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SEAWEED *HYPNEA* SP. CULTURE IN COX'S BAZAR COAST, BANGLADESH

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Abstract: The culture of a red seaweed *Hypnea* sp. in three locations of Cox's Bazar coast, Saint Martin Island, Inani and Bakkhali with net method of 4 × 4 m coir rope net was evaluated. Seaweed was partially harvested at 15 days interval during December 2015 to January 2016. Daily growth rate of cultured *Hypnea* sp. was significantly higher (3.21 ± 0.01% day^{-1}) in Saint Martin while Inani had the lowest (0.41 ± 0.06% day^{-1}). Biomass yield of *Hypnea* sp. (3.81 ± 0.04 kg fresh $wt.m^{-2}$) gained highest in Saint Martin than in Bakkhali (3.34 ± 0.10) and Inani (2.70 ± 0.02). Growth rate of seaweeds had a significant correlation with NO_3-N ($p < 0.05$) but not with PO_4-P. Culture of seaweed along those sites added a new dimension of prospect and possibility of seaweed mariculture in Bangladesh coast.

***Key words**:* Seaweeds, *Hypnea* sp., St. Martin, Inani, Bakkhali.

INTRODUCTION

Macroscopic marine algae, generally known as 'seaweeds', has morphological characteristic to attach and colonize over the hard substratum and shallow water zone of the coast which is suitable for their enormous growth. In recent time, seaweeds regarded as a high profile commercial marine biota for its magnitude of uses, like raw materials of bio-chemicals (agar, agarose, algin, carrageenan), dyes, food, feed, enzymes, drugs and hormones (Athithan 2014).

Most common seaweed in Bangladesh coast *Hypnea* sp. is a red algae, inhabited in shallow tropical and subtropical marine environments (Guist *et al.* 1982) and harvested in Burma, India, Philippines, Vietnam, Senegal, Brazil, USA and Bahamas (Boer 1981). Due to its tolerance over a wide range of water temperatures, salinities and light intensities (Dawes *et al.* 1976), *Hypnea* cultivation has been initiated in many countries (Humm and Kreuzer 1975, Mshigeni 1976, Guist *et al.* 1982). In India, culture of *Hypnea musciformis* was adopted in the lagoon of Krusadai Island (Rao and Subbaramaiah 1980). CMFRI has also developed a commercial cultivation technique of seaweeds using coir rope nets (Chennubhotla *et al.* 1987, Kaliaperumal *et al.* 1992). Tropical countries with coastlines are searching for seaweed cultivation as a sustainable alternative livelihood for coastal people. Seaweed farming is a

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major source of livelihood in Mindanao, Philippines (Bardach *et al.* 1972). The objective of the present study was to adapt seaweed culture technique using horizontal coir rope floating net in suitable sites of the Cox's Bazar coast of Bangladesh.

MATERIAL AND METHODS

Seaweed: Young, growing fragments of *Hypnea* sp. collected from the Saint Martin Island was used as initial seedlings. In Inani, the seedlings of *Hypnea* sp. were transplanted from seedlings of St. Martin, whereas the seedlings in Bakkhali site were transplanted from the same site. Seeding was done by inserting the young fragments of *Hypnea* sp. with an average of 4 ± 0.5 kg fw (fresh weight) and average 5 cm length in the twists of the coir ropes. Culture net were anchored by bamboo poles and held afloat at the surface level with plastics floats. The frame was tied loosely to the poles and fixed in a submerged so as to facilitate its going up and down vertically accordingly to the tide. Seaweeds were partially harvested at 15 days interval when they reached an average 30 ± 5 cm length. Partial harvest was done by clipping the algae hanging on the rope leaving the base on the rope to grow further. A total of 12 partial harvests of *Hypnea* sp. were done in three culture sites, with 4 partial harvest in each site. Daily growth rate (DGR % day^{-1}) was calculated every 15 days of culture following the formula of Hung *et al.* (2009).

Seaweed biomass expressed as fresh weight of seaweed per unit culture area (Kg m^{-2}) and computed with the following modified formula of Doty (1986): $Y = (W_t - W_0)/A$ where: Y = biomass production; W_t = fresh weight at day t; W_0 = initial fresh weight; A = area of 4 m^2 net.

Initially 1 ± 0.01 kg fwm^{-2} and average 5 cm length were used for each seedling density. A 4 × 4 m net was used for each seedling density. Four partial harvests were made including excess materials leaving the same initial density on the rope for further growth.

In order to understand whether the duration of the cultivation period influenced the DGR and biomass, yield was harvested at 15, 30, 45 and 60 days intervals of culture period. At first harvest, fresh seeding was done on new rope on 15 day. After second harvest, fresh seeding was done once after two harvests of 15 days. In third harvest, fresh seeding was done after three harvests of 15 days. Likewise, culture period up to 60 days was investigated and in each set fresh seeding time was extended by 15 days.

Study sites: Three experimental sites were selected on sheltered intertidal zones of Saint Martin Island (N20°37.043, E092°19.715), Inani (N21°13.941, E092°02.596) and Bakkhali river estuary (N21°28.500, E091°57.941) of Cox's

Bazar district (Fig. 1), along the north-eastern coast of the Bay of Bengal. The experimental site of Saint Martin Island was sandy and rocky bottom, protected by the coral reef with slanting; wave action was weak, which supported favorable environment for several seaweed species (Hossain *et al.* 2007). Inani beach site has sandy bottom with boulders, pebbles, broken shells and naturally occurring *Padina, Enteromorpha, Gracilaria, Hypnea* species were observed in this area (Umamaheswara 1974). Bakkhali river estuary site was sandy to muddy bottom with luxuriant growth of different seaweeds (Hena *et al.* 2013). The experiment was conducted for a period of 60 days from December 2015 to January 2016.

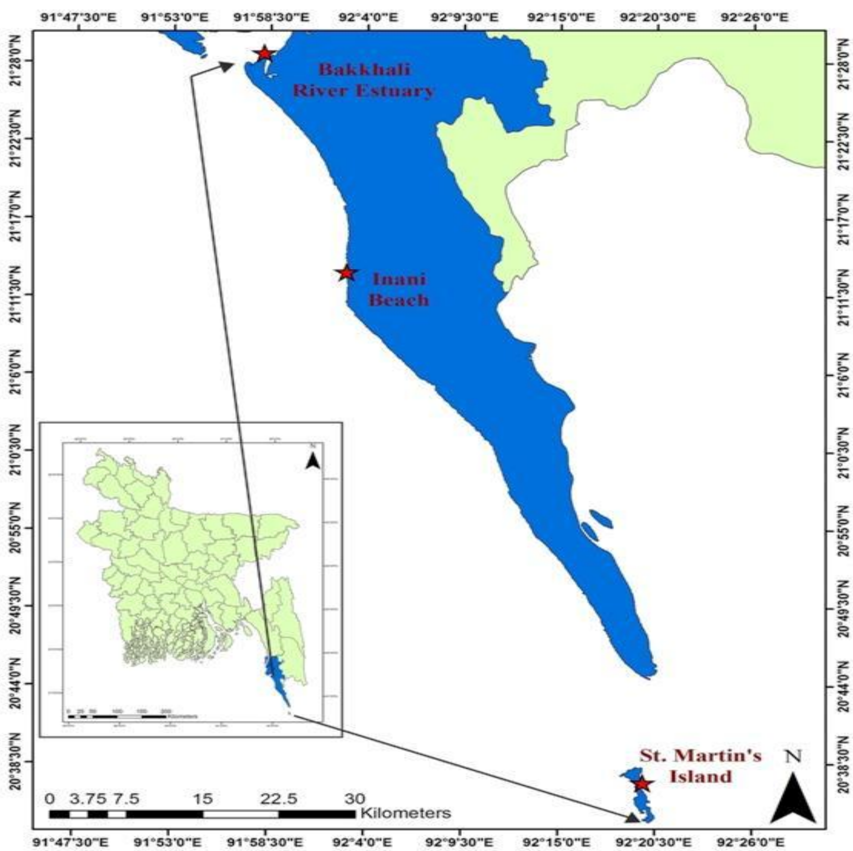

Fig.1. Seaweed culture sites at Saint Martin, Inani and Bakkhali of Cox's Bazar.

Experimental system: For seaweed culture, a square net frame of 4 × 4 m size fabricated with coir rope (obtained from coconut husk) was chosen as a support since it has loose braids to hold the seaweed seedling. Coir rope with 16 mm diameter was used and the mesh size of the coir rope nets on the culture frame was maintained 20 cm.

Study design: This was a 3 × 4 factorial study with three replicates. The experimental variables were three culture sites (St Martin, Bakkhali and Inani) and four culture durations (15, 30, 45 and 60 days).

Water quality variables: Seawater temperature, salinity, water transparency, pH and dissolved oxygen concentration were recorded at the time of partial harvest. Temperature was measured by using a standard thermometer and salinity with a hand held refractometer (Atago, Japan), water transparency by using a Secchi disk (30 cm in diameter) and pH with a pH meter (s327535, HANNA Instruments USA). Dissolved oxygen was measured immediately after sampling by standard method (APHA 2000). Dissolved inorganic phosphate (PO_4-P) and nitrate (NO_3-N) were determined according to the method described by Grasshoff *et al.* (1999). Seawater samples (in triplicate) were collected every 15 days and nutrients were analyzed in the laboratory of Marine Fisheries & Technology Station, BFRI, Cox's Bazar.

Statistical analysis: Data were analyzed by using Predictive Analysis Software (PASW) Statistics 18 and Microsoft Office Excel 2013. One-way ANOVA was done following Tukey's HSD post-hoc for multiple comparisons. Level of significance considered in this study was $p < 0.05$. Spearman's correlation was used to determine the correlation between the water quality variables and growth rate.

RESULTS

Daily growth rate (DGR): Daily growth rate of cultured seaweed was in similar trend in 3 locations except in Inani for the first 30 days. The mean daily growth rate of cultured *Hypnea* sp. attained a peak of 3.21 ± 0.01 % day^{-1} in 60-day of culture period from Saint Martin's Island. Whereas, the lowest DGR of 0.41 ± 0.06 % day^{-1} accounted from Inani during 15-day culture. Spatial variation in DGR showed a linier trend of increment from Saint Martin to Inani and found significantly higher in St. Martin than Inani after Bakkhali (Table 1). Temporal observation of DGR marked a very conventional linier increasing trend from 15 to 60 days whereas, mean DGR of *Hypnea* sp. in 60-day was significantly higher than the other culture durations (15, 30 and 45 days) with a non-significant variation between 15 and 30-day. DGR in St. Martin suddenly dropped between 15 and 30 days and resume to increase gradually up to 60 days.

Partial harvest: Mean harvest of cultured *Hypnea* sp. *was* maximum19.24 ± 0.15 kg fresh wt in 60-day of culture period from Saint Martin's and minimum 4.24 ± 0.04 kg fresh wt. collected from Inani in 15-day (Table 2). A significant increment of partial harvest resembled from 15-day to 60-day whereas, the total harvest in 60-day found exponentially 3 times higher than 15-day's harvest.

Spatial difference also illustrated by a linier no-significant variation of 48.13, 43.40 and 35.27 kg fresh wt. from Saint Martin, Bakkhali and Inani, respectively.

Table 1. Daily growth rate (Mean ± SEM) of *Hypnea* sp. in Cox's Bazar coast during 60-day culture period

Duration (day)	Daily growth rate (% day^{-1}) of seaweed in 3 locations			Total
	St. Martin	Bakkhali	Inani	
15	2.71 ± 0.09^{a}	1.71 ± 0.05^{a}	0.41 ± 0.06^{a}	4.83 ± 0.20^{a}
30	2.24 ± 0.03^{a}	1.87 ± 0.04^{a}	0.89 ± 0.02^{a}	5.00 ± 0.09^{a}
45	2.66 ± 0.03^{b}	2.47 ± 0.02^{b}	2.01 ± 0.01^{b}	7.14 ± 0.06^{b}
60	3.21 ± 0.01^{b}	2.97 ± 0.02^{b}	2.64 ± 0.02^{b}	8.82 ± 0.05^{c}
Total	10.82 ± 0.16^{a}	9.02 ± 0.13^{b}	5.95 ± 0.11^{c}	25.79 ± 0.40

DGR of *Hypnea* sp. was significantly different among the sampling sites and sampling periods. Different letters indicate significant variation ($p<0.05$).

Table 2. Partial harvest (Mean ± SEM) of *Hypnea* sp. in Cox's Bazar coast during 60-day culture period

Duration (Day)	Partial harvest of seaweed in 3 locations			Total
	St. Martin	Bakkhali	Inani	
15	5.89 ± 0.05^{a}	5.10 ± 0.04^{a}	4.24 ± 0.04^{a}	15.23 ± 0.13^{a}
30	8.36 ± 0.09^{b}	7.39 ± 0.10^{b}	5.38 ± 0.04^{b}	21.13 ± 0.23^{b}
45	14.64 ± 0.02^{c}	13.53 ± 0.14^{c}	10.83 ± 0.04^{c}	39.00 ± 0.20^{c}
60	19.24 ± 0.15^{d}	17.38 ± 0.21^{d}	14.82± 0.08^{d}	51.44 ± 0.44^{d}
Total	48.13 ± 0.31^{a}	43.40 ± 0.49^{b}	35.27 ± 0.20^{c}	126.8 ± 1.00

Partial harvest of *Hypnea* sp. was significantly different among the sampling sites and sampling periods. Different letters indicate significant variation ($p < 0.05$).

Biomass yield: Mean biomass yield found to be increased from Saint Martin to Inani, although there is no significant variation appeared among them. On the contrary, it was varied distinctly, along the study period; where lowest value was recorded in 15-days culture and then, it increased significantly till 60 days (Table 3). Maximum biomass yield of 3.81 kg m^{-2} was found in 60-day culture duration from St. Martin and minimum 0.06 kg m^{-2} obtained from Inani in 15-day sampling. Biomass yield in every culture duration had similar changing trend, which gradually increased from Inani to St. Martin.

Water quality variables: The mean water quality variables are presented in Table 4. The water temperature, salinity, pH, DO and transparency were high in Saint Martin with low PO_4-P and NO_3-N among the 3 cultured sites.

In Saint Martin seawater temperature variations ranged from 23.8°C to 25.6°C. Salinity values varied from 31.1 to 32.5‰. Water pH ranged from 6.8 to 7.5. Dissolved oxygen content ranged from 5.5 to 6.2 mg/l. Water transparency values varied from 67 to 76 cm. Phosphate content varied from 0.10 to 0.21 mg/l and the Nitrate content varied from 0.22 to 0.44 mg/l.

Table 3. Biomass yield (Mean ± SE) of cultured *Hypnea* sp. in Cox's Bazar coast during 60-day culture period

Duration (Day)	Yield (kg m^{-2}) of seaweed in 3 locations			Total
	St. Martin	Bakkhali	Inani	
15	0.47 ± 0.02^{a}	0.27 ± 0.01^{a}	0.06 ± 0.01^{a}	0.8 ± 0.04^{a}
30	1.09 ± 0.02^{b}	0.84 ± 0.03^{a}	0.34 ± 0.01^{b}	2.27 ± 0.06^{b}
45	2.66 ± 0.06^{c}	2.38 ± 0.04^{b}	1.70 ± 0.01^{c}	6.74 ± 0.11^{c}
60	3.81 ± 0.04^{d}	3.34 ± 0.10^{b}	2.70 ± 0.02^{d}	9.85 ± 0.16^{d}
Total	8.03 ± 0.14^{a}	6.83 ± 0.18^{b}	4.80 ± 0.05^{c}	19.66 ± 0.37

Biomass yield of *Hypnea* sp. was significantly different among the sampling sites and sampling periods. Different letters indicate significant variation ($p<0.05$).

Water temperature variations ranged from 21.7 to 24.1°C in Bakkhali culture site. Salinity values varied from 27.1‰ to 29.9‰. Water pH variations ranged from 5.1 to 6.3. Dissolved oxygen content ranged from 4.1 to 4.9 mg/l. Water transparency values varied from 50 to 55 cm. Phosphate content varied from 0.22 to 0.33 mg/l and Nitrate content varied from 0.74 to 1.1 mg/l.

In Inani, water temperature ranged from 22.1°C to 25.5°C. Water salinity varied from 29.5‰ to 30.5‰. pH variations ranged from 5.5 to 7.1. Dissolved oxygen content ranged from 5.0 to 6.1 mg/l. Water transparency values varied from 60 to 67 cm. Phosphate content varied between 0.19 to 0.26 mg/l and Nitrate content varied between 0.25 to 0.64 mg/l during the culture period.

Table 4. Mean values (± SE) of water quality parameters from three culture sites of *Hypnea* sp. in Cox's Bazar coast

Sites	Temp. (°C)	Salinity (‰)	pH	DO (mg/l)	PO4-P (mg/l)	NO3-N (mg/l)	Transparency (cm)
St. Martin	24.62±0.76	31.72±0.59	7.22±0.28	5.88±0.28	0.17±0.04	0.34±0.08	71.4±3.51
Bakkhali	22.92±0.94	28.82±1.15	5.7±0.49	4.5±0.32	0.28±0.05	0.89±0.13	52±2.12
Inani	23.86±1.35	29.94±0.38	6.4±0.62	5.58±0.43	0.23±0.03	0.43±0.15	62.8±3.11

Water temperature showed a significant positive correlation with water salinity, pH, DO and water transparency (Table 5). Salinity also had a significant positive correlation with pH, DO and water temperature. pH showed significant positive correlation with DO and water transparency but had a significant negative correlation with PO_4-P. Concentration of DO displayed a significant negative correlation with PO_4-P and NO_3-N, but had a significant positive correlation with water transparency. PO_4-P positively correlated with NO_3-N and negatively correlated with water transparency. Growth rate of seaweeds had a significant negative correlation with PO_4-P and NO_3-N concentrations resembled a significant correlation with water transparency (Table 5).

Table 5. Correlation matrix among growth and different water quality parameters

G. rate	W.T.	PO_4-P	NO_3-N	Sal.	pH	DO	Parameters	Temp.
							1.00	Temp.
						1.00	0.691*	Salinity
					1.00	0.710**	0.748**	pH
				1.00	0.870**	0.633*	0.765**	DO
			1.00	0.757**	0.792**	0.835**	0.829**	W.T
		1.00	–0.632*	–0.694*	–0.714**	–0.532	–0.337	PO_4-P
	1.00	0.834**	–0.656*	–0.607*	–0.521	–0.472	–0.220	NO_3-N
1.00	–0.413	–0.643*	0.154	0.056	0.236	0.228	–0.165	G. rate

Temp.= Water temperature, Sal.= Salinity, pH= pH, DO= Dissolve oxygen, W.T= Water transparency, G. Rate= Growth rate. *Correlation is significant $p<0.05$ (2-tailed) ** Correlation is significant $p<0.01$ (2-tailed)

DISCUSSION

In 60-day culture of *Hypnea* sp. with net and suspended rope methods, growth rate of 1.06 and 0.95 cm/day, respectively was observed in Saint Martin (Zafar 2007). In the present study about 3% day^{-1} was obtained, which is higher than the previous study. In the Gulf of Mannar, Bay of Bengal coast, India favorable period for cultivation of *Hypnea musciformis* was observed from July to January with the peak during August to September (Reddy *et al.* 2014). In our coast, we observed seaweeds abundance during winter with the peak during December to January. If cultured period could be prolonged, then the seaweed DGR might be higher. The lower DGR in Inani for the first 30 days could have resulted from environmental adjustment of the newly transplanted seaweed seedlings. Culture of *Hypnea musciformis* has been practiced successfully in north-eastern Brazil with DGRs near 5% day^{-1} (Wallner *et al.* 1992).

Among 3 seaweed culture locations, 3-4 folds increases was observed from partial harvest of *Hypnea* sp. Partial harvest resulted highest yield at 45-day culture period among 3 locations. Frequent harvests also allow seaweed farmers to take advantage of varying market conditions by managing supply of produce when demand is high, although seaweed maturity considerations are important (Barta 2008). Partial harvest, allows for several harvests without the need for replanting, which substantially increases biomass yield of seaweed.

The observed higher yield of red algae *Hypnea* sp. in St. Martin in the present study could have resulted due to favorable water variables of culture sites. Generally, Bakkhali and Inani are located in the upstream, where, magnitude of water quality parameters does not remain stable like Saint Martin and does not have a vast substratum facilities to form an enormous colony of seaweeds. *Hypnea* sp. was cultivated mostly in quite waters preferably in lagoons of Gulf of Mannar, India (Reddy *et al.* 2014). According to water transparency result, it has been appeared that surrounding water of Saint Martin is most translucent than Bakkhali and Inani. This is one of the main factors of lower biomass in Bakkhali and Inani compared to Saint Martin. Four-folds increase in biomass was obtained after 25 days growth with *Hypnea musciformis* in the lagoon of Krusadai Island, India on long line ropes (Rao and Subbaramaiah 1986) which is in agreement with our findings.

Temperature is a promising environmental factor that controls the growth rate of seaweeds, although this effect can be varied to species to species. Guist *et al.* (1982) found that the biomass of *Hypnea musciformis* increased by 20% when the water temperature remain 18 - 24°C. *Hypnea* is autotrophic and cannot live without photosynthesis in any conditions, as the physiological processes had a close and strong affinity to the trend of temperature change. Ding *et al.* (2013) revealed high growth rate of *Hypnea* at the temperature ranges from 15 - 25 °C. In present study temperature ranged from 22 – 26 °C, which is very suitable to culture the *Hypnea* sp. and this optimum temperature may have prominently accelerated the normal growth of *Hypnea* sp. in three culture sites. Among the three culture sites, Saint Martin showed stable and significant higher temperature within optimum level that may be another effective cause of higher biomass yield of cultured *Hypnea* sp. in this site.

Salinity of sea water is very prudent and potential factor to grow seaweeds, as it is the key determinate of osmotic balance. Nutrient accumulation from water, severely stimulated by the osmotic balance that governed by water salinity, although, range of optimum salinity differs from species to species. Zafar (2007) observed lower growth of *Hypnea* sp. from Saint Martin, when salinity dropped below 24 ‰ and better growth reported when salinity increased (>30 ‰). In the present study, the observed salinity ranged from 27 – 33 ‰ along three culture sites which did not deplete below 30 ‰ in St. Martin. Therefore, this study suggests a stable and moderate salinity as one of the key factor for having highest biomass yield in Saint Martin. Moreover, water salinity has a strong positive correlation with water pH, DO and water transparency; that means salinity plays a vital and effective role to these water quality parameters.

Significantly lower pH as observed in Bakkhali may be caused by lower salinity in this site. Concentration of DO found significantly higher in Saint Martin rather than in Bakkhali and Inani. That is another cause of higher *Hypnea* production from Saint Martin.

Transparent water allows adequate light intensity to facilitate the growth of algae. Photosynthesis process totally depends on light, so adequate light intensity is very much essential to seaweed growth and this light intensity totally depends on water transparency (Ding *et al.* 2013). In this study highest average water transparency recorded in the Saint Martin, followed by Inani and the lowest in Bakkhali.

Highest growth rate of *Hypnea musciformis* was resulted with supplementation of nitrogen and phosphorous in mariculture (Guist *et al.* 1982). The present study revealed that growth rate of *Hypnea* sp. have a correlation with PO_4-P, which indicates that phosphorus can play an effective role on seaweed growth.

CONCLUSION

Taking all these factors into account, present study recommended that cultivation of seaweeds *Hypnea* sp. on coir rope is viable and fisher folk can take up such activities as an income generating activity and thus reduces the fishing pressure in our coast. Present study also revealed that seaweed could be cultured in our coast particularly in Bakkhali river estuary and Inani beach in winter season.

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