

Comparative Evaluation of *Toddy (Borassus flabellifer SAP)* on pH Reduction and Control of Plankton Bloom in two Brackishwater Shrimp Culture Ponds

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Abstract: Water quality parameters of modified extensive shrimp (*Penaeus monodon*) culture pond were monitored. Nowadays with the development of water Probiotic, cultured shrimps performance has improved to great extent. Present experiment was carried conducted from 26th May 2010 to 14th October 2010 in shrimp grow out ponds at Diu (U.T) region of Saurashtra coast. The stocking density of tiger shrimp *Penaeus monodon* was 7.5 no/m² in both the culture ponds. Palmyra palm trees fermented toddy lead to formation of acetic acid having pH 2-3, when pH in culture water rises to about 8.7, its use can instantly reduce the pH to 2-4 and is eco-friendly. Good survival rate (79.21%), ADG (0.29), total biomass (2019.6 kg) with less Feed Conversion Ratio (FCR) (1.28:1) was recorded compare to control pond survival rate (72.02%), ADG (0.28), total biomass (1737.1) and FCR (1.28:1) from control pond P1. There were no significant differences in temperature, pH, dissolved oxygen, turbidity and salinity, whereas micro-nutrients in water parameter vary during last month of culture period. The present investigation concluded that used of toddy is practically useable by shrimp farmers and is cheaper compare to the aquaculture products available in the market for pH control. Toddy is rich in micro nutrient, which can boost up the tiger shrimp growth with low operation cost.

Key words: *Penaeus monodon*, palm tree sap, plankton, pH, shrimp culture.

Introduction:

Aquaculture is one of the fastest growing industries around the world. Capture fisheries and aquaculture supplied the world with about 106 million tons of food fishes in 2004; of this total, aquaculture accounted for 50% in 2004(1). There is an increasing demand for seafood in international market and will continue to grow in future. In India, commercial shrimp farming started during the mid-eighties. Shrimp culture in ponds has been one of the major sources of livelihoods for the coastal farmers. Diu is island were 65 ha of lands is under shrimp culture (2). At the global level, people have understood the bad effects of antibiotics and they are now shifting over to the natural products (3). Plants are the storehouse and sources of safer and cheaper chemicals. Herbs may be used as preparations containing herbal mixture or as individual ingredients in the diet (4). Natural plant products have been reported to promote various activities like anti-stress, growth promotion, appetite stimulation, tonic and immune stimulation, and to have aphrodisiac and antimicrobial properties in finfish and shrimp larviculture due to the active principles such as alkaloids, flavanoids, pigments, phenolics, terpenoids, steroids and essential oils (5,6).

Palmyra palm trees are widely grows naturally throughout the semi arid to subhumid regions of Africa, South Asia, South America, Australia and in other tropical countries (7) and in Diu Island. The sugar palm tree (*Borassus flabellifer*) is a source of material for producing a variety of product among that,

most important product of sugar palm is the sap or juice. The drink is a rich nutrient medium containing sugars, protein, amino acids, alcohol and minerals (8). In addition, emphasis on the consumption of natural foods has resulted in the use of palm sugar concentrate as an alternative sweetener (9). It also contains a dense population of yeasts (10). When it is allowed to stand, fermentation converts the sugars to ethanol and subsequently to acetic acid, leading to loss of sweetness, shortened shelf life and decreased acceptability (11) for direct human consumption. Fouling by sap is attributed to the accumulation of macromolecular or colloidal, foulant deposition increases with time (12). This study was therefore carried out to suppress the pH rise and its side effect on planktonic bloom in shrimp ponds.

Materials and Method:

Experiments were carried out at Unit No.9 (pond No 1 and 2) shrimp grow out ponds situated at North latitudes 22° 44' and 22° 41' and East longitudes 71°00' and 70°52' Patelwadi village of Diu regions, Saurashtra. Shrimp seeds stocking was done from 26th May 2010 to 14th October 2010 in ponds no. 1, 2 of 0.8 hector WSA (water spread area). Sea water was pumped from Chassi creek adjacent to pond (Fig 1). Initial water filling was to the level of 1m than bleaching was done @18-20 ppm. Pond preparation, addition of fertilizer and supplementary feeds were applied as per the standard (13). Water level of the pond was maintained approx. 1.5 to 1.7 m.



Fig 1 : Map showing the study area

The *P. monodon* seeds post larvae (PL 16) were procured from a commercial hatchery, packed @ 1500 no/plastic bag, transported to farm site having initial weight of 0.06 g. They were acclimatized to Fiberglass Reinforced Plastic (FRP) tank of 1000 L capacity, after two hours of acclimatization, the seeds were released into two ponds by 2" diameter hose pipe. Immediately after stocking the seeds were fed with commercial starter feed twice in a day as per feed manual. Pond no 1 acted as control whereas pond no 2 was treated pond with toddy (palm sap). Toddy was applied during last seven weeks of culture because organic matter/ nutrient load of feed, fecal wastes and other Probiotics application have direct co-relation with pH rises, which again affect bloom density. Toddy was usually applied at 9- 11 hr @10-25 l day⁻¹ depending upon the pH level.

Growth of *P. monodon* were assessed fortnightly i.e average daily growth (ADG) and biomass calculated as per (14). Water quality parameters like temperature, salinity, dissolved oxygen (DO) and pH were monitored every fortnightly throughout the culture period (142 days) at 06:00 hr. Salinity was measured using a hand refractometer (ATAGO, Japan), temperature was recorded by alcoholic thermometer, pH using a pH pen and DO (Winkler's method) were determined according to (15). Every month quantitative estimation of phytoplankton was done by "Direct census method" (16) and planktonic groups were identified by following (17,18,19,20).

pH level of creek seawater:

Monsoon season is the offseason for fishing activity, so all the fishing vessels i.e canoes to big trawling boat are lifted by crane on jetty for dry docking i.e boat repair, maintenance of hull etc. Every fisherman during monsoon period, washed thoroughly the fishing vessels, gears and other boat accessories by creek seawater, which ultimately lead to contamination of seawater, which directly affect the pH of the whole creek seawater. During high tide this seawater through meandering reaches, spread at the periphery of culture pond areas. Seawater through gravity reach reservoir,

were sea water is stored. Stored seawater is pumped for pond filling i.e seepaging, evaporation and water exchange of cultured pond. Most of the time pH of creek seawater is 9.2 to 9.4.

Importance of *Borassus flabellifer* tree:

Starch is one of the main reserve foods for green plants. Available starch is segregated and sun dried, which is than powdered for food purposes, like khir, kanji, payasam, kesari, uppuma, vaangibath, sago curd bhath, vadam (pappad), macaroni and spaghetti. Other than starch it is use as fuelwood (21) as per (22) it is also an important fiber-yielding woody plant.

Method of collection of toddy from palm tree:

Initially, palm trees with inflorescence are selected. The inflorescence of palms is tapped with razor sharp knife and apical tissue sliced off. As a result of which, the sap flow from the damaged inflorescence. The sap is collected in wide mouth earthen pots that are hanged close to the inflorescence. The collected sap is pooled and allowed to ferment for few hours. The thin layer of micro-flora remaining in the pot is sufficient to bring about a rapid fermentation. After fermentation, the ferment is filtered and consumed, beyond 24 hours of ferments, the pH becomes too low and the product taste sour. Preservation procedures are primarily aimed at preventing this increase in acidity, as uncontrolled development of acidity may also lead to inhibition of yeast activity and a poor alcohol to acid ratio. It is a highly perishable liquid consisting mainly of water, sugar, vitamins and many aroma and flavor components in very small amounts.

Toddy (Palm sap)

The sap, commonly referred to as 'sweet toddy' or 'neera', contain 10-16.5% w/v sugar, mainly in the form of sucrose (Table 5). This sugar is converted into ethyl alcohol during fermentation by wild yeasts and bacteria usually found in 'toddy' collecting pots (23).

Table 5 Nutritional Composition of Palmyra Sweet Sap, *Borassus flabellifer*

Specific gravity	1.07	Calcium	Trace
pH	6.7-6.9	Phosphorus(g/100 cc)	0.14
Nitrogen (g/100 cc)	0.056	Iron (g/100 cc)	0.4
Protein (g/100 cc)	0.35	Vitamin C (mg/100 cc)	13.25
Total sugar (g/100 cc)	10.93	Vitamin B1 (IU)	3.9
Reduced sugar (g/100 cc)	0.96	Vitamin B complex	Negligible
Minerals as ash (g/100cc)	0.54		

Source: (24)

Results

The water quality parameters recorded in this study are temperature, salinity, dissolved oxygen, pH and ammonia (Table 1). The temperature varied from both ponds 28.1 to 28.6. The average pH recorded 7.9 to 8.4 in pond P1 and P2. The salinity was ranged from 29.2 to 30ppt in both stations. The dissolved oxygen was recorded during the culture period in the pond P1 was

4.9ppm and pond P2 it was 4.6ppm. The ammonia was recorded 0.01 mg/l in both ponds.

During the study period the density of plankton recorded in before and after application of toddy (Table 2a). After application of toddy the plankton density decreased in every application 76.12, 75.37, 81.84, 67.34, 69.04, 73.29, 70.18, 59.34 and 68.21 respectively in pond P2.

Table 1 Average water quality parameters in control (P1) and toddy (sap) treated (P2) ponds.

Parameters	Control (P 1)	Before Sap Treated (P 2)	Control (P 1)	After 5 hr. sap Treated (P 2)
Temp (°C)	28.6	28.1	28.6 ± 1.73	29.0 ± 3.64
pH	8.2	8.4	8.2 ± 0.33	7.9 ± 0.42
Salinity (‰)	29.2	30	29.2 ± 2.94	29.2 ± 2.89
DO (ppm)	4.9	4.6	4.9 ± 0.61	4.6 ± 0.82
Ammonia (mg/l)	0.01	0.01	0.01 ± 0.2	0.01 ± 0.00

Table 2a Comparison of planktonic density before & after application of toddy.

Month 2010	Before 3hr. of sap application		After 5 hr. of toddy application			
	Control (P1) (no x 10 ⁶ m ⁻³)	Sap treated (P2) (no x 10 ⁶ m ⁻³) L1	Control (P1) (no x 10 ⁶ m ⁻³)	Date of Toddy application	Sap treated (P2) (no x 10 ⁶ m ⁻³) L2	Plankton no reduce after sap applied. L1 - L2 = (no x 10 ⁶ m ⁻³)
May	62.28	74.28	63.24	-	NA	-
June	97.52	114.54	102.38	-	NA	-
July	89.74	95.22	87.15	-	NA	-
August	74.51	83.17	75.37	16th Aug	76.12	7.05
	68.28	79.23	71.08	21th Aug	75.37	3.86
	73.18	84.51	76.27	28th Aug	81.84	2.67
September	67.57	71.29	67.43	4 th Sept	67.34	3.86
	72.18	73.07	73.18	9 th Sept	69.04	4.03
	59.91	76.81	61.37	20 th Sept	73.29	3.52
	66.19	72.47	64.28	29 th Sept	70.18	2.29
October	60.18	66.12	61.28	1 st Oct	59.34	6.78
	63.28	70.38	63.22	9 th Oct	68.21	2.17
Avg.	71.23	80.09	72.18	-	71.19	4.02

Table 2b Planktonic density with specific to season

Phytoplankton Density				
		No. of spp. Specific to Season		
Algal groups	% (cells $\times 10^6 \text{ ml}^{-1}$)	Summer	Monsoon	Spring
Bacillariophyceae	33.20	7	11	5
Cyanobacteria	20.71	16	12	9
Chlorophytes	43.06	11	9	13
Euglenoids	<1.00	1	-	1
Dinoflagellates	<1.00	1	1	1
Miscellaneous*	1.03	Less	More	Moderate
Grand Total	100.00	36	33	29

* Shells, fish egg, shrimp larvae, foraminiferans, molluscans and plant materials.

The maximum production was reported in pond P2 (2019.6kg) and minimum was recorded in pond P1 (1737.1 kg) (plate 2).The good survival was recorded

79.21% in pond P2. The average FCR was calculated 1.29 in both ponds (Table 3).

Table 3 *P. monodon* recorded after harvesting of P1 and P2.

Harvest record	Pond1 (P1)	Pond2 (P2)
Total stocking (no.)	60000	60000
Survival%	72.02	79.21
Avg. wt. of shrimp (g/pcs)	40.2	42.5
Count (no/kg)	24.8	23.5
Total biomass (kg)	1737.1	2019.6
FCR	1.3:1	1.2:8
Feed utilized (kg)	2258.1	2585

**Plate 2 Harvested shrimps**

Table 4 Quantity of toddy applied during pH rise in shrimp pond

Sr. no	pH rises	Toddy pH	Qty. of Toddy applied
1	9.3 – 9.1	2.2-2.3	30-35 lt
2	9.0 - 8.8	2.4-2.5	25-30 lt
3	8.7 – 8.5	2.4-2.5	20- 25 lt
4	8.4 – 8.2	2.4-2.5	15- 20 lt

Table 6 Plankton recorded in shrimp pond (P1 & P2)

Phytoplankton	<i>Chaetoceros spp.</i> , <i>Cosinodiscus spp.</i> , <i>Nitzshia spp.</i> , <i>Gyrosigma spp.</i> , <i>Chlorella spp.</i> , <i>Navicula spp.</i> , <i>Chlamydomas spp.</i> , <i>Daphnia spp.</i> and <i>Dinoflagellates</i>
Zooplankton (Copepods like Calanoid, Cyclopoid, Harpacticoid)	<i>Oithona brevicornis</i> , <i>Microsetella spp.</i> , <i>Pseudodiaptomus spp.</i> , <i>Paracalanus spp.</i>
Rotifers	<i>Branchionus spp.</i> , <i>Keratella spp.</i> , Nauplii and eggs.
Euglenoids	<i>Euglena spp.</i>
Miscellaneous	Shells, fish egg, shrimp larvae, foraminiferans, molluscans and plant materials.

During the study period in both ponds the plankton samples were collected and analyzed (Table 6). The phytoplankton species recorded in both ponds are *Chaetoceros spp.*, *Cosinodiscus spp.*, *Nitzshia spp.*, *Gyrosigma spp.*, *Chlorella spp.*, *Navicula spp.*, *Chlamydomas spp.*, *Daphnia spp.* and *Dinoflagellates*. The zooplankton species recorded in both ponds are *Oithona brevicornis*, *Microsetella spp.*, *Pseudodiaptomus spp.*, *Paracalanus spp.*, *Branchionus spp.*, *Keratella spp.* Copepods, Nauplii and eggs, *Euglena spp.*, Shells, fish egg, shrimp larvae, foraminiferans, molluscans and plant materials.

Discussion

Water parameter was conducive in both the pond P1 & P2 with small variation having (Table 1) temp (°C), pH, salinity (ppt), DO (ppm) and Ammonia (mg/l), where P1 28.6 ± 1.73 , 8.2 ± 0.33 , 29.2 ± 2.94 , 4.9 ± 0.61 and 0.01 ± 0.2 but when pH rises above 8.4 onwards, toddy was applied in treatment pond 2, after 5 hours of application, slightly change in water parameters were recorded with compare to control P1 where P2 water parameter were 29.2 ± 3.64 , 7.9 ± 0.42 , 32.4 ± 2.89 , 4.3 ± 0.82 , 0.01 ± 0.0 . In both the pond, optimal salinity values range between 19- 29.8 ppt, whereas temperature at a different of 23–33°C are well tolerated (25) same trend was observed where temperatures vary with 0.1 – 1.3 °C when toddy was applied to the P2. *P. monodon* may be one of the most efficient osmotic and ionic regulators among Penaeid

species (26); same trend was observed when salinity, during monsoon season lowers upto 14 ppt.

(27) reported that in natural populations, the pH may vary from 5 to 9, but for aquaculture facilities it is recommended to be in the range 6.5–9.0; present study during early days of culture (DOC) pH was stable 7.8- 8.2., he further added that low pH increases nitrite toxicity to fish and may have similar effects on shrimp. pH of brackish water is usually not a direct threat to the health of the shrimp, since brackish water is well buffered against pH changes, and pH mostly remain within the range of 7.8 to 8.3. (28) reported that in highly intensive prawn ponds, the pH decreases only by 0.7- 1.3 unit to about 7.3 at the end of the crop; same trend was observed in present study with pH rise of 0.2 to 1.1 in normal condition, pH will of-course vary with the water source where Chassi creek seawater has pH of 8.9 – 9.8 therefore, adverse effects on shrimp culture are uncommon, again pH of pond soil was between 6.6 – 6.9, which does not affect the pH rise of cultured water. Fish and other vertebrates have an average blood pH of 7.4, pH higher than 7 is not lethal to *P. monodon* (29). Maximum pH 9.3 was recorded in P2 and was controlled by application of 35 l toddy. Seawater normally has pH value between 7.8 and 8.3 and has a good buffering capacity due to the available free bicarbonate. (30) reported that pH within the requirements of 7.5 – 8.5 has best result; in present study pH was balance by application of toddy sap pH 2.2- 2.5 level and survival of 70% was recorded. (31) reported that increases in % of unionized ammonia in a given ammonia concentration may cause more toxic

form of ammonia; In the present observation were ammonia has not raised above 0.02 ppm, whereas according to (23) contain fermenting wild yeasts and bacteria; in present observation toddy sap contain fermenting bacteria, which may have suppress the nitrification bacterial process in the pond.

Initially plankton density in control pond (P1) was $62.28 \times 10^6 \text{ m}^{-3}$ and (P2) $74.28 \times 10^6 \text{ m}^{-3}$, but in the month of August plankton density in P1 was $74.51 \times 10^6 \text{ m}^{-3}$ and plankton density rise to $75.37 \times 10^6 \text{ m}^{-3}$ whereas in P2 before use of toddy sap plankton density was $83.17 \times 10^6 \text{ m}^{-3}$ and after 5 hrs of treatment plankton density reduces to $76.12 \times 10^6 \text{ m}^{-3}$ so reduction of plankton density was $7.05 \times 10^6 \text{ m}^{-3}$ (Table 2a). Maximum. Plankton density reduce was on 16th August by $7.05 \times 10^6 \text{ m}^{-3}$ and was the highest plankton reduced no. in the record. When P2 culture water pH recorded to 9.3, the toddy used was 32 lit having pH 2.2. Planktonic density (Table 2a) shows that application of toddy may have boost up few plankton density (acidophilic plankton) but in other case, it's acidic effect have reduces the plankton density whenever toddy was applied in the range from $2.17 \times 10^6 \text{ m}^{-3}$ to $7.05 \times 10^6 \text{ m}^{-3}$, whereby after application of toddy, a white foam was observed, which indicate the bloom crash (Plate 1). In control pond the plankton density remains the same with the maximum rise during month of June 2010 by $4.86 \times 10^6 \text{ m}^{-3}$ and min. density reduction by $0.14 \times 10^6 \text{ m}^{-3}$ during 4th September, whereas plankton was reduce during 29th September 2010 by $-1.91 \times 10^6 \text{ m}^{-3}$ numbers.

(32) reported that percentage of Bacillariophytes decrease with decrease in salinity;

same trend was recorded, before stocking of *P. monodon* seeds Bacillariophyta density (28.37%) which increases to (38.04%) the salinity was around 17- 23 ppt (Table 2b). The avg. population of Chlorophyta (43.06%) was highest followed by Bacillariophyta (33.20%) and Cyanophyta (21.74%), Euglenoids and dinoflagellates had fewer species with mean densities of less than $1.0 \times 10^3 \text{ cells ml}^{-1}$ whereas miscellaneous like broken shells, fish egg, shrimp larvae, foraminiferans, molluscs and plant materials were recorded 1.03%. (33) reported that change of watercolor or its intensity indicates changes of phytoplankton flora and densities and light availability to phytoplankton is more important for organic production; same trend was observed in present study, here water color change was observed after application of toddy i.e from dark grey to light grey or brown hue to light brown or sometime from greenish to bright green. Variations were recorded in the phyto and zooplankton populations. Phytoplankton were represented by *Chaetoceros* spp. *Coscinodiscus* spp., *Nitzschia* spp. *Gyrodinium* spp. *Chlorella* spp. *Navicula* spp. *Chlamydomonas* spp. and *Daphnia* spp. While rotifers, copepods and mysids were the predominant zooplankton. Rotifers were represented by *Branchionus* spp., *Keratella* spp. Nauplii, copepodids and eggs. Copepods were dominated by *Pseudodiaptomus annandalei* *Paracalanus aculeatus* *Oithona brevicornis* *Microsetella* spp. Mysids were represented by *Acetes* spp and *Mesopodopsis* spp. Miscellaneous groups of plankton mostly consisted of calcareous shell, shrimp larvae, foraminiferans, molluscan larvae, fish egg and plant materials (Table 2c).



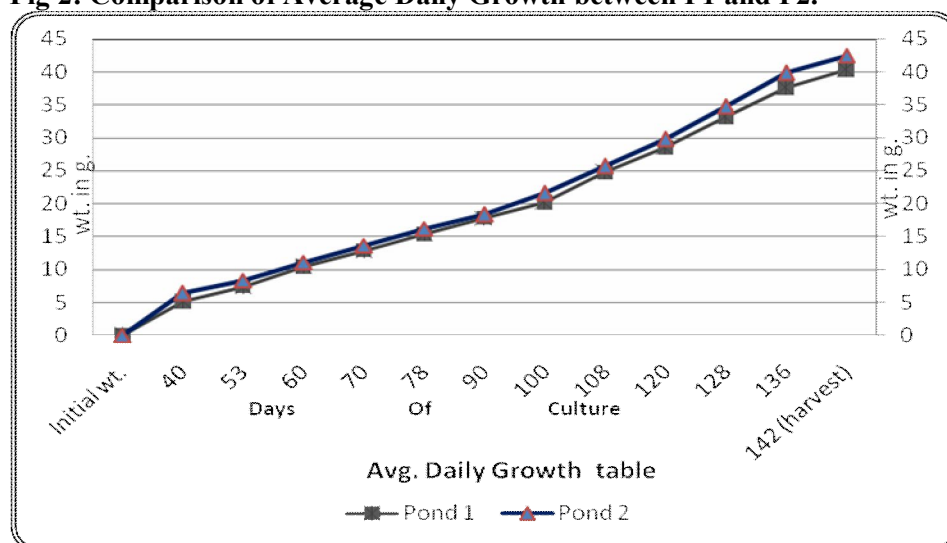
Plate 1 Plankton crash in the pond P2

(34) reported that farmers rarely use inorganic fertilizers to stimulate natural productivity; in present study dominance of phytoplankton was noticed during initial days of pond fertilization and rainfall started the density of Bacillariophytes raises to (38.04%) which reduce, drastically on salinity rises. Chlorophyta were higher during first-month with approx. $43.06 \times 10^6 \text{ m}^{-3}$ and declined thereafter $27.41 \times 10^6 \text{ m}^{-3}$, and copepod were moderately dense particularly during initial days which reduces decently as culture progress. (35) reported that progressive increase in the density of mysids during the culture may be attributed to their predation on zooplankton; in present study, Mysids were low in the first month ($3.46 \times 10^4 \text{ m}^{-3}$) and their density increased progressively ($5.97 \times 10^4 \text{ m}^{-3}$) these may be due to monsoon season, mysids larvae density proliferate in creek, which by chance may have enter the culture water through water pumping or given out young ones. (36) reported that in the gut contents of *P.monodon* generally algal, micro-crustaceans, molluscans and polychaete worms occurred; In the present observation plankton, pellet feed and sand grain constituted the gut contents of *P. monodon*. As this species is an omnivore scavenger, its gut content showed detritus and the presence of sand. The gut contents of the shrimps, further indicated that *P. monodon* feeds on organisms available abundantly in the pond. (37) also reported that wild *P. monodon* feeds on a variety of organisms according to their seasonal availability. (38,39) reported that culture animal has their own size-selectivity for prey and

displayed size selectivity during the youngest stages; in present observation after stocking of *P. monodon*, bloom density place a major role for speedy growth and to overcome size variation among seeds stocked. As per (40) Shrimps are fed 3–4 times per day and the ration is adjusted according to shrimp size and appetite; in present study as per check tray observation, feeding quantity was reduce during molting period and increased when feed was consumed from check tray. (41) reported that by adding *Kappaphycus alvarezii* sap to *P. monodon* feed good survival, ADG, total biomass (kg) and low FCR with 89.70%, 0.277, 2007.48 kg and 1.26:1 was recorded; in present study (Table3) 79.21%, 0.29, 1763 kg and 1.28:1 respectively. Avg. Daily Growth was comparatively higher with 0.29 gday^{-1} (Fig 2), this may be due to conducive water parameter by using toddy for pH control.

The major chemical reactions occurred during the heating process of sugar riched products such as palm sugar concentrate are Inversion, Maillard and Caramelisation reactions (42). Micro-organisms in palm sap also play important role in producing reducing sugars (glucose and fructose) from sucrose via hydrolysis reaction; in present study toddy's effects on pH control was excellent and it's side residual effects on water parameter, produce conducive environment for tiger shrimp culture, where we got excellent result of 1772.4 kg total harvest (Table 3).

Fig 2: Comparison of Average Daily Growth between P1 and P2.



Conclusion

Results showed that use of toddy a herbal sap for pH control to shrimp culture water can contribute to the production of culture animal without any adverse effect to culture organisms. This is supporting that inclusion of toddy sap has resulted in higher shrimp growth and survival. Use of Palmyra palm trees toddy is practically useable by fish farmers and toddy is cheaper compare to the aquaculture products available in the market for pH control. Toddy herbal effects can be useful for fertilizing few planktonic specific species, which may be directly depending upon the acetic acid to some extent. Sap quantity may be increased or decreased according to the pond pH level and loss of toddy nutrient in seawater cause conducive environment for culture organisms. Use of Palmyra

palm toddy in animal production can provide livelihood for many coastal agricultural farmers for generating additional income as well as aquaculturist procure an eco-friendly herbal composition for pH control.

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