Bull. Mar. Sci. Fish., Kochi Univ.
No. 12, pp.9-16, 1992

Seasonal Variation of Reproductive States of the Agar-producing Seaweed, *Gracilaria changii* (Xia & Abbott) Abbott, Zhang & Xia in Thailand

Anong CHIRAPART[1], Khanjanapaj LEWMANOMONT[2] and Masao OHNO[1]

[1]*Usa Marine Biological Institute, Kochi University, Usa-cho, Tosa, Kochi 781-11, Japan*
[2]*Department of Fishery Biology, Faculty of Fisheries, Kasetsart University, Bangkok 10903, Thailand*

Abstract : Samples of the agar-producing seaweed *Gracilaria changii* (Xia & Abbott) Abbott, Zhang & Xia including environmental parameters were collected monthly from three stations, Laem Tian, Laem Sok and Ao Cho, Changwat Trat from January to December 1987.

Plants of *G. changii* from Laem Sok were found throughout 12 months of the collection, while those from Laem Tian and Ao Cho occurred only from January to July and from March to September respectively.

All plants from the three investigating areas composed of male and female gametophytic, tetrasporic and sterile plants at the same period of time. The ratio of male : female gametophytic plants from all of the three stations was approximately 1 : 1 during the early sampling period, these ratios changed at the end of the growing season. The ratio of gametophytic : tetrasporic plants of materials collected from the three stations showed markedly different pattern during the growing period.

Key words : Ecology ; *Gracilaria* ; Reproduction ; Abundance ; Seasonality ; Thai seaweeds

Introduction

Gracilaria changii (Xia & Abbott) Abbott, Zhang & Xia has been placed in the genus *Polycavernosa* (Xia and Abbott, 1987). After the reexamination of the species of *Polycavernosa* from the western Pacific by Abbott, Chang and Xia (1991), this species was transfered to the genus *Gracilaria.*

In Thailand, large quantity of *Gracilaria changii* is harvested from the east coast of the Gulf of Thailand for human food and for agar extraction. At present, the major source of plants from natural condition has been decreasing and there are insufficient wild stock for extracting agar (Lewmanomont, 1981).

Studies on seasonal variation of reproduction have been conducted on several species of *Gracilaria.* For temperate species they were done by Kim (1970) on *G. verrucosa* (Hudson) Papenfuss in Chile, Bird (1976) on *Gracilaria* sp. in Nova Scotia, and Penniman (1977) on *G. foliifera* (Forssk.) Borg. in New Hampshire. The tropical species have been reported in many aspects, such as states of reproductive plants by Hoyle(1978) on *G. bursa-pastoris* (Gmelin)Silva and *G. coronopifolia* J.Agardh from Hawaii, Umamaheswara Rao (1973) on *G. sjoestedii* Kylin from India, Trono and Azanza-Corrales (1981) on *G. verrucosa* (Hudson)Papenfuss, *G. salicornia* (C. Agardh)Dawson and *G. coronopifolia* J. Agardh from the Philippines, Hay and Norris (1984) on six sympatric species of Caribbean Panama *Gracilaria.* Most of them reported the dominance of tetrasporic over others reproductive stages.

However, there are not record on biological studies of *Gracilaria* species in Thailand. The purpose of this study was to find out the seasonal variation of reproductive stages of *G. changii,* parti-

cularly the ratio of gametophytic, tetrasporic and sterile plants.

Materials and Methods

This studies were conducted from January to December 1987 along the coast of Changwat Trat, the eastern part of the Gulf of Thailand . Three stations were established for regular monthly monitory, station 1 Laem Tian, station 2 Laem Sok and station 3 Ao Cho (Fig. 1).

Quantitative sampling was done at one month interval during low tide. Randomly quadrat method was used for this study. A 50 x 50 cm. quadrat was laid in ten representative areas of each station. All plants within a quadrat were removed and placed in a labelled plastic bag and transported to the laboratory of the Department of Fishery Biology, Kasetsart University in Bangkok. Collected materials were determined for their reproductive status. Number of tetrasporic, male and female gametophytic and sterile plants were determined. All samples were prepared as herbarium specimens for taxonomic inspection which based on Xia and Abbott (1984).

Temperature, salinity and transparency of seawater were recorded during each sampling period, using a thermometer, salinity refractometer and secchi-disc respectively.

Results and Discussion

The condition of the study areas are sandy-muddy shores, alternated with rocky shores. The bottom is intermingled with gravels, shells (*Cerithium* sp.), bivalves and hermit crabs. *Gracilaria changii* grew in rather turbid seawater (St.1 and St.2) and highly turbid seawater (St.3). The fluctuations of temperature at each station ranged from 25.5 to 35 ℃ at St.1, 26 to 37.5 ℃ at St.2 and 26 to 36 ℃ at St.3 during growing period (Table 1). Salinity of seawater ranged from 6.0 to 34.0 ‰ at St.1, 10 to 32 ‰ at St.2 and 0-33 ‰ at St.3. The ranges of transparency at St.1 which is muddy, varied remarkedly from 21 to 106 cm. while at St.2 and 3 showed lower values from 5 to 56 cm. and 2 to 61.5 cm. respectively.

Table 1. Temperature. salinity and transparency at station 1,2 and 3 from January to December 1987.

Months	St.1 Laem Tian			St.2 Laem Sok			St.3 Ao Cho		
	temp. (℃)	sal. (‰)	t.p. (cm.)	temp. (℃)	sal. (‰)	t.p. (cm.)	temp. (℃)	sal. (‰)	t.p. (cm.)
January	32.7	34.0	—	31.8	31.0	12.0	32.0	33.0	12.5
February	28.8	30.5	55.0	29.2	32.0	8.0	—	—	—
March	31.4	30.0	87.9	32.4	29.0	36.7	32.8	31.0	47.5
April	30.0	27.0	58.4	36.5	29.0	11.9	34.5	29.0	54.4
May	35.0	12.0	66.6	36.0	29.0	5.9	35.5	29.0	12.5
June	29.0	17.0	21.0	27.0	10.0	5.0	29.0	2.0	2.0
July	30.0	25.0	46.6	37.5	15.0	9.0	36.0	11.0	5.0
August	29.0	16.0	93.0	33.0	23.0	14.0	31.0	22.0	13.0
September	25.5	6.0	41.0	27.5	22.0	10.0	26.0	0	2.0
October	26.5	15.0	106.0	29.5	22.0	42.0	29.0	22.0	61.5
November	26.5	23.0	49.0	26.0	25.0	56.0	26.5	22.0	28.0
December	26.0	18.0	52.0	26.5	23.0	15.5	26.0	25.0	12.5

- no data

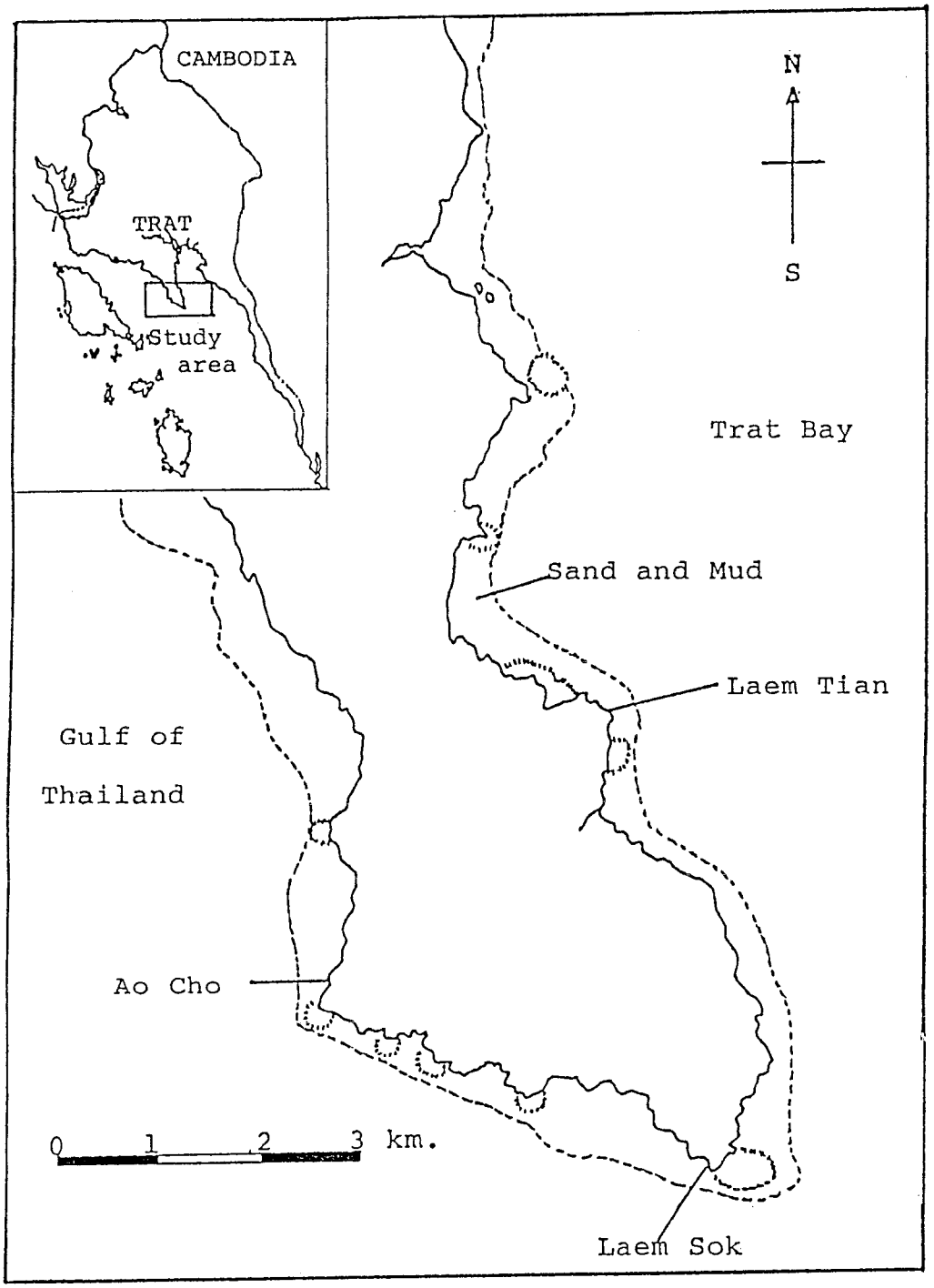

Fig.1. Map of study sites of St.1 Laem Tien, St.2 Laem Sok, and St.3 Ao Cho, Changwat Trat Thailand.

Observation on general condition of thalli was found to be clean and healthy during early growing period with numerous branching. Thalli were degenerated in the late of growing period with stump branches, roll thalli, surface gnarled and few branching.

The proportion of fertile (male, female and tetrasporic) plants from St.1 Laem Tian were reached their maximum abundance of 95.4 % in June and decreased to the minimum of 58.8 % in January, in contrast, sterile plants were found at the maximum of 41.2 % in January and the minimum of 4.6 % in June (Table 2). The percentage of fertile plants at St.2, Laem Sok, showed the maximum and minimum of 81.2 % in December and 45.5 % in September respectively, while sterile plants reached the maximum percentage of 54.6 % in September and the minimum of 18.8 % in December. Fertile plants at St.3 Ao Cho were occurred in the primary peak of 91.5 % in May and the secondary peak of 91.1 % in June, for the minimum of 62.4 % in September, while sterile plants were increased to the maximum proportion of 37.6 % in September and decreased to the minimum proportion of 8.5 % in May. These results are coincident with those of Umamaheswara (1973) on *Gracilaria edulis* and *G. foliifera* and also of Trono and Azanza- Corrales (1981) on *G. verrucosa* of which fertile plants were found throughout the year.

Table 2. Proportion of fertile and sterile thalli at studied areas in 1987.

Stations	Months	Number of sample	Fertile thalli			Total (%)	Sterile thalli (%)
			Male (%)	Female (%)	Tetrasporic (%)		
	January	600	12.2	12.8	33.8	58.8	41.2
	February	1,175	16.7	17.4	39.6	73.6	26.4
St.1	March	199	13.9	17.6	47.7	78.9	21.1
(Laem Tian)	April	740	13.4	17.3	48.8	77.4	22.8
	May	419	12.9	22.9	83.9	89.7	10.3
	June	588	8.2	31.1	58.1	95.4	4.8
	July	445	4.3	21.8	62.3	88.1	11.9
	January	1,347	11.43	13.1	29.4	53.9	46.1
	February	824	13.96	18.3	39.2	71.48	28.5
	March	721	13.5	15.8	47.4	76.7	23.3
	April	555	12.1	17.5	44.9	74.4	25.6
	May	1,046	13.4	13.5	51.3	78.2	21.8
St.2	June	453	9.7	9.1	56.1	74.6	25.2
(Laem Sok)	July	868	10.8	10.7	36.5	58.1	41.7
	August	2,049	9.2	15.7	44.2	69.1	30.9
	September	1,455	4.5	10.9	30.1	45.5	54.6
	October	1,538	15.7	23.7	25.8	65.2	34.8
	November	1,473	17.0	14.5	24.5	56.0	43.99
	December	1,192	18.5	14.9	47.8	81.2	18.8
	March	556	18.2	17.1	53.1	88.3	11.69
	April	942	15.4	16.7	42.1	74.2	25.8
St.3	May	531	18.6	16.4	56.5	91.5	8.5
(Ao Cho)	June	293	16.0	15.7	59.4	91.1	8.9
	July	312	18.6	14.1	35.3	67.9	32.1
	August	556	16.0	20.9	27.5	64.4	35.6
	September	527	15.8	18.0	28.7	62.4	37.6

From this studies, young plants were found throughout the sampling period, and showed the abundance during January and September. This result is different from the report of Raju and Thomas (1971) on *G. edulis*, in nature, young plants were seen in the months of June and July and again in December, January and February.

In consideration on sex ratio, St.1 and 2 showed the ratio of male : female gametophytic plants with seasonal variation during the sampling period, that was from January to February, at St.1 sex ratio was not different (base on an expected 1 : 1 ratio) and from March-April, this ratio variated, but no significant differences ($P<0.05$) by chi-square analysis (Table 3). Thereafter the ratio showed significantly different ($P>0.05$ and $P>0.01$) from May-July. During this period, there were conspicuous more female than male plants while at St.2 sex ratio showed no difference in early period (January and March) and middle period (May-July), but in August and October this ratio showed significant difference. In December, however, male plants were found in large number than female plants and the ratio was significantly different. On the other hand, the samples from St.3 exhibited

Table 3. Male : female gametophytic ratio at studied areas in 1987.

Stations	Months	no. of male gametophytic	no. of female gametophytic	male : female gametophytic	Chi-square @
St.1 (Laem Tian)	January	73	77	1 : 1	0.06
	February	196	204	1 : 1	0.1225
	March	27	35	1 : 1.3	0.7903
	April	99	128	1 : 1.3	3.454
	May	54	96	1 : 1.8	11.206 **
	June	48	182	1 : 3.8	76.90 **
	July	19	96	1 : 5	39.79 **
St.2 (Laem Sok)	January	157	180	1 : 1.1	1.436
	February	115	151	1 : 1.3	4.605 *
	March	97	114	1 : 1.2	1.213
	April	67	97	1 : 1.4	5.128 *
	May	140	141	1 : 1	0.000
	June	44	41	1.1 : 1	0.047
	July	94	93	1 : 1	0.000
	August	188	321	1 : 1.7	34.232 **
	September	65	159	1 : 2.5	38.612 **
	October	241	365	1 : 1.5	24.968 **
	November	251	213	1.2 : 1	2.950
	December	221	177	1.3 : 1	4.646 *
St.3 (Ao Cho)	March	101	95	1.1 : 1	0.1276
	April	145	157	1 : 1.1	0.400
	May	99	87	1.1 : 1	0.651
	June	47	46	1 : 1	0.000
	July	58	44	1.3 : 1	1.657
	August	89	116	1 : 1.3	3.298
	September	83	95	1 : 1.2	0.6798

@ Propability 0.05, degree of freedom at 1 is 3.841 and propability 0.01, degree of freedom at 1 is 6.635

* Significantly different

* * Significantly greater different

sex ratio with no difference from an expected 1 : 1 ratio throughout the growing period. Hoyle (1978) reported that *G. coronopifolia* showed significantly more male than female plants, while *G. bursa-pastoris* was reached an expected 1 : 1 sex ratio. Furthermore, Hay and Norris (1984) reported on 6 species of *Gracilaria* from Caribbean subtidal sand plain, Panama, only one specie was exhibited a significantly higher number of female than male plants while all others showed no significant difference.

On the gametophytic (male and female) : tetrasporic plant ratio, there was significant difference in all of the 3 stations (Table 4). Tetrasporic ratio were predominant higher than gametophytic ratio which corresponded with Hoyle (1978)on *G. bursa-pastoris* and *G. coronopifolia* from Hawaii. Although, in the present study, tetrasporic ratio were found higher than gametophytic ratio in most of the sampling period, except at St.3 where gametophytic ratio were conspicuous than tetrasporic ratio in August.

Table 4. Gametophytic : tetrasporic ratio at studied areas in 1987.

Stations	Months	number of gametophytic	number of tetrasporic	gametophytic tetrasporic	Chi-square@
St.1 (Laem Tian)	January	150	203	1 : 1.4	7,660 **
	February	400	465	1 : 1.2	4.7352*
	March	62	95	1 : 1.5	6.5224*
	April	227	346	1 : 1.5	24.3002**
	May	150	226	1 : 1.5	14.9602**
	June	230	329	1 : 1.4	17.1806**
	July	115	277	1 : 2.4	66.1250**
St.2 (Laem Sok)	January	337	404	1 : 1.2	5.8786*
	February	266	323	1 : 1.2	5.3242*
	March	211	342	1 : 1.6	30.5606**
	April	164	249	1 : 1.5	17.0848**
	May	281	537	1 : 1.9	79.4928**
	June	85	254	1 : 3	83.256 **
	July	187	317	1 : 1.7	33.0178**
	August	509	906	1 : 1.8	110.8240**
	September	224	438	1 : 1.96	68.5332**
	October	606	397	1.5 : 1	43.1346**
	November	464	361	1.3 : 1	12.6110**
	December	398	570	1 : 1.4	30.2076**
St.3 (Ao Cho)	March	196	295	1 : 1.5	19.560 **
	April	302	397	1 : 1.3	12.6410**
	May	186	300	1 : 1.6	26.2736**
	June	93	174	1 : 1.9	23.970 **
	July	102	110	1 : 1.1	0.2312
	August	205	153	1.3 : 1.1	7.2654**
	September	178	151	1.2 : 1	2.0548

@ Propability 0.05, degree of freedom at 1 is 3.841 and propability 0.01. degree of freedom at 1 is 6.635

* Significantly different

* * Significantly greater different

With respect to the study on the proportion of fertile and sterile plants at St.1 showed higher percentage of fertile plants than at St.2 and 3 in the same period (January-July) and St.3 exhibited the predominance of fertile plants than St.2 during March, May-July and September. The proportion peak of fertile plants at St.1 are discernable in June. This proportion, thereafter, was decreased with declining of thalli in July and was apparently absent from August to December, while at St.2 showed the high proportion in May and December, as well as St.3 indicated the high proportion from May-June together with increasing of tetrasporic plants. The reproduction was decreased in the month onwards with the decrease of tetrasporic plants.

It was obvious that, male, female and tetrasporic plants were found throughout the sampling period but tetrasporic plants were found in large number than those of male and female plants. In this case, Isaac (1956) reported on *G. confervoides* from South Africa which showed cystocarpic plants throughout the year and tetrasporic plants for most of the year. Kim (1970) reported on *G. verrucosa* from Chile that male plants were found very scarce and the major part of which was vegetative throughout the year while female and tetrasporic were approximately equal percentage. In contrast with this present study, male plants could be found throughout the sampling period.

In the case of male : female ratio in all of the three stations exhibited 1 : 1 sex ratio during the early period of sampling, while in late of sampling period showed the ratio with more female than male. This may due to the decreasing of growth rate and degenerated of the male thalli after discharging of spermatia. In contrast, after female thalli were fertilized, they have been developed into carposporophyte phase. Female thalli have begun to decline after the discharge of carpospores.

It was observed that samples from St.2 and 3 can be found gametophytic plant growing on tetrasporic plant. This may be because of the mature tetraspores could not released due to an unsuitable condition. Ultimately, the mature spores developed into gametophytes on optimum condition. Therefore, it could be found both of the male and female germinating on tetrasporic plants.

It was mentioned by Kim (1970) that the growth of *Gracilaria* was increased at high water temperature and high illumination and this favored the fruiting of tetrasporangium and carposporangium. In the present study, it was shown that under high water transparency and temperature apparently increased in the proportion of sterile plants. At low temperature and transparency the proportion of sterile plants were decreased while the total proportion of fertile plants were increased. This may due to the effect of high light intensity and temperatures on the increment of photosynthetic rate of sterile plants and developed into reproductive status. Over irradiances and temperature, however, may contribute limit growth of this plants while under optimum turbidity, turbid water where nutrients are plentiful (Hoyle, 1975).

The effect of salinity on photosynthetic rate was reported by Ogata and Matsui (1965). Also Liangnin reported that the optimum salinity for photosynthesis of *Gracilaria* species was 26-32 ‰ and their maximum respiration was in a salinity of zero and was decreased with the increasing of salinity in an inverse proportion.

The results of this study indicated that the mature fertile plants were mostly found in the salinity range of 22-31 ‰. At lower salinity, less than 22 ‰, most of the plants turned to be stumped. It may due to the effect of lower salinity on reduction of photosynthetic rate but increase in respiration rate. Trono and Azanza-Corrales (1981) observed on *Gracilaria verrucosa* and found out that the fertile plants predominated over the sterile plants during the months of high salinity and the optimum salinity was between 30-35 ‰ and the population was mostly vegetative when the salinity was 20-25 ‰.

References

ABBOTT, I.A., C.F. CHANG and B.M. XIA 1991. *Gracilaria mixta,* sp. nov. and other western Pacific

species of the genus (Rhodophyta : Gracilariaceae). *Pacific Science,* **45** (1), 12-27.

BIRD, N., 1976. Studies on *Gracilaria* : Ecology of an attached population of *Gracilaria* sp. at Barrachois Harbour, Colchester Co., Nova Scotia. *Proc. N.S. Inst. of Sci.*, **27**,144-158.

HAY, M.E. and J.N. NORRIS, 1984. Seasonal reproduction and abundance of six sympatric species of *Gracilaria* Grev. (Gracilariaceae, Rhodophyta) on a Caribbean subtidal sand plain. *Hydrobiologia,* **116/117**,63-94.

HOYLE, M.D., 1975. The literature pertinent to the red algal genus *Gracilaria* in Hawaii. Marine Agronomy U.S. Sea Grant Program, Hawaii, 340 pp.

HOYLE, M.D., 1978. Reproductive phenology and growth rates in two species of *Gracilaria* from Hawaii. *J. Exp. Mar.Biol. Ecol.*, **35**, 273-283.

ISAAC W.E., 1956. The ecology of *Gracilaria confervoides* (L.) Grev. in South Africa with special reference to its ecology in the Saldanha-Langebaan Lagoon. In, *Second International Seaweed Symposium,* edited by Trygve Braarud and N.A. Sorensen, Pergamon Press, London, pp. 173-185.

KIM, D.H., 1970. Economically important seaweeds in Chile-I. *Gracilaria. Bot. Mar.*, **13**, 140-162.

LEWMANOMONT, K., 1981. The utilization of seaweeds in Thailand. Paper presented at the Symposium on Culture and Uses of Algae in Southeast Asia, Tigbauan, Iloilo, Philippines, 7 pp.

LIANGMIN, H., 1986. Relationship between photosynthesis or respiration of some seaweeds and temperature or salinity. *Acta Oceanological Sinica,* **5** (2), 291-299.

OGATA, E. and T. MATSUI, 1965. Photosynthesis in several marine plants of Japan as affected by salinity, drying and pH, with attention to their growth habitats. *Bot. Mar.*, 8, 199-217.

PENNIMAN, C.A., 1977. Seasonal chemical and reproductive changes in *Gracilaria foliifera* (Forssk.) Borg. from Great Bay, New Hamshire (U.S.A.). *J. Phycol.*, **13** (suppl.), 53.

RAJU, P.V. and P.C. THOMAS, 1971. Experimental field cultivation of *Gracilaria edulis* (Gmel.) Silva. *Bot. Mar.*, **14**, 71-75.

TRONO, G.C., Jr. and R. Azanza-Corrales, 1981. The seasonal variation in the biomass and reproductive states of *Gracilaria* in Manila Bay. In, *Xth International Seaweed Symposium* edited by Tore Levring, Water de Gruyter & Co., Berlin. pp. 743-748.

UMAMAHESWARA RAO, M., 1973. Growth and reproductive in some species of *Gracilaria* and *Gracilariopsis* in the Palk Bay. *Indian J.Fish.*, 20, 182-192.

XIA, B.M. and I.A. ABBOTT, 1984. The genus *Polycavernosa* Chang et Xia (Gracilariaceae, Rhodophyta) : A comparison with *Gracilaria* Grev., and a key to the species. In, *Taxonomy of Economic Seaweeds with reference to some Pacific and Caribbean species.* California Sea Grant College Program, Washington, edited by I.A. Abbott and J.N. Norris, California Sea Grant College Program, Washington. pp. 157-162.

XIA, B.M. and I.A. ABBOTT, 1987. New species of *Polycavernosa* Chang & Xia (Gracilariaceae, Rhodophyta) from the western Pacific. Phycologia 26 (4), 405-418.

(Received 11 September,1991 ; Accepted 5 February, 1992)