov, 1986). The latter occurs because the mechanics of digestion and adsorption are incomplete due to rapid passage of formulated food through the digestive tract (Jobling, 1986; dos Santos and Jobling, 1988). Wells and Wells (1989) have demonstrated that the digestive tract of octopuses is a principal site for fluid absorption as well as for the digestion and the absorption of nutrients. The greater solubility of the pelleted diet may result in faster passage and reduced absorption in cephalopods. It is possible that both the increased nutrient content and gastric overloading may be working in the described octopus and cuttlefish growth studies. The higher energy content of the pellet may be limiting ingestion and/or the digestibility of the pellet may be lower than for live or fresh control diets because of more rapid passage through the gut. However, the problem seemed less significant for the cuttlefishes when compared to the octopuses, suggesting differences in their digestive physiology or nutritional requirements.

Neuroanatomical evidence also supports the observation that octopuses did not find the pellets as palatable as cuttlefishes. There are large areas of the octopus brain (primarily the frontal lobes) associated with processing taste and texture information coming from sensory cells on the arm suckers (Young, 1971). By comparison, comparable areas of the brain in squids and cuttlefishes are not as well developed, suggesting that these animals are less concerned with texture and touch learning during the feeding process. It appears that cuttlefishes rely primarily on visual inspection of a potential food while octopuses will grab food more quickly but inspect it closely with the suckers.

Feeding Costs.—Another practical consideration of any pelleted diet developed for cephalopods will be the diet's impact on reducing animal production costs. We have estimated that 50% of the labor required to culture a population of octopuses or cuttlefishes through the life cycle is involved in collecting, holding and feeding live food organisms (100 man-hours$\cdot$month^{-1}). By collecting and freezing food organisms, food-related labor costs are reduced by about one-third (33 man-hours per month^{-1}). The labor required to prepare a one-month supply (30 kg) of pelleted diet for 100 cuttlefishes (average weight 100 g) is 4–6 man-hours. The cost of raw ingredients ($21.19/kg) and the labor to prepare a one month supply results in a final cost of $680.00 or 6.80\cdot$cuttlefish^{-1}. While this provides a clear cost savings of 20% compared to live food, further reductions can be achieved by producing larger batches, by using commercial production methods and by reducing the percentage of expensive marine meals and raw shrimp used. Bulk prices for commercial pelleted feeds that are intended for fish and crustacean culture are routinely $500–$1,000$\cdot$MT^{-1}. Based on these prices, a realistic cost estimate for feeding a cuttlefish from hatching to maturity (1.0 kg bw) would be less than 2.75\cdot$cuttlefish^{-1}.

Conclusions

The major accomplishment has been the development of a pelleted diet that will be eaten, sustain life and promote growth of cuttlefishes. It is now possible to begin the process of incremental diet modification, examining nutrient requirements, digestibility and assimilation. The differences in feeding behavior, particularly ingestion and growth between cuttlefishes and octopuses indicate a need for different diet formulations. Future research will focus on several areas. Development of techniques for presenting pelleted diets to younger cephalopods and the differences in nutritional requirements for young maturing cephalopods will require additional diet formulations. Furthermore, formulation of pelleted diets for squids may be possible based on the results of this research. The pelleted diets can be used for oral administration of antibiotics (Forsythe et al., 1990); this is a critical need for the future development of commercial mariculture of cephalopods.

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