

Field culture of American strain of *Artemia* in India

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Pilot scale culture of *Artemia* was carried out in a 0.22 ha condenser unit of salt pans. Pond was fertilized with inorganic salts. The nauplii of San Francisco Bay, USA strain were inoculated. Within a period of 5 months a production of 20.6 kg dry cyst.ha⁻¹ was achieved. Harvested cysts had a diameter of 201 µm with a hatching efficiency of 235000 nauplii.g⁻¹. The level of highly unsaturated fatty acid (W3 HUFA) was 27.8 mg.g⁻¹ dry wt.

About 58000 ha of brackish water area along Indian coastline is presently under commercial aquaculture producing about 30000 tonnes of shrimps annually. The futuristic projection is addition of about 70000 ha more by the year 2000. To realise this objective shrimp hatcheries have been established in all maritime states.

Notwithstanding the technical progress that has been made in running shrimp hatcheries, supply of feed-live or artificial-still remains the most important constraint to the success of such hatcheries. Formulated feeds are increasingly being used, but less attractive compared to live feed especially the *Artemia* (as nauplii) in terms of edibility, nutritional value and conversion efficiency. As a consequence, dependence on *Artemia* for shrimp hatcheries is unavoidable and bound to increase with the projected increase in brackish water aquaculture for shrimp. In view of the limited availability of quality cysts, culture of *Artemia* was undertaken to assure a constant supply of cysts for shrimp hatcheries. With this objective, a pilot scale culture of *Artemia* was successfully carried out at the salt pans at Singach (lat. 23°30'N; long. 70°12'E) and the results are presented.

Materials and Methods

Singach salt work is located 50 km west of Jamnagar, Gujarat covering an area of 1214 ha comprising of reservoirs, condensers, brine tanks and crystallizers. Seawater (sal. 35 × 10⁻³) is pumped into reservoirs and circulated by gravity flow through a series of condenser pans and finally evaporated to obtain salt crystals. Charging of crystallizers with high density brine commences by October and salt production starts from December/January and lasts till June.

Water level in the culture pond (area 0.22 ha) was maintained at 75 cm. The intake water to the pond was passed through nylon screen (400 µm) to prevent predators. The salinity of the pond was 100 × 10⁻³ in early October 1989. The pond was fertilized at the rate of 50 kg.ha⁻¹ of diammonium phosphate (DAP) and 25 kg.ha⁻¹ of urea initially with intake water and subsequently at monthly intervals.

San Francisco Bay strain (SFB) cysts of *Artemia* used for inoculation were hatched under laboratory conditions in normal seawater according to Sorgeloos *et al.*¹. Freshly hatched Instar I nauplii were introduced into the culture pond at a density of 10 nauplii.l⁻¹.

For *Artemia* population studies, quantitative sampling was done on the following day of inoculation and thereafter at 5 d interval for a period of 35 d. Sampling was carried out between 0800-1000 hrs to avoid patchiness in the distribution of *Artemia* due to wind action. Ten samples of 1 litre each were taken from different parts of the pond so as to minimise heterogeneity of the samples. They were pooled and preserved in 5% formaldehyde. Length measurements were made with dissecting microscope. Brood pouches were dissected out and the number of offsprings and cysts/brood were counted.

Taking advantage of its ovoviviparous mode of reproduction in lower salinities, the *Artemia* population was maintained at 100 × 10⁻³ for a period of 2 months i.e., upto November 1989, by which time the population density also increased. From December 1989 onwards, the salinity was gradually allowed to increase and at 120 × 10⁻³ *Artemia* population started to reproduce oviparously (cyst production). The cysts released by the popu-

lation started accumulating along the corners of the pond and were collected periodically using scoop net. Hatching efficiency (number of nauplii hatching out of 1 g of dry cysts) which is one of the criteria for assessing the quality of cysts was calculated². Biochemical constituents such as carbohydrate, lipid and protein were estimated following standard procedures³⁻⁵. To evaluate the nutritional quality, fatty acid analysis⁶ was carried out.

Results

The changes in temperature and salinity during different months are shown in Fig. 1. The average noon temperature recorded was $25.9 \pm 3.48^\circ\text{C}$ with the highest temperature in April (29.71°C) and the lowest 21.1°C in December. Mean salinity recorded in the pond was 122.6×10^{-3} with a maximum 153×10^{-3} in April and the minimum 109×10^{-3} in October. Between February and April a steady increase in salinity was observed.

Population density (no. 1^{-1}) of *Artemia* from the 5th to 35th day of inoculation was -16, 40, 53, 100, 160, 140, 210 respectively. Length measurements showed a steady increase reaching 8 ± 0.46 mm in 28 d. Size of the adult males ranged from 7 to 8 mm and those of females 8 to 8.5 mm. Riding couples were observed as early as 5th day of inoculation. Egg pouch in females appeared on the 7th day after inoculation and the first batch of nauplii through ovoviviparous reproduction was released on the 10th day. Percentage of egg bearing females varied from 23.33 to 92.6. The number of offspring/brood varied from 79 to 127.

Cyst production (Table 1) was high in February and March, 2-3 months after the salinity was raised to 120×10^{-3} . The total cyst production was 20.6 kg ha^{-1} . Table 2 lists cyst and naupliar characteristics as well as biochemical composition of nauplii. Mean diameter of hydrated untreated ($210 \mu\text{m}$) and hydrated decapsulated cyst ($130 \mu\text{m}$) recorded in the present study is less than the reported range⁷. Hatching efficiency (Table 2) compared well with reported values for SFB strain harvested from Mundra⁸. In the present study, the naupliar length is smaller as compared to the values reported for different geographical strains⁷. This trait is of advantage as smaller nauplii are readily preferred by predators. Biochemical constituents did not vary much from what has been reported elsewhere both for bisexual and parthenogenetic strains⁹. Fatty acids especially highly unsaturated fatty acids (HUFA) showed significantly higher values with eicosapen-

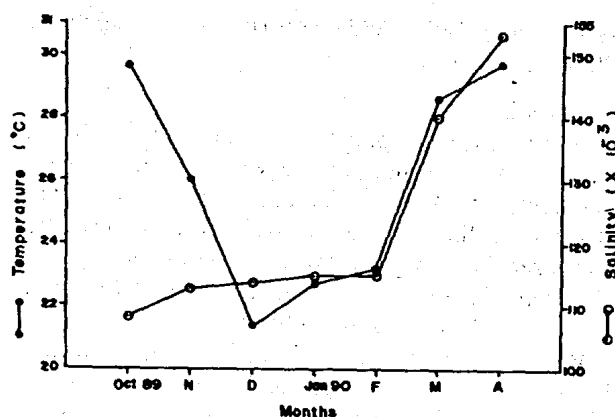


Fig. 1 - Monthly variation of surface temperature and salinity in the experimental pond

Table 1 - Production of *Artemia* cysts (kg dry wt) in the experimental pond.

December '89	0.84
January '90	0.51
February	1.22
March	1.53
April	0.43
Total	4.53
Cyst produced ha^{-1}	20.59
Cyst produced $\text{ha}^{-1} \cdot \text{month}^{-1}$	4.12

Table 2 - Cyst and naupliar characteristics of *Artemia*

Cyst diam. (μm)	
Untreated hydrated	201.11 ± 20.21
Hydrated-decapsulated	129.60 ± 07.70
Cysts (no. mg^{-1})	286 ± 24
Individual cyst dry wt (μg)	2.2 ± 0.31
Hatching efficiency	
(no. of nauplii hatched. g^{-1} cyst)	235000
Naupliar length (μm)	374 ± 29
Naupliar survival after 7 d	98.26%
Naupliar biochemical composition	
(values in % dry matter)	
Protein	56.45 ± 1.46
Lipid	25.27 ± 1.57
Carbohydrate	7.81 ± 0.16
Ash content	10.47 ± 0.93
Naupliar fatty acid content	
($\text{mg} \cdot \text{g}^{-1}$ dry wt)	
20:5W3	20.5
Total W3 HUFA	27.8

tanoic acid (20:5W3) contributing as high as 20.5 mg.g⁻¹ dry wt.

Discussion

About 300 geographical strains of *Artemia* were known throughout the world¹⁰ with several occurring in India¹¹, including parthenogenetic strain from Singach saltern itself. Nevertheless the San Francisco Bay (SFB) strain supplied by the *Artemia* Reference Centre, Belgium (Ref. No. 1090) was chosen for this culture.

Brine shrimps were alive in supersaturated brines¹² at salinity as high as 340×10^{-3} . At such high salinity, the *Artemia* barely manages to survive as the physiological and metabolic activities do not function efficiently. Though *Artemia* can tolerate salinity as low as 45×10^{-3} , it prefers a salinity where there are no predators. Hedgpeth¹³ mentioned that for several species of marine fish and invertebrates the upper salinity tolerance levels can be as high as $80-100 \times 10^{-3}$. Hence, inoculation of the pond with freshly-hatched nauplii was carried out at sal. $> 100 \times 10^{-3}$.

Under laboratory conditions SFB strain of *Artemia* can become adult in 2 week's time¹⁴. Great Salt Lake (GSL) and Macau strains of *Artemia* have been reported to have attained sexual maturity within 13-14 d and the Chinese strain¹⁵ in 19 d. In the salt pans in Thailand, riding couples were observed within 9 d of inoculation of SFB strain. In the present study riding couples were noticed on the 5th day after inoculation, thereby indicating early maturity. Fertilisation of pond at regular intervals ensured food availability for the *Artemia* population. Availability of quality food, appropriate salinity and water temperature are known to affect the growth and reproduction of *Artemia*¹⁷. It is likely that these factors have probably contributed for good growth in the present study.

The number of offsprings per brood varied from 79-127. Compared to the number of offsprings produced by Macau, GSL and Chinese strains¹⁸ the number recorded in the present study is higher.

Salinity increased from December-April, when cyst production also increased upto March and declined in April. Reduction in cyst production in April probably be attributed to high salinity. This has been found to be true by Berthelemy-Okazaki and Hedgecock¹⁹ who have shown that salinity above 120×10^{-3} inhibits cyst production.

Cyst production data from different countries is given in Table 3. Best production has been recorded from Philippines²⁰ followed by some of the sites in Thailand²¹ and India⁸. (Mundra). Ear-

Table 3 - Production of *Artemia* cysts from different countries

Country/Reference	Area (ha)	Harvest (kg.month ⁻¹)
1. India ⁸ Mundra	0.3	12.40
2. Philippines ²⁰ Negros Oriental (Commercial salt pan)	20.00	20.00
3. Thailand ²¹ Chonburi, Samut Sakorn	3.2 0.3	15.40 05.67
Phetburi	1.4	3.4
Chachoeng-Sao	1.4	17.50
Samut Songkram	1.0	15.30
4. Vietnam Cam Ranh Bay, Pond 1		6.8
Pond 2		2.2
Pond 3	0.031	2.6
Pond 4		1.9
Pond 5		0.6
5. India, Singach (Present work)	0.22	4.12

lier²² a cyst production of 17 kg.ha⁻¹ was estimated from a naturally occurring population of *Artemia* in Singach which is very close to the realised yield from culture population.

The quality of the *Artemia* cysts is assessed on the basis of the presence of highly unsaturated fatty acids (HUFA) especially the eicosapentaenoic acid (20:5W3) and decosahexaenoic acid (22:6W3) in substantial quantities. The analysis of the I instar nauplii in the present study showed a high level of 20:5W3 and in general a good percentage of total HUFA. Hence, this culture strain is highly nutritive as a live feed for hatcheries.

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