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ABBREVIATION : *J. mar. biol. Ass. India*

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## STUDIES ON *ASPARAGOPSIS TAXIFORMIS* (DELILE) COLLINS AND HERVEY FROM INDIAN WATERS

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### ABSTRACT

*Asparagopsis taxiformis* (Delile) Collins and Hervey has been found to be restricted to the north Gujarat Coast and southeast coast of India. Sexual plants were found to occur in both the localities. However *Falkenbergia* bearing tetraspores has not been recorded so far from Gujarat Coast.

At Port Okha, plants begin to appear on the reef in the month of October - November, attain normal morphology and maximum length in December and dry weight in January and then gradually disappear from the reef after April - May. In nature, plants appear to regenerate by vegetative means by persistent part of the rhizomatous branches and by the production of the propagules.

Shedding of carpospores was observed in April and shedding was completed within ten days in culture. Mode of shedding of carpospores, their germination in culture and *in situ* have also been studied.

### INTRODUCTION

AMONGST the conventional sources of Iodine in India *Asparagopsis taxiformis* found to contain high amount of Iodine (Dave *et al.*, 1967).

*Asparagopsis taxiformis* and tetrasporic phase of the alga *Falkenbergia hillebrandii* have been reported earlier from Dwarka and Port Okha (Boergesen, 1933; Srinivasan, 1969), Okha, Kanyakumari and Krusadai Island (Krishnamurthy and Thomas, 1971).

The alga occurs only in a few places in India and no account of regeneration, growth, reproduction and phenology of the alga occurring in nature has been studied on our coast. Therefore an attempt has been made to study these aspects. Distribution and description of the alga studied have also been given. Simultaneously some environmental factors operating in nature and likely to influence the alga have also been considered.

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#### *Asparagopsis taxiformis* (Delile) Collins and Hervey

During the present investigation *Asparagopsis taxiformis* was collected from Kanyakumari on the southeast coast of India and on the Gujarat Coast from Armada, Mithapur, Port Okha (Pl. I) Adatra reef (Okha) and near Okha in Beyet Island from Hanumandandi reef. The alga does not occur from Dwaraka upto Diu in the Gujarat Coast.

At Kanyakumari plants were found at the level of low water of spring tides and below, attached to the rocks near Vivekananda rock. A few colonies were also found attached to the sand stone, foot-steps of the bathing pool situated near the Kanyakumari temple. Plants were collected in March 1970 and these were either vegetative or male only. At Hanumandandi reef, Adatra reef, Port Okha reef, and at Armada and Mithapur reefs plants were found to occur single or many plants close together at the level of low water of spring tides and below, attached to the corals and stones covered with a thin layer of fine sand. At Adatra severity of wave action is less as compared to the other localities.

Plants 5 cm to 35 cm in length, bushy, deep rose in colour and attached with the help of rhizoids arising from the creeping branches. Arising from the creeping branches are many erect branches of unlimited growth, these branch alternately with short laterals of limited growth upto the 3rd order. Branching is sparse at the basal part of the erect axis but dense in the upper parts. Spines small delicate upto 3 mm in length and 0.1 mm in diameter occur usually in the lower parts of the erect branches and on the creeping branches. Such spines are commonly present in *A. taxiformis*.

Both antheridial and cystocarpic plants could be collected from Okha. Antheridial plants have been described here for the first time from Gujarat Coast. The antheridial clusters (Pl. II A) occur on the distal parts of the shoots generally borne on the branches with three pericentral cells and a central cell and lie among whorls of branches with three pericentral cells only. Antheridial clusters are clavate about 770  $\mu\text{m}$  to 1232  $\mu\text{m}$  long and 523  $\mu\text{m}$  to 616  $\mu\text{m}$  broad. The cystocarps are nearly spherical about 446  $\mu\text{m}$  to 646  $\mu\text{m}$  in diameter born terminally upon a short thick branch which may be either secondary or tertiary in order. Carpospores are oval or pyriform 246  $\mu\text{m}$  to 308  $\mu\text{m}$  long and 92.0  $\mu\text{m}$  to 138  $\mu\text{m}$  broad.

Besides sexual plants of *Asparagopsis*, the tetrasporic phase of this alga *Falkenbergia hillebrandii* was found to occur as small pinkish balls, consisting of clusters of coarse filaments attached to the exposed reef at Kanyakumari. Plants were bearing well developed cruciate tetraspores at regular intervals. The filaments are 39.6  $\mu\text{m}$  to 61.6  $\mu\text{m}$  long and 14.4  $\mu\text{m}$

to 18.0  $\mu\text{m}$  broad with typical bell shaped apical cell about 10.8  $\mu\text{m}$  x 7.2  $\mu\text{m}$  to 14.4  $\mu\text{m}$  x 7.2  $\mu\text{m}$  in size.

The germlings of *Falkenbergia* attached to the branches of *Asparagopsis* were also collected from Okha in April 1968.

#### PHENOLOGY AND GROWTH

An attempt was made to study the phenology and growth of *Asparagopsis taxiformis* occurring at Port Okha. The rate of growth of the plants was studied by determining the length of the erect branches and dry weight of 10 plants collected from Port Okha at regular intervals of one month and for five months. These determinations were correlated with the state of development of the plants and ecological factors such as temperature of the sea water, air temperature, sunshine value, etc. (Fig. 1).

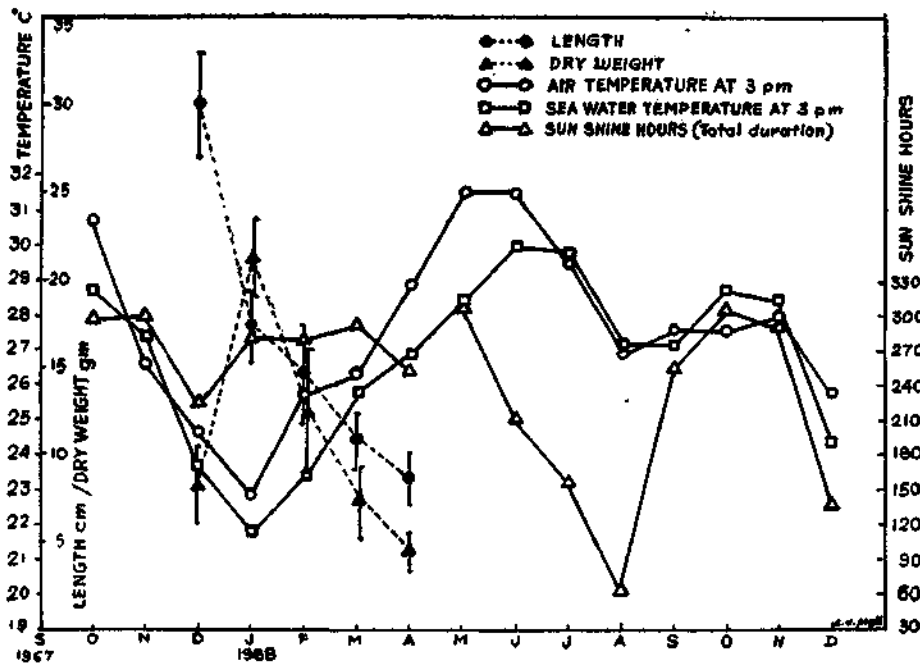


Fig. 1. Monthly variation in length and dry weight with standard deviation (vertical lines) in *Asparagopsis taxiformis* (for five months) and temperature of air, seawater and sunshine hours at Port Okha during 1967 - 1968.

**Observations:** Observations were started from October 1967. During October and November 1967, only a few small vegetative plants were available and these were measuring 3 cm to 5 cm in length. However, in December many plants appeared on the reef and during the period erect branches of the plants had grown rapidly, attaining a height of 16 cm to 35 cm. Erect branches were either vegetative or bearing antheridial clusters only. The erect system of the plant was well developed as compared to the creeping system.

During January many clusters of the plant were found adrift and these were more or less similar to the plants found in attached condition. The plants during this month were quite healthy but some of their branches with antheridial clusters had shed, thereby reducing the length of the plants. Thus their length varied from 10 cm to 22 cm. At the base of the plant a number of creeping branches were developed. These were bearing small vegetative erect branches. Probably due to this well developed creeping system the dry weight of the alga found to be maximum in January.

In February both antheridial and cystocarpic plants could be distinguished. Cystocarpic plants were rare while antheridial plants were common. Many of the taller branches bearing antheridial clusters had shed further and dry weight and lengths of the plants also declined. Algae found attached to *Asparagopsis* branches were *Ceramium rubrum*, *Cladophora rupestris*, *Ulva lectuca* and a member of the Bangiophyceae.

In March and April cystocarpic plants were mature and showed liberation of carpospores. Some of the cystocarps were found to contain germinating carpospores. During these months also both antheridial and cystocarpic plants were available but antheridial plants were more common.

In April, bleaching and shedding of the branches were severe. Branches were turning brick red and subsequently getting completely bleached. Some of the branches were also infested with bacterial growth which generally start on the reproductive structure first. Many branches were completely shed leaving stumpy axes with rhizomatous branches at the base. During this month many of the creeping branches had also become reduced. The dry weight and length of the plants were found to minimum in this month. In May only a few battered clusters of the alga were found in a drift on the shore. Thereafter gradually plants disappeared from the reef. The plants appeared again on the reef in October following. Although only one year's observations are presented here, further observations have been made since then and these are largely confirmatory.

#### REGENERATION

During field studies on *Asparagopsis taxiformis* in the months of December and January, it was observed that a large number of hook-shaped structures were produced from the apex of the ultimate branches bearing finer branches (Pl. II B). As many as 5855 hook-shaped structures were counted in a single plant (total count from five branches) in December. The hook-shaped structures when branch show slight distinction into the erect and the creeping branches (Pl. II C). These were examined further by maintaining the plants in culture. And it was observed that each of the ultimate branches having three pericentral cell and a central cell is capable of producing at its apex a hook-shaped structure, which appears to be morphologically distinct from the parent axis. These vegetative structures (which may be called as propagules) get detached from the parent axis and develop into young plantlets of *Asparagopsis* (Pl. II D, E). The same phenomenon occurred when these propagules were removed mechanically and cultured in the laboratory. Therefore, it is presumed that these propagules may serve as starting material for the fresh growth of *Asparagopsis* in nature. The fragments of the erect or creeping branches were not found to regenerate from the cut ends in culture.

## SHEDDING OF CARPOSPORES AND THEIR OUTPUT

To study the shedding rhythm of carpospores and carpospore output, healthy plants with nearly mature cystocarps were collected from Okha in April 1967. Of these a healthy shoot bearing 26 cystocarps was suspended with the help of a glass rod and a thread in sterile enriched seawater (Schreiber medium - Provasoli, 1964) contained in a beaker. The carpospores liberated were collected in a petridish placed at the bottom of the beaker.

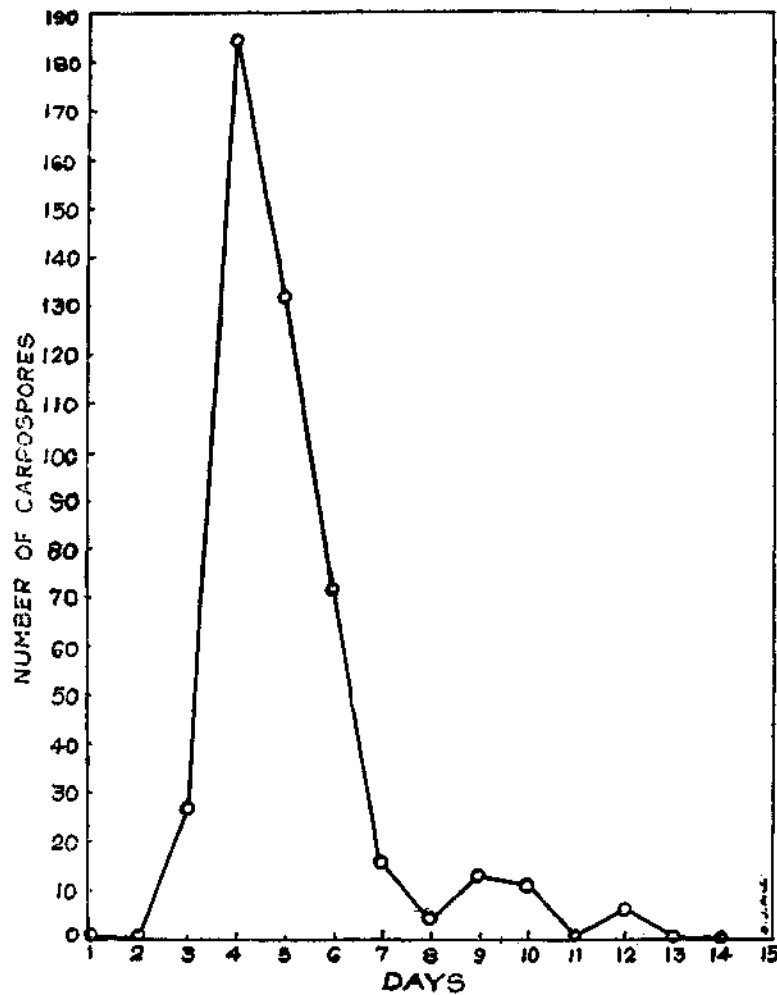


Fig. 2. Shedding rhythm of carpospores in *Asparagopsis taxiformis*.

Experiments were conducted under controlled conditions of continuous illumination 620 lux and temperature  $24 \pm 2^\circ\text{C}$ .

Carpospore liberated were counted every day with the aid of a microscope and a counter. And the petridish was replaced daily in the beaker

to collect freshly liberated carpospores. Shedding rhythm of carpospores liberated during the experiment is presented in (Fig. 2).

**Observations:** The carpospores when ripe become detached from the gonimoblasts filament on which they are produced and lie freely within the cystocarps. Sooner or later these are liberated freely through the ostiole. A slight pressure on the cystocarp wall has the effect of ejecting the carpospores through the ostiole. Occasionally carpospores were liberated enclosed in a mucilagenous sheath held at the mouth of the ostiole or lie freely at the bottom of the petridish. Soon the mucilagenous sheath gets dissolved in seawater and then a number of carpospores were liberated simultaneously.

It was observed that shedding of the carpospores was initiated on the third day after the start of the experiments. Initially a small number of carpospores were shed, but suddenly their number was increased and maximum number of carpospores was shed on the fourth day and then gradually carpospores ceased to liberate (Fig. 2). Shedding of the carpospores was completed within ten days. As many as 466 carpospores were shed from about 26 cystocarps. About 212 cystocarps were counted on a single plant indicating that about 3799 carpospores could have been shed from a single plant.

Percentage of germination of the carpospores was found to be 84% of the carpospores shed on the fourth day in the experiment.

#### GERMINATION OF CARPOSPORES

Carpospores liberated in the preceding experiments were followed to study their germination.

A ripe carpospore is pyriform or globular about  $246\ \mu\text{m}$  to  $808\ \mu\text{m}$  in length and  $92.0\ \mu\text{m}$  to  $138\ \mu\text{m}$  broad slightly narrower at one end (Fig. 3 a). The contents of carpospores are dense with reserve material and rose red in colour.

During germination a carpospore gets rounded off and soon a distinct polarity is established. The first visible sign of germination is marked by the appearance of a small hyalin region of the basal end which is slightly narrower (Fig 3 b, c). Sooner or later within 24 hours a single transverse wall is laid down towards the apex, delimiting a dome-shaped upper cell from a larger basal cell (Fig 3 c). The second division is also transverse and in the basal cell, cutting a small lence shaped cell at the base (Fig. 3 d). During subsequent development the basal cell gives rise to a tubular cell called a rhizoidal cell which subsequently divided by transpartition walls into a rhizoid filament of 3 to 4 cells with a thick mucilagenous outer wall (Fig. 3 e, f). Meanwhile the upper most cell of the three celled germling functions as apical cell of the erect filament. First it divides transversely into a short series of two or three cells germling (Fig. 3 e), this is followed by a vertical division in the above regenerants resulting in the typical *Falkenbergia* axis with three hexagonal pericentral cells surrounding a central cell, a bell shaped apical cell and vesicles (Fig. 3 f).

Meanwhile the middle cell of the three celled germling also undergoes transverse and longitudinal division forming a small basal attaching disc

from which 2 to 4 erect filaments are produced in a 20 days old germling (Fig. 3 g). Subsequently this may grow into typical *Falkenbergia* plant.

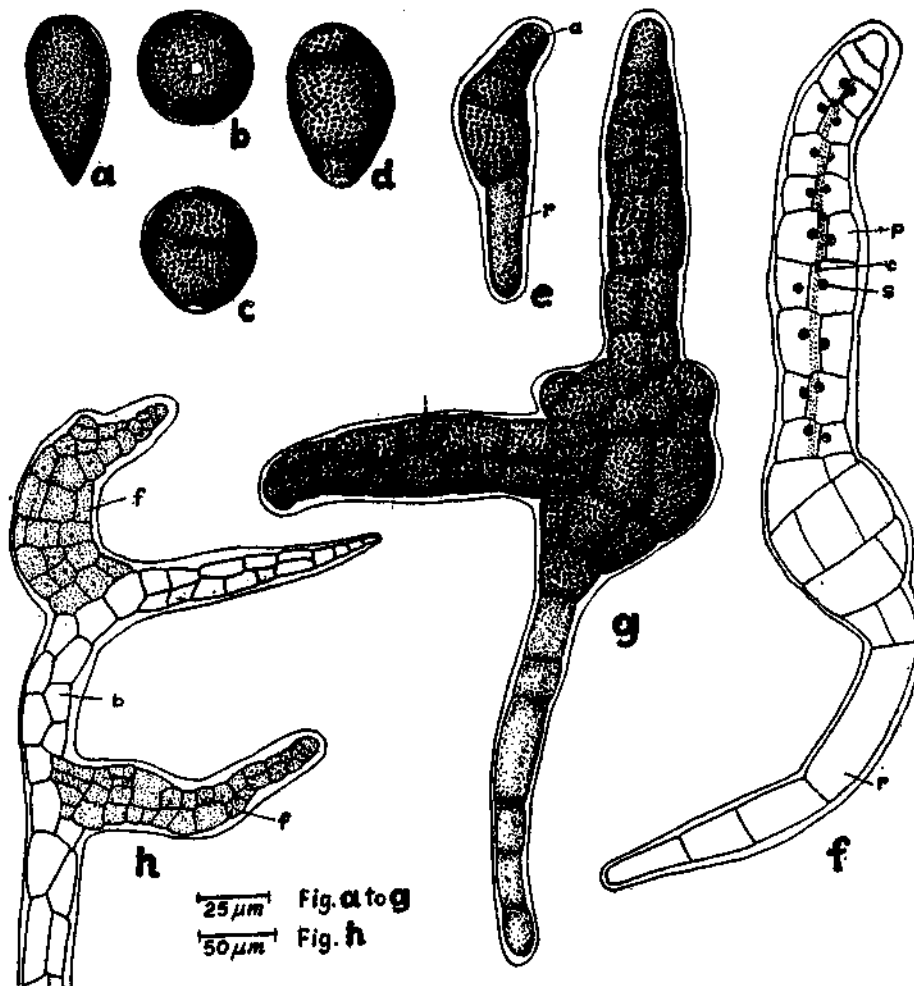


Fig. 3. Successive stages in the germination of carpospores in *Asparagopsis taxiformis*: a. carpospore before germination, b. rounded off carpospore with hyalin region, c, d. first and second divisional stages of the carpospore respectively, e. few celled germling with an apical cell (a) and a basal rhizoidal cell (r), f. *Falkenbergia* filament showing central cell (c), pericentral cell, vesicular cell (s) and apical cell, g. twenty days old *Falkenbergia* filament and h. young filaments of *Falkenbergia* (f) attached to the *Asparagopsis* branch (b).

It may be mentioned here that some of the carpospores were found under various stages of germination, while still enclosed within the cystocarp wall. In these germlings rhizoidal cell found to be very little

developed. And the proportion of the germlings having irregular segmentation was greater in enclosed germlings than those developed freely in culture. *Falkenbergia* filaments developed on *Asparagopsis* branches are shown in Fig. 3 h.

#### DISCUSSIONS

Chihara (1961) has studied the seasonal phenomena of *Asparagopsis taxiformis* occurring at Ezu Peninsula in Japan. Plants reach to a length of 15 cm to 20 cm in March. Antheridia preceding cystocarps in occurrence are met within February-May while cystocarps are found in April-June. Mature cystocarps are found after. And after May-June, number of plants decreased rapidly. The present investigations (at port Okha) reveal that the plants make their appearance in the month of October and November and continue to occur on the reef upto April-May. During October and November, only few small plants were available. However in December and January many plants appeared on the reef and these grew rapidly and attain their normal morphological features and maximum length in December. Dry weight of the alga was found to be maximum in January when the creeping system was well developed and temperature of the seawater reached to minimum 24.8°C. Thereafter due to the shedding of the branches plants gradually decline in their length and dry weight until minimum was reached in April when temperature of the seawater reached 29.9°C. Antheridial plants appeared earlier in December than the cystocarpic ones. Cystocarpic plants started appearing from February and reached to maturity in March and April. During the period October to April, temperature of air and the seawater varied from 22.2°C to 30.7°C and 21.8°C to 26.9°C (at 3.00 p. m) respectively. Sunshine values varied from 226 hours to 300 hours (Fig. 1).

Defoliation and shedding of the branches was associated with the maturation of the reproductive structures. Severe shedding of the branches was found in April, after the plants reached maturity and the reproductive structures were shed. Thereafter plants gradually disappeared from the reef. But these plants reappeared in October following. During this period (May to October) temperature of the air and the seawater remained higher and varied from 26.9°C to 31.5°C and 27.2°C to 30.1°C respectively. While sunshine values are comparatively lower and varied from 63 hours to 306 hours. Salinity of the seawater at Port Okha varied from 35.10‰ to 36.73‰ (Chauhan, 1967; Mairh, 1974). It may be mentioned here that in the laboratory culture plantlets remained healthy in the salinity ranging from 30.6‰ to 40‰, indicating that the salinity of the seawater remained favourable (throughout) the year for the growth of *Asparagopsis* in nature.

Fertile plants of tetrasporic phase *Falkenbergia* have not been recorded so far from Okha. But in nature cystocarp germinate and produce *Falkenbergia* filaments. Therefore life cycle of the alga occurring at Okha may be close to that known for British Coast (Dixon, 1965). Dixon (1965) has pointed out that the *Falkenbergia* phase never produced tetraspores in British Coast and Ireland. The plants propagate independently by vegetative means. And the actual life cycle in British waters may not be the functional sequence of sexual carposporic and tetrasporic phases as demonstrated by Chihara (1963) from Japan.





PLATE I. *Asparagopsis taxiformis* (Delile) Collins and Harvey. Habit (O. P. Mirah No. 406, Port Okha, 18.1.1968).

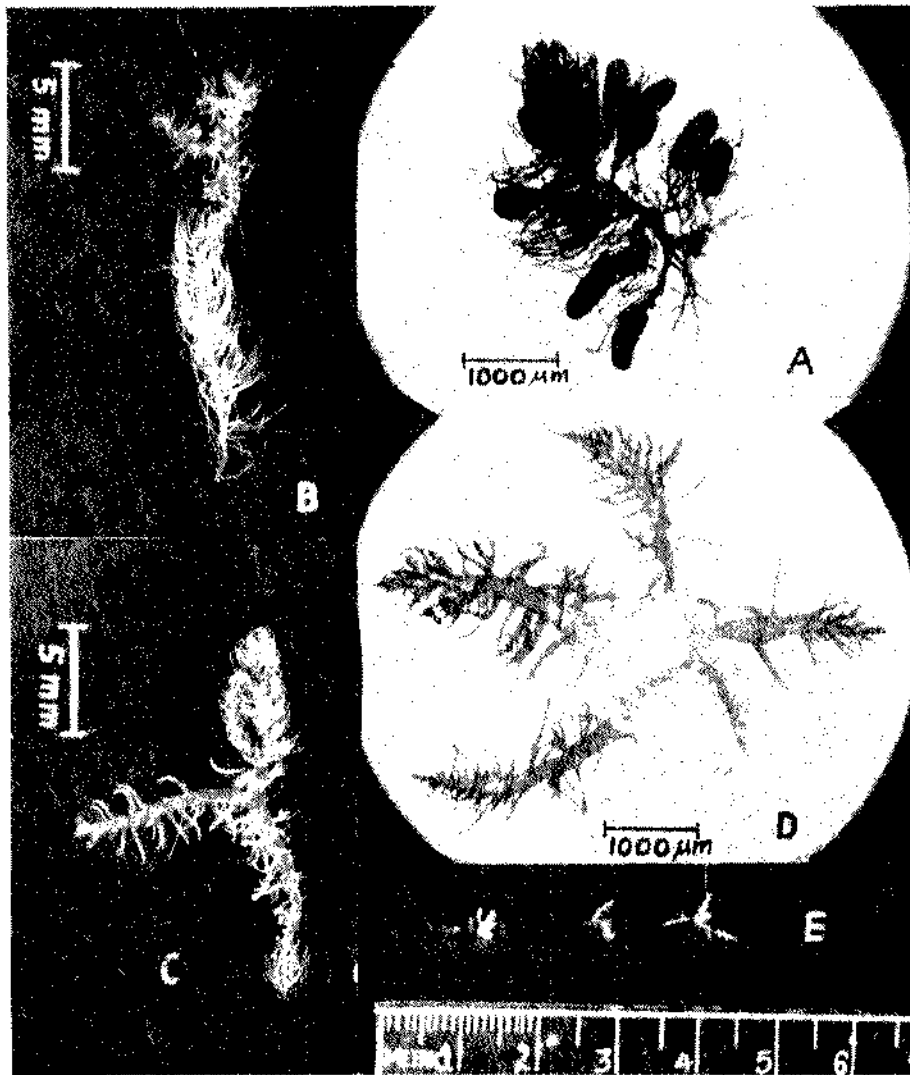


PLATE II. *Asparagopsis taxiformis* - A. Branch bearing antheridial cluster, B. Hook-shaped vegetative structure (Propagule) produced by the apical portion of the branch bearing finer branches, C. Hook-shaped structure showing distinction into aerial and creeping branches, D. Detached propagule from laboratory culture 29 days old and E. Plantlets from laboratory culture 56 days old.

Krishnamurthy and Thomas (1971) has indicated occurrence of complete life cycle on the southeast coast of India as far as Kanyakumari where reproductive plants of both *Asparagopsis* and *Falkenbergia* could be collected.

Regarding vegetative propagation in the alga, Dixon (1965) has pointed out that specimens with normal basal attachment never appear to have been reported in British Isles or Ireland which suggests that the whole population may well have been derived by fragmentation of the thalli.

At Port Okha reef, plants were bearing well developed rhizomatous attachments to the reef and after the period of shedding of the branches the remaining attached part of the plant may regenerate again in the following season. Besides this the propagation of the alga appears to be mainly by the production of propagules. Production of propagules has not been reported so far in *Asparagopsis*. Production of the tendrils in *A. hemifera* has been reported earlier (Fritsch, 1945).

Shedding of the carpospores (in culture) was completed within 10 days. Majority of the carpospores were shed within the first 4 days and maximum number of carpospores were shed on the second day of the entire shedding period (or on the fourth day from the start of the experiment). Shedding of a total of 3799 carpospores have been estimated per plant.

During germination of carpospores a comparatively long rhizoidal filaments is formed, otherwise studies on the germination of carpospores made here in *A. taxiformis* in culture and *in situ* are more or less similar to the studies made earlier by Chihara (1961) in *A. taxiformis*, by Feldmann and Feldmann (1939) in *A. armata* in culture and by Velera *et al.* (1964) in *A. armata* *in situ*.

Thus, the present studies provide further evidence to the view that these two species *A. taxiformis* and *A. armata* (which have been generally accepted as distinct species) may appear to be merely growth forms of a single taxon as earlier suggested by Schiffner (1931) and recently appreciated by Dixon (1965). It has already been shown that there is no apparent morphological difference between *Falkenbergia rufiolanosa* and *F. hillebrandii* the alternate phase in the life cycle of *Asparagopsis armata* and *A. taxiformis* respectively (Feldmann and Feldmann, 1942).

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