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Feeding ecology, reproductive biology and aspects related to selected marine ornamental fish; *Abudefduf vaigiensis* (Quoy & Gaimard, 1825)

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Abstract

Sri Lanka is one of the pioneers in the collection and export trade of ornamental reef fish and this industry almost depends on wild populations. *Abudefduf vaigiensis* is one of the most popular aquarium reef fish in the world and they are exploited in large numbers from coastal waters of Sri Lanka. Present study is carried out to identify their feeding and reproductive biology in order to explore the possibilities to breed them in captivity. Morphological features of 25 *Abudefduf vaigiensis* collected from coastal waters of Dondra, Sri Lanka were studied and they were related to their food preferences. Its small terminal mouth correlates to their feeding in the middle of the water column. Incisiform teeth having saw-edged cutting surfaces on both upper and lower jaws indicate their involvement in cutting the food items during ingestion process. Closely packed filament like gill rakers on the first gill arch while others bear short and blunt gill rakers, indicates their planktoivorous feeding habit. Pharyngeal teeth on their pharyngeal pads are a character useful in crushing the food before they enter the esophagus. Increase of mean relative gut length with increasing body size indicates their change of food habits with increasing size. Decreasing gastro somatic index (GaSI) from immature (developing) gonadal stage to ripe (mature) gonadal stage, indicates their low feeding rate at maturity. Stomach contents were analysed by occurrence method, number method, volume method and index of relative importance. Statistical differences in diet composition as a function of size, was assessed by chi-squared test. Food items found in the gut were mainly categorized into 5 groups: copepods, decapods, algae, unidentified fleshy parts and other items, and they confirm the omnivorous feeding habit of *Abudefduf vaigiensis*. Algae and unidentified muscle parts were the most common food items.Copepods were dominant food item, when considering the numbers but low in volume. Decapods were found in the food of large size classes. Histological studies of the ovary revealed 6 oocyte developmental stages and villi-like structures on zona radiata of oocytes and latter indicates the sticky nature of eggs. Frequency distribution studies of oocytes at different diameters indicated that they are serial spawners.

Keywords: Abudefduf vaigieunsis, stages of gonadal development, feedies ecology.

Introduction

Marine fauna (fish & invertebrates) are becoming more popular as mini-reef systems, are technologically & economically feasible (Larkin & Robert 2001). Marine Ornamental Fish trade includes around 1000 species of fish and the main supplying countries are the Philippines & Indonesia. Sri Lanka is also included among the other important exporting countries (Wood, 2001). The taxonomic profile of global trade clearly indicate the importance of fish species belong to Family Pomacentridae and also it shows that the species of pomacentridae dominate, accounting for 42%, of all fish sold internationally. *Abudefduf vaigiensis* (Plate 1) is one of the most popular aquarium reef fish in the world and they are exploited in longer numbers from coastal waters of Sri Lanka.

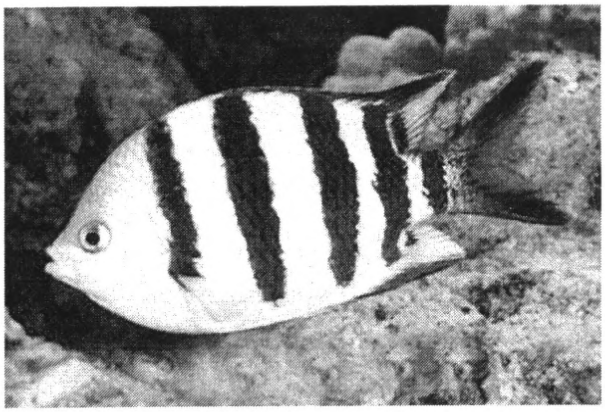


Plate 1. *Abudefduf vaigiensis* (1x1/3)

In order to help the sustainability of wild population it is important to explore the possibilities of breeding these economically important species in captivity. Prior to breeding and rearing them in captivity it is a prerequisite to have a good knowledge on the feeding and reproductive biology and ecology. Present study is designed with a view to identify the feeding biology, morphological characters related to feeding, food preference, reproductive biology and gonadal development of Abudefduf vaigiensis, which are the most important information required to develop feeding and breeding strategies for any species of fish top be reared and bred in captivity.

Material & Methods

Sampling and Measurements Taken

A. vaigiensis were collected from Doandra the southern coastal belt of Sri Lanka and 25 fish collected were immediately transported to the laboratory Total length, standard length and total weight (TW g) were measured. 25 fish studied were divided into four size classes; ≤68mm, 68-93mm, 93-111mm and ≥111mm, considering the total length. After dissecting the fish, fresh gut length (GL mm) somatic weight (SW g); fresh weight after removal of the visceral organs (gonads and other organs) and weight of stomach with contents (SW ±0.01g) were measured. Stomachs and gonads were preserved in 6% and 8% formaldehyde respectively. Stomach was cut opened and contents were preserved in 70% ethanol solution.

Relative Gut Length (RGL) and Gastro Somatic Index (GSI)

Gastro-Somatic-Index (GSI) and Relative Gut Length (RGL) were calculated according to the methods adopted by (Chiu and Pan, 2002).

Relationship between RGL and GSI with respect to size classes and maturing stages were subjected to investigation.

Methods for Gut content analysis

The contribution of each feeding item found among gut contents was determined by the frequency of occurrence (f %) (Allen, 1935), numerical composition (C_n%) (Radforth, 1940), Index of Relative Importance (Pinkas et al., 1971) and by their volume composition. Statistical analyses on effects of differences in diet composition on *A. vaigiensis* was carried out as a function of size by using a chi-square test.

Spawning pattern

Sex of *A.vaigiensis* was not detectable by means of visual examination of external characters. Ovaries were classified according to the development stage of ovaries and relative abundance of oocytes and spawning rhythm also determined (Maer, 1974).

Histology of Gonads

Histological examinations were carried out by using the methods described by Cumaranatunga (1986) and Hinton (1990) and sections were stained by haematoxylin & eosin.

Results

Morphological features related to feeding

Abudefduf vaigiensis has a short deep compressed body & have small terminal mouth (Plate 1) with Compressed incisor teeth were present in lower and upper jaws; the upper margins of the teeth that formed continuous cutting surfaces (Plate 2A). A tongue was present but there were no special modifications.

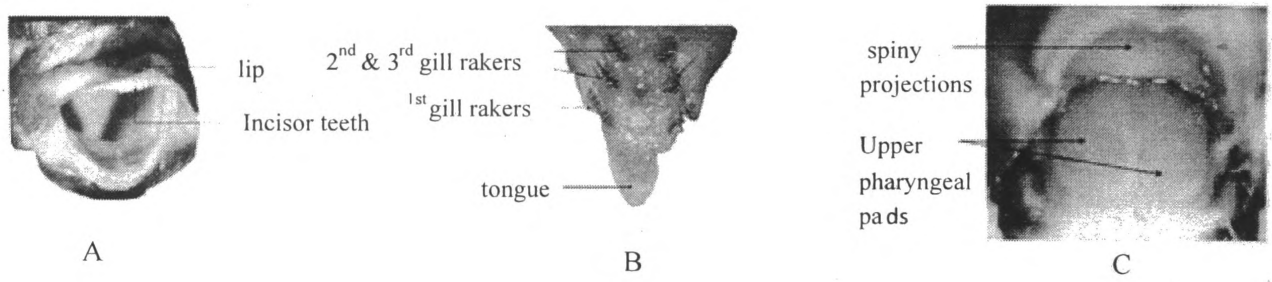


Plate 2. A-Upper jaw of *A. vaigiensis* showing the compressed incisor teeth were present in lower and upper jaws; the upper margins of the teeth that formed continuous cutting surfaces, **B- First gill arch**, showing filamentous gill-rakers and second gill arch with sturdy teeth like gill rakers and C-Upper pharyngeal teeth **of** *Abudefduf vaigiensis.*

1st gill arch possessed filament-like gill rakers and an area with spiny serrated projections were seen on either side of the filaments (Plate 2B). They were closely packed together. In second and third gill arches short sturdy teeth like gill rakers were observed. There were two pads with a 'U" shape in Upper pharyngeal teeth & its taller and prominent than the lower pharyngeal teeth. Lower pharyngeal pad is triangular in shape. Lateral view of the pharyngeal teeth gives them a spiny structure (Plate 2C). Mean values of Relative gut length (MRL) and Gastro-Somatic –Index (GaSI) for all length groups of fish are given in Table 1 and in Figure 1

Analysis of gut contents

Tables. 2, 3 & 4 provides the dietary composition in the stomach contents of A. vaigiensis of size class I (≤ 68mm), size class II (75-93mm) & size class III (98-111mm) respectively. In most stomach contents a pulpy mass was observed and its source is not clear. In addition to the items observed in the stomach, undigested spiny skeletons (were observed within the last 1/3rd of the gut (Plate 3). A chi–square test revealed that there is a relationship among *A. vaigiensis* size class with food items recorded in the stomachs of different size classes ($\chi 2 = 49.544$, df = 8, $P > 0.05$).

Table 1. Mean values of relative gut length (m.RGL) and gastro somatic index (GaSI) of *A. vaigiensis* of different size classes ≤68mm (I), 68-93mm (II), and 93-111mm (III) & ≥111mm (IV) and maturity stages.

Size class	I	II	III	IV
Maturity stage	Virgin	Developing	Maturing	Ripe
RGL	1.0462	1.1804	1.2586	1.191
GaSI	0.9925	1.0152	0.7284	0.3092

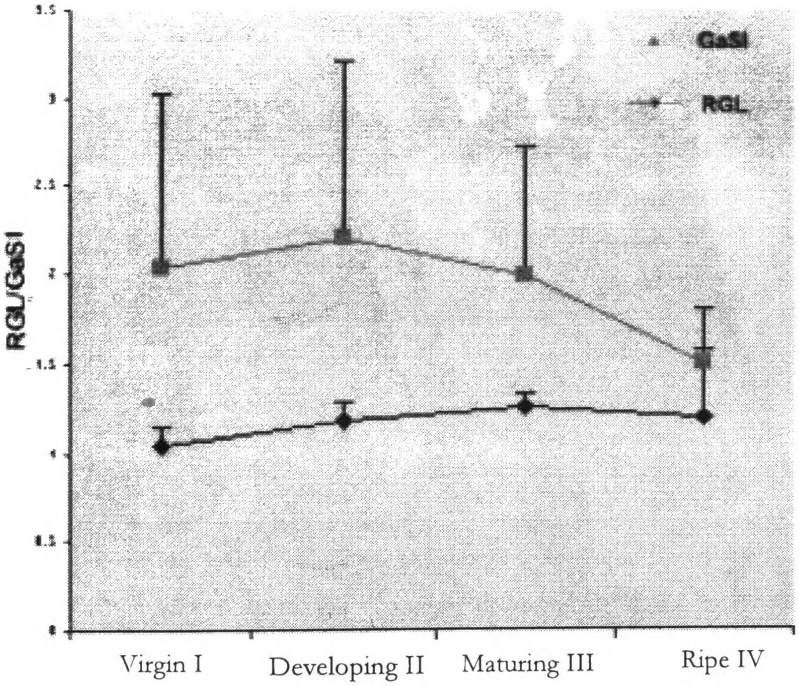


Figure 1. Changes in mean Relative Gut Length (RGL) & mean Gastro Somatic index (GaSI) according to size classes and maturity stages of *A. vaigiensis*

Table 2. Composition of stomach contents of *A. vaigiensis* of size class II (68-93mm) Number of stomachs examined [n (st)], number of individuals of a specific prey [n (i)], frequency of occurrence (f %), numerical composition (Cn %), volume composition, (Cv %) and index of relative importance (IRI).

Food items	n(st)	n(i)	f (%)	Cn (%)	Cv (%)	IRI
Copepods	5	24	83.33	27.27	0.3625	2302.61
Calanus sp.	5	3	83.33	3.409		
Cyclopoids	3	8	50	9.0909		
Harpacticoids	4	15	66.66	17.045	0.1654	1147.24
Decapoda :Naupli stages	1	6	16.67	6.8181		530.41
Decapod: body parts	1	2	16.67			
Muscle parts	2	10	33.33	11.36	16.66	933.91
Semibalanus	1	3	16.67	3.409		
Pulpy masses	1	4	16.67	4.5454	16.66	353.49
Algal remains	3	15	50	17.045	37.5	2727.25

Table 3. Composition of stomach contents of A. vaigiensis of size class III (98-111mm) Numberof stomachs examined [n (st)], number of individuals of a specific prey [n (i)], frequency of occurrence (f %), numerical composition (Cn %), volume composition, (Cv %) and index of relative importance (IRI).

Food items	n(st)	n(i)	f (%)	Cn (%)	Cv (%)	IRI
Calanoids	5	208	33.33	35.495	0.417	2431
Microcalanus sp.	2	4	13.33	0.683		
Paracalanus sp.	2	2	33.33	0.341		
Calanus sp.	4	8	26.66	1.365		
Labidocera sp	2	3	13.33	0.521	3.645	
Harpactecoids	3	22	20	3.754	0.032	
Unidentified copepods	8	120	53.33	20.478	0.26	
Decapoda						311
Natantia	6	7	40	1.194	6.593	
Pisidia sp.	1	1	6.66	0.171		
Body parts	8	18	53.33	3.072		
Muscle parts	12	37	80	6.314	37.912	3538
Naupli stages	3	3	20	0.512		
Cumaceans	2	4	13.33	0.683		
Polychaeta	1	2	6.66	0.341		
Insect parts	2	3	13.33	0.512		
Scales	5	8	33.33	1.365	2.747	137
Semibalanus	2	6	13.33	1.024		
Algae	15	127	100	21.672	48.352	7002.12

Table 4. Composition of stomach contents of A. vaigiensis of size class III (98-111mm) Numberof stomachs examined [n (st)], number of individuals of a specific prey [n (i)], frequency of occurrence (f %), numerical composition (Cn %), volume composition, (Cv %) and index of relative importance (IRI).

Food items	n(st)	n(i)	F (%)	Cn (%)	Cv (%)	IRI
Copepods	2	13	50	29.54	0.3626	1495.13
Decapoda	1	7	25	15.9	15.26	779
Muscle parts	3	9	75	20.45	38.16	4395.75
Scales	1	1				
Pulpy masses	3	7	75	15.9	7.63	1764.75
Insect parts	1	1				
Algal remains	2	8	50	18.18	30.534	2435.7

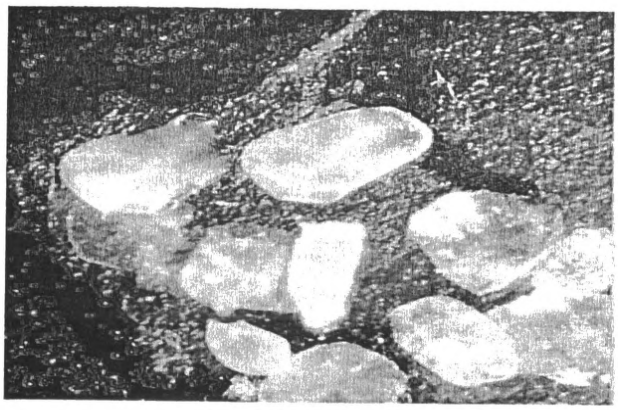


Plate 3. Undigested skeletal parts observed among stomach contents observed in the hind 1/3 of the gut of *A. vaigiensis*

Maturity stages of gonads and gametes, their histology & spawning pattern

Typical hollow structure. In immature stages two lobes of the ovaries are combined together and appear as a single structure & mature specimens the two lobes are clearly separated and they are not equal in length. Ovaries were classified into four maturing stages: Virgin, Developing, and Maturing & Ripe (Table 5). Testes of A. vaigiensis are paired structures located in the mid dorsal region of the peritoneal cavity, between the kidneys and intestine. During sexual maturation, testes show changes in colour, size and shape and they are given in Table 6.

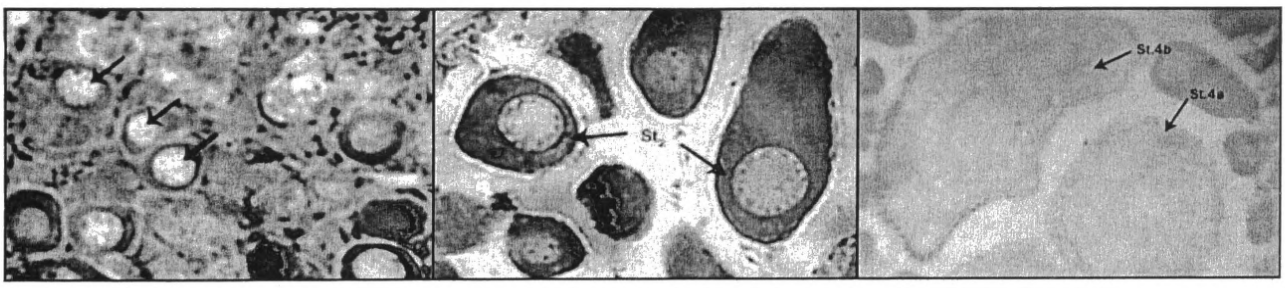
Table 5. Macroscopic structure of ovaries at different maturity stages and changes in the ovarian development of *A. vaigiensis*

Maturity stage	Macroscopic appearance
Virgin (I)	Small symmetric ovaries just recognizable as a female reproductive organ. R eddish-pink in colour
Developing (II)	Asymmetric swollen ovaries with superficial blood vessels. Eggs are clearly visible. Salmon pink in colour.
Maturing (III)	Granular slightly asymmetric ovaries. Yellow-pink in colour. opaque oocytes
Ripe (IV)	Asymmetric swollen ovaries with superficial blood vessels. Eggs are clearly visible. Salmon pink in colour

Table 6. Macroscopic structure of testes at different maturity stages and changes in the testes development of *A. vaigiensis*.

Maturity stage	Macroscopic appearance
Immature (I)	Symmetric, Yellowish-red, string like testes
Developing (II)	Elongated whitish-yellow, ribbon like testes
Mature (III)	Elongated, whitish and creamy testes.
Running-Ripe (IV)	Creamy white soft testes, presence of milt

6 oocyte developmental stages described by (Cumaranatunga, 1986) were also observed in the ovaries of *A. vaigiensis and their h*istological structures are shown in Plate 4. T*heir sizes, ratio between nucleus & cytoplasm (N/C %) and cytological structure is given in Table 7.* Histological observations indicated that the zona radiata is formed during the vitellogenic stage. Light microscopic observations showed that ZR has filamentous villi like processes. Spawning pattern of *A.vaigiensis* was recognized by using the pattern of frequency distribution of oocytes having different diameters (Figure 2) and it indicates that they are serial spawners.



A B C

Plate 4. Ovarian sections of *A. vaigiensis* belong to Size group I showing all stages below stage 2a (A); Ovarian section of size class II showing oocytes at stage 2b (B) and Section of an ovary of Size class III showing oocytes at stage 4 (C)

Table 3. Sizes, ratio between nucleus & cytoplasm (N/C %) and cytological structure of oocytes at different stages of development in the ovaries of A. vaigiensis

	Stage	Diameter (μm)	N/C %	Cytological structure
1.	Chromatin nucleolar stage	8-10	80	Single or multiple oogonia with large nucleus observed in the stroma of ovigerous folds
2a.	Early Perinucleolar stage Oocyte	18-30	65-80	Large central nucleus with small peripherally located several small nucleoli. Basophilic cytoplasm distributed like a ring in the nucleus.
2b.		40-50		Balbiani bodies found to be distributed only within the cytoplasm close to the nucleus. Cytoplasm appears to be more basophilic
2c.		60-90		Several nucleoi was observed & located close to the nuclear membrane, within the movements of balbiani bodies their basophila was found progressively reduced.
3.	Late Perinucleolar	150-220	50-60	Nucleus shown an irregular outline, nucleoli are still visible.Cytoplasm in less basophilic
4.	Endogenous Vitellogenic stage	200-265	45	Presence of cytoplasmic vesicles. In early stage of yolk vesicles formation (4a) vesicles appeared at the periphery of the ooplasm. At later (4b) whole ooplasm was filled with vesicles giving a net like appearance.
5.	Exogenous vitellogenesis	250-375	35-40	Small yolk granules are visible in the periphery of the oocyte & its srongly basophilic.
6.	Yolk granule stage	520-600		Oocytes have an irregular shape, Zona radiata increase in width and consist of filamentous villi like processes.

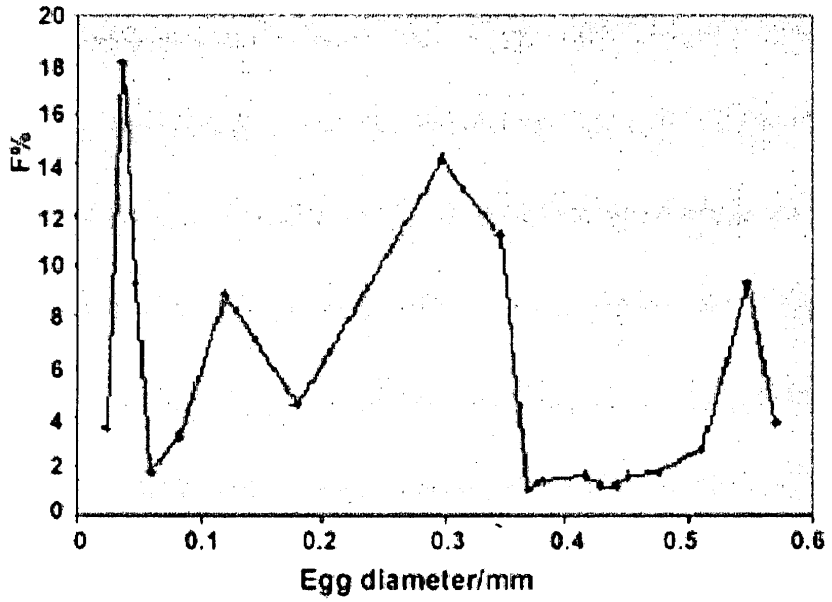


Figure 2: Frequency distribution of oocytes having different diameters

Discussion

Present study revealed several biological and ecological factors that are useful to establish captivity breeding and rearing programmes for *A. vaigiensis*. Morphological features considered in the present study in *A. vaigiensis* clearly indicated adaptations with respect to their food preference. Their terminal mouth that opens to the front is a clear indication of their feeding in the middle of the water column. This species display a bewildering array of adaptation in teeth and gill rakers, which could be considered as adaptations for successfully, capture and ingest food items. Presence of fragments of algae with holdfasts and other plant matter and fragments of muscles in the stomach suggest that saw edged incissiform teeth may play an important role in cutting fragments of algae, other plants and animals from their feeding substrata. According to Evans & Claiborne (1960), algal croppers

typically have incissiform teeth that form continuous cutting surfaces. Filamentous like gill rakers on the first gill arch, with the filaments located very close to each other pronounced in fish that feed on small planktonic organisms. Presence of planktonic microcrustacean such as *Labidocera* sp, *Microcalanus* sp, *Centrophagus* sp, *Tigirips* sp is a clear indication of their filter feeding habit. Appearance of blunt teeth on the 2nd and 3rd gill arches contributing to their carnivorous feeding habit, as Hoar *et al* (1984) have made similar observations in carnivorous fish species. According to Helfman *et al.*,(1997) pharyngeal teeth on the upper palate is mostly seen on piscivorous fish and also fine recurved teeth are present in phytoplankton feeders, which confirm the piscivorous and phytoplankton feeding habit of *A. vaigiensis*. Length of the gut is known to be a variable entity, which varies sensitively with changes in feeding conditions and maturity stages (Kapoor *et al.*, 1975). Mean relative gut length (RGL) appears to increase with increasing body size and after reaching a maximum with further increase in size it decreased again. When fish reached maturity and gonads reach the ripe stage m.rgl decreased. During the present study all the ripe fish studied had empty stomachs, which suggests that when they become ripe they refrain from feeding or take easily digestible food. Kader, *et al.*(1988) have observed a relationship of intensity of feeding of *Gobiodes rubincundus* to maturity stages and size classes and gastro somatic index has been observed to be high in maturing individuals while low in the case of ripe individuals. Observations made on the *A. vaigiensis* are in agreement with the above observations. Increase in Gastrosomatic index (GaSI) is an indication of increasing feeding frequency and food is required for development of gonads and therefore this can occur until the fish reaches maturity. According to Dadzie *et al.* (1998), spawning of the species in Kuwait waters take place from May to August in females. This period, characterized by a high gonado somatic index (GSI) and coincided with a low gastro somatic index (GaSI). Analysis of gut contents in the present study indicates that *A. vaigiensis* is an omnivorous species which agrees with the observations made on *A. vaigiensis* in the Mediterrenean Sea (Allen, 1991). Fishelson, (1970) noted that *A. vaigiensis* feeds on floating algae; whereas Emery (1973) reported that it feeds on benthic algae. Although algae were observed in the gut contents of *A. vaigiensis* during the present study, it was difficult to decide whether those were benthic or floating in nature because structural parts required for identification were lacking. A study carried out by Hiatt & Strasberg (1960) in the Marshall Islands, classified this species as a browsing herbivore, even though they found many small crustaceans among the algae in its diet. During the present study, unidentified muscle parts, semibalanus and cumaceans, were observed in the gut contents, but such items are reported in the diet of those captured in Marshall Islands or Mediterranean Sea. According to Booth & Peters (1972) noted that *A. vaigiensis* has a cleaning symbiotic activity with green turtle in the Indian Ocean. Observation of undigested spiny skeletons within the last 1/3rd of the gut indicate that they are feeding on organisms with spiny skeletons, and while the soft body parts are digested hard parts are left undigested. They graze algae as well as molted skin therefore pulpy masses found in juveniles stages could be accounted for material scraped from green turtles' skin. Proximate analyses of the ingested material should be carried out to understand the chemical composition of food required for rearing this fish species in captivity.

Frequency distribution of oocytes in mature ovaries of *A. vaigiensis* indicates their serial spawning habit and therefore if they are bred in captivity, suitable arrangements should be made to collect the eggs laid in batches or fry emerging from them. In small size classes primary stages of oocytes were observed. Fish belong to the same size class showed similar ovarian development stages, which is a good indication of their synchronous reproductive development. These damsel fishes are egg layer and they stick these eggs on substrates. (Allen, 1991). So modifications are important in sticking eggs to substrates. Observation of filamentous villi on the zona radiata in vitellogenic oocytes is a clear indication of the presence of structures helping the attachment of eggs to a substratum. Similar observations have been made by Shalanipour and Iteriaari (2004) and it has been concluded that in fish species laying adhesive eggs have a ZR with globules filament villi or honey comb like pores whereas non adhesive eggs had smooth ZR with pore canals. Therefore if *A. vaigiensis* are to be bred in captivity a suitable substratum to attach eggs should be provided.

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