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Effect of salinity on the life span and reproductive characteristics of brine shrimps in the salt pans at Tuticorin

N. Soniraj

Tuticorin Research Centre of CMFRI, Karapadu, Tuticorin,Tamil Nadu, India
E-mail: *Soni_raj2000@yahoo.com*

Abstract

Effect of salinities in the range of 20 - 170 ppt on reproductive and lifespan characteristics of *Artemia franciscana* Kellog, a non endemic sexually reproducing species and *A. parthenogenetica* Bowen, the endemic asexual species, from salt pans of Tuticorin were studied at ambient temperature with *Chlorella* as feed. *A.franciscana* showed significantly faster rate of maturation and longer 'reproductive period' compared to the local species at all the tested conditions. At higher as well as lower salinities the pre-reproductive period of both the species were prolonged. The values per female of the 'broods', 'total offspring' and 'nauplii' in *A.franciscana* were significantly higher especially between 20 - 100 ppt. The 'brood size' of sexual and asexual species varied with salinity, the former showed higher values at 20-100 ppt and the latter at 120-170 ppt. Only sexual species produced significant cysted offspring which showed an increasing trend with higher salinities (120,145 and 170 ppt) while both the species produced maximum nauplii at lower ranges (20, 45, 80 and 100 ppt). The results presented may generate useful management tips to those who take up *Artemia* culture in saltpans of Tuticorin.

Key words: Effect of salinity on lifespan, brine shrimps

Introduction

Variation in habitat ecological conditions can have major effect on life history and reproductive characteristics of *Artemia* species occurring in different geographical areas (Browne *et al.*, 1988). There are reports of co-occurrence of both sexual and asexual population in some Spanish salina (Amat, 1980, 1982) where one population dominates the other in accordance with the climatic changes. In India, at Tuticorin, presence of both the native asexual species *A. parthenogenetica* and the exotic sexual form *A. franciscana* have been recorded from the same ecosystem (Rajamani *et al.*, 1998). The latter has been reported to have contaminated the native parthenogenetic population in the salt pans at Tuticorin from the culture ponds, where earlier attempts of commercial culture with this species were done by some industrialists. The lifespan, reproductive characters and the encystment rate were studied to find out how these aspects vary in relation to different salinity stress. Such aspects on various *Artemia* spp. have been studied earlier by many workers at temperate conditions (Triantaphyllidis *et al.*,

1995; Browne *et al.*, 1984; Wear *et al.*, 1986; Wear and Haslett, 1986). As information on the reproductive characteristics of *Artemia* species is not available from the tropical environment a detailed investigation was made.

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Material and methods

The matured females of *A. parthenogenetica* and the exotic sexual form *A. franciscana* were collected from the saltpans of Tuticorin and maintained at laboratory in filtered brine from saltpans as stock culture. The first generation nauplii (F1 generation) released by these females were collected and then transferred to 20, 45, 80, 100, 120,145 and 170 ppt for studying the effect of these salinity levels on the life span and reproductive characteristics. Required salinities were made by diluting concentrated filtered brine collected from salt pans. Cultures of *Chlorella* sp. maintained in algal laboratory were used as feed. An average algal concentration of 1.5 million cells/ml of culture was maintained at all the experimental units. *Artemia* rearing jars were maintained under natural lighting at ambient temperature. Accumulated faecal matter and algal deposits were removed every day. Culture medium was replaced every 5th day. Separate sets of twenty nauplii each from wild broods were reared in triplicates in one litre transparent plastic jars at the required salinity medium. After attaining maturity, three adults from each nauplii rearing jars were transferred separately to three 500 ml jars containing 300ml brine. For asexual species, one female was placed in a jar and for sexual species one female and male were kept in a single jar. Every day jars were examined for nauplii, cyst or dead animal. Dead males were replaced with fresh ones from the stock culture.

Entire life span of the female was divided into three phases viz. pre-reproductive, reproductive and post-reproductive. Pre-reproductive period is the average days taken by 50% of nauplii in 1st instar to mature and release the 1st batch of their nauplii/cyst. Reproductive period is in days from the release of 1st brood to the last brood. Post- reproductive period represents the number of days from the last brood to the death of animal. 'Total female life span' was counted as total number of days taken by nauplii to mature from 1st instar to reproduce and die. The 'broods per female' was counted as the number of broods obtained from each female in its entire life span and the 'interbrood period' as the average days taken by female to mature and release offspring from one brood to next. 'Offspring per brood' (brood size) was measured as total number of nauplii / cysts obtained from single brood and 'total offspring per female' as total offspring produced by individual female in its reproductive cycle (Browne *et al.*, 1984,1988).

Readings were taken from individuals maintained at salinities of 20, 45, 80, 100, 120, 145 and 170 ppt. As females of both species did not attain maturity at 170 ppt, reproductive characteristics at this salinity were studied using mature specimens collected from the culture maintained at 145 ppt. Total number of animals used for pre-reproductive period were 60 (20 nauplii in 3 replicates each) for each experiment. As three matured animals from each replicate were reared till death, rest of the characters were studied from 9 females from each set. Results were analysed by a single factor ANOVA using the statistical software Systat 7.0. For all the statistical analysis values for % were normalized by arcsine transformation.

Results and discussion

Effect of salinity on life span components

The results of the present investigation carried out in a tropical environment clearly indicated that a salinity range of 80 - 100 ppt is most favourable for maturation. At 170 ppt females of both the species died even before maturation. Maturation was faster for sexual species at 20, 45, 80 and 100 ppt salinities compared to 120 and 145ppt for the asexual form. There was a significant increase in the maturation period as the salinity deviated from 80 and 100 ppt on either side for both the species, while within the range there was no significant differences (Table 1). According to Browne and Hoopes (1930), in a temperate climatic region total mortality occurred only at 230 ppt contradicting the results of the present investigation which may be due to the high temperature prevailing during most part of the year at Tuticorin region. Rearing *Artemia* at salinities above 200 ppt was found to be difficult as reported by Wear *et al.* (1986) and Dana and Lenz (1986). According to Triantaphyllidis *et al.* (1995) maturation period of *A. franciscana* strain and a parthenogenetic strain from China were found to be longer at higher salinities and maturation rate was found inversely proportional to salinity especially above 140 ppt thus agreeing with the observations in the present investigation.

In the present study, salinities between 45 and 120 ppt were found to be favourable for the asexual species and 20 and 100 ppt for the sexual form for longer reproductive life. The shortest pre- reproductive period also was at these salinities (Figs.1&2).

Reproductive period of both the species was found to decline with an increase

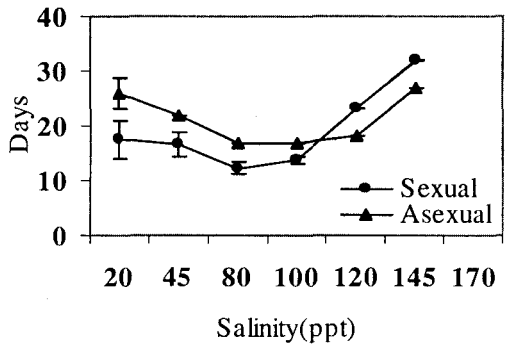

Fig. 1. *'Pre-reproductive period' of sexual and asexual species of* Artemia *at different salinities*

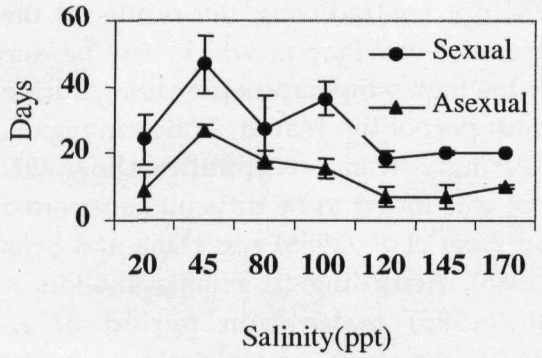

Fig. 2. *Reproductive period of sexual and asexual species of* Artemia *at different salinities*

in salinity which differed significantly between the species, with sexual species having longer days at all the tested salinities (Fig. 2). In both the species reproductive period was relatively longer in the lower salinity ranges (20, 45, 80 and 100 ppt). Total life span' was also reported to be longer for *A.sanfranciscana* (Fig.3). The sanfrancisca sexual strain generally showed a longer reproductive period than the parthenogenetic strain from China (Triantaphyllidis *et al.*, 1995). Post-reproductive period of both the species in the

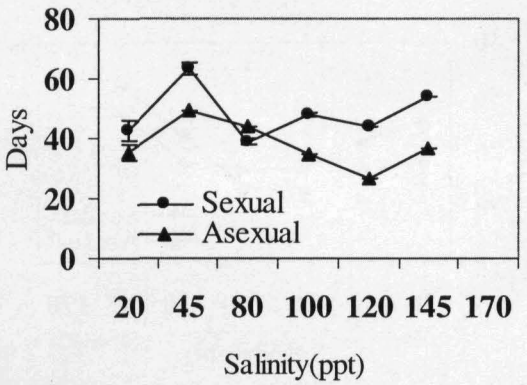

Fig. 3. *'Total lifespan' of sexual and asexual species of* Artemia *at different salinities*

present study was found not significantly varying with the change in salinity as observed by Browne *et al.* (1984). Maximum 'life span' was at 45 ppt for both the species, 63.3 ±8.3 and 49.5 ±2.1 days respectively (Fig. 3). 'Reproductive period' constituted a major part (>50%) of the lifespan for sexual species at 20, 45, 80 and 100 ppt salinities but for the other species it was only at 45 ppt (Table 2). For both the species lowest 'percentage of reproductive period' was at higher salinity (145 ppt). It was highest (76%) for sexual at 100 ppt and 54.7% for asexual species at 45 ppt.

Effect of salinity on the reproductive traits

The reproductive output of *Artemia* species is controlled mainly by the differences in reproductive characteristics like 'brood numbers', ' interbrood period', 'offspring production' and '% encystment'. At salinities 20 to 100 ppt sexual species had significantly better 'brood number', 'total offspring per female' and 'nauplii per female' and at 120 ppt and above there was no significant difference between the two (Figs. 4, 6 & Table 2). For both the species 'brood size' was better at salinities 20,45,80 and 100 ppt (Fig. 5). Above 100 ppt there was no statistically significant variation in the offspring production between the species as well as among the salinities (Table 1). At salinities 20, 45, 80, 100 and 120 ppt the sexual form produced significantly more offspring than the asexual at 145 and 170 ppt (Fig.5). Knight (1974) reported that in

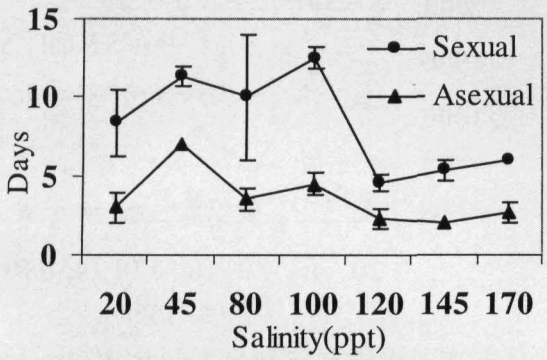

Fig. 4. *'Broods per female' of sexual and asexual species of* Artemia *at different salinities*

the field, with little variation upto 215ppt salinity, *A. franciscana* was found to carry 45 to 55 eggs per females. The number steeply reduced to 20 eggs when salinity increased to 282 ppt. Dominant influence of salinity on fecundity was also reported by Wear *et al.* (1986). The 'reproductive period' showed significant difference between two species. The reproductive characteristics like, 'total offspring per female', 'brood number' and 'nauplii per female' showed significantly higher values for sexual form only between 20 to 100 ppt.

The reproductive characters of parthenogenetica population from China and sexual strain from San Francisco Bay were significantly different at 35, 60 and 100 ppt (Triantaphyllidis *et al.*,1995). Above 140 ppt the reproductive out put showed reduction. This was found almost

Table 1. *Results of ANOVA indicating significant differences (P<0.05) for Life span and reproductive characteristics of* A.franciscana (A.f) *and* A.parthenogenetica (A.p) *from Tuticorin salt pans reared at different salinities. Species that share the same alphabetic letters (a,b,c....k) for each characteristics are not significantly different. A.: Pre-reproductive period, B: Reproductive period, C: Post-reproductive period, D: Total life span, F: Broods per female, G: Inter brood period, H: Total offspring per female, I: Offspring per brood and K : Nauplii per female.*

Species	‰	A	B	C	D	F	G	H	I	K
A.p	20	a	Ah	ae	ea	ah	acfg	a	edac	a
A.f	20	b	bcef	ae	abc	b	acfg	b	b	b
A.p	45	c	C	ae	ci	c	cde	c	c	c
A.f	45	d	D	ae	d	df	acf	d	b	d
A.p	80	bde	Ei	b	abc	aeik	acd	a	eda	a
A.f	80	f	Cf	ae	abe	bd	f	c	edc	c
A.p	100	bde	Aij	ce	abe	aeg	acf	a	gec	a
A.f	100	f	G	ace	ci	f	gf	b	fc	b
A.p	120	be	A	ac	f	ae	d	a	ga	a
A.f	120	c	Bej	e	g	aej	ed	a	a	a
A.p	145	a	Ha	ace	e	hi	acf	a	eda	a
A.f	145	g	Bej	e	i	ce	acd	a	eda	a
A.p	170		A	e	be	ahk	acd	a	a	a
A.f	170		Bej	d	hg	cgj	acf	a	a	a

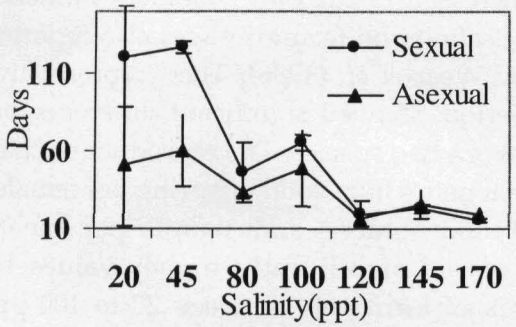

Fig. 5. *'Offspring per brood' of sexual and asexual species of* Artemia *at different salinities*

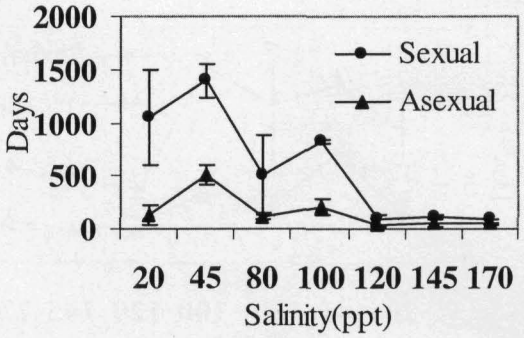

Fig. 6. 'Total offspring per female' of *sexual and asexual species of* Artemia at different salinities

true in the tropical conditions in the present study. The 'broods per female', 'offspring per brood' and 'total offspring per female'showed higher values at low salinities (<100 ppt) and *vice versa*. In general, response of these characteristics in the two species with different salinities was similar (Figs.2, 4, 5 and 6).

With increase of salinity from 120 ppt the nauplii production decreased in the case of sexual species while the encystment showed an increase. Asexual species could produce cysts only at 100 and 170 ppt, but in few numbers (Table 2). Increase in salinity brought by evaporation was known to induce adult *Artemia*

Table 2 *Mean and s.d. of reproductive and life span characteristics for the* A. franciscana *(A.f) and* A. parthenogenetica *(A.p) species reared at different salinities Sps: Species, A: Post reproductive period, B: Percentage reproductive period to life span, C: Inter brood period, D: Cysts per female, E : Nauplii per female and F: Percentage encystment.*

Species	Salinity (ppt)	A (days)	B (%)	C (days)	D number	E (number)	F (%)
A.p	20	0.67±1.2	23.2	3.9	-	126.66+94.1	-
A.f	20	0.67±0.6	62.7	3.3±0.1	-	1041.67+450.7	-
A.p	45	0.5±0.7	54.6	4.3	-	501.5+98.3	-
A.f	45	1.0	72.8	3.93±0.6	0.6	1395.0+161.1	0.53
A.p	80	9.0±4.2	40.9	4.5±0.7	-	501.5+98.3	-
A.f	80	0.3±0.5	70.0	2.88±0.1	178.67+91.5	329.67+280.0	41.29+13.2
A.p	100	3.0±1.0	42.8	3.88±0.1	9+15.6	198+94.7	5.67
A.f	100	1.5±2.1	76.0	2.95±0.1	51.5+50.2	820.0+12.7	6.24+6
A.p	120	1.3±2.3	27.4	4.71±0.2	-	33+10.6	-
A.f	120	2.67±1.5	41.6	5.28±1.0	20.67+27.6	68.33+22.6	16.8+18.2
A.p	145	2.67±1.5	16.2	4.5±0.7	-	53.67+40.2	-
A.f	145	2.33±1.5	35.8	4.42±0.3	68.66+22.3	43.33+3.2	59.9+11
A.p	170	2.67±2.1	-	4.5±0.5	13+22.5	36.33+14.6	4.2
A.f	170	5.33±0.6	*	3.87±0.1	82.67+17.1	12.33+10.8	89.65+11.7

for oviparous reproduction (Sorgeloos *et al.*, 1975; Amat, 1983). In the San Francisco Bay strain, cyst production was found most favourable at 35,140 and 180 ppt (Triantaphyllidis *et al.*, 1995).

Competitive ability between the *Artemia* population is highly related to their life history traits (Browne, 1980a; Browne *et al.*, 1984). Pre-reproductive period that can also be called as 'generation time' is a key factor in determining the population growth rate and subsequent competitive exclusion among the population in a resource limited environment (Cole, 1954). In many species the competitive exclusion requires in the order of 10 to 100 generation, but for *Artemia* where the generation time is approximately 20 days the exclusion occurs in 2 to 3 generation (Carpelan, 1957). According to Browne and Halanych (1989), sexual population from Old World were always out competed by parthenogenetic species indicating that the outcome of sexual and parthenogenetic competition are linked to whether sexual population is of New or Old World origin. At Tuticorin, sexual strains are reported to have come from Sanfrancisca population (New World population) (Rajamani *et al.*, 1988). In the present study in almost all the experimental conditions, especially below 100 ppt salinity, sexual species had the dominance with respect to their reproductive characteristics over the asexual parthenogenetic one. Sexual females also had lesser generation time and longer reproductive days indicating its possible dominance over *A.parthenogenetica*.

There are certain factors, which are also to be considered in a mixed population. The lifespan of the male is an important factor in determining the growth of sexual population and is comparatively short as reported by Browne (1982). In a mixed population there is a possibility of sperm robbing (Browne, 1980b; Lynch, 1984) which may eliminate sexual species while the parthenogenetic females can reproduce in the absence of male. Hence a complete exclusion of native asexual population by sexual exotic population may not occur. But there is every possibility that the purity of the native population will be lost. This is not due to the genotypic variations of the local strain, which happens rarely in *Artemia*. The mixing of the cysts produced by both the populations would affect its marketability as the hatchability and the required hatching conditions of both the species vary greatly. The present results are encouraging as cyst production in large salt pans could be controlled by salinity manipulations. The results indicate that sexual species can be tried at salinas of Tuticorin with salinities higher than 120 ppt to have a good cyst harvest.

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