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EFFECTS OF TEMPERATURE AND SALINITY ON THE DECAPSULATION OF *ARTEMIA* CYST

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Abstract: A study was performed to determine to effect of salinities (20, 25, 28, 30, 32, 35 and 40 ppt) and temperatures (20, 24, 28, 30 and 32°C) on the decapsulation of *Artemia* cyst. The dry *Artemia* cyst were hydrated up to 2 hrs in fresh water at (26 ± 1ºC) temperature with a density 0.3 g/ 50 ml water. Saline water was prepared using salt (NaCl) and hatching rate of *Artemia* cyst was counted after 36 hours of incubation. Highest hatching rate (72.59 ± 5.03), (75.84 ± 3.67), (74.45 ± 0.83), (89.38 ± 0.96), (76.46 ± 4.63), (70.58 ± 9.51) and (69.17 ± 0.70)% were observed in 20, 25, 28, 30, 32, 35 and 40 ppt salinities at 32, 28, 28, 24, 24, 24, and 28°C temperature, respectively. Lowest hatching rate (57.03 ± 3.67), (47.97 ± 8.06), (46.20 ± 8.17), (39.58 ± 10.12), (47.36 ± 7.04) (25.86 ± 8.10) and (20.57 ± 2.71) % were measured in 20, 25, 28, 30, 32, 35 and 40 ppt salinities at 30, 32, 32, 32, 32, 32 and 32°C, respectively. The maximum hatching rate (89.38 ± 0.96)% was found in 30 ppt salinity at 24°C temperature and minimum hatching rate (20.57 ± 2.71) % was observed in 40 ppt salinity at 32°C temperature. Therefore, 30 ppt salinity and 24ºC temperature could be the optimum salinity-temperature for *Artemia* cyst decapsulation.

Key words: Temperature, salinity, decapsulation, *Artemia*

INTRODUCTION

Artemia is a crustacean live food item distributed all over the world including tropical, subtropical and temperate zones. It is widely used as live food for larvae of finfish and shellfish especially *Macrobrachium rosenbergii* (De Man 1879) and *Penaeus monodon* (Fabricius 1798) (Lavens and Sorgeloos 1996, Triantaphyllidis *et al.* 1998, Lavens *et al*. 1986). Every year more than 2000 metric ton *Artemia* cyst are marketed all over the world for aquaculture production purposes. The demand of *Artemia* cyst is increasing day by day in aquaculture industry and it comprises almost 40% of total larval food in aquaculture production (Sorgeloos *et al.* 2001).

In Bangladesh, *Artemia* cyst are used in a large scale specially in shrimp and prawn hatchery due to its higher nutritional value and ease of uses (Naser *et al.* 2016). Generally *Artemia* cyst are imported from different countries like USA, China, Brazil etc. and cyst are decpsulated in the hatchery. The decapsulation rate of *Artemia* cyst greatly depends on the salinity, temperature, aeration and

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pH of water (Ahmed *et al.* 1997, Lavens and Sorgeloos 1996). A small number of works has been conducted to observe the effect of salinity and temperature on the decapsulation of cyst (Ahmed *et al.* 1997, Sharahi and Zarei 2016). But, there was lack of research work of precise temperature and salinity or their combined effect on cyst decapsulation. That is why, an experiment was conducted to find out the influence of salinity and temperature on the decapsulation of cyst.

MATERIAL AND METHODS

Artemia cyst was purchased from the local market shops at Kataban, Nilkhet, Dhaka, Bangladesh. The origin of the cyst was GSL (Great Salt Lake), USA. *Artemia* cyst is protected by a hard shell that encysts the dormant *Artemia* embryo. This *Artemia* embryo was completely removed from the cyst by a short term incubation in saline solution with aeration. This procedure is called decapsulation of cyst (Lavens and Sorgeloos 1996). Total three stages involved in decapsulation procedures.

The first step of decapsulation procedure was hydration of *Artemia* cyst. Hydration of cysts allowed for separation of the nauplii from the chorion, facilitating the decapsulation process. For this purpose, *Artemia* cysts were kept in a beaker with fresh water at room temperature for approximately two hour, using a concentration of 0.25 g of cysts per 50 ml of water (Sorgeloos and Skujlasekarapandian 1984). The hydration procedure was maintained at 26 ± 1°C temperature. The second step of decapsulation was removal of live embryo in saline solution from its brown shell. For this purpose, decapsulation solution was prepared using 500 ml of water in a beaker with iodine free salt. The amount of salt was calculated using following formula:

$$\text{The amount of salt needed (g)} = \frac{\text{Desired salinity (ppt)} \times 500}{1000}$$

Seven types of decapsulation salinity solution were prepared (e.g. 20, 25, 28, 30, 32, 35 and 40 ppt). Three replicates were maintained for each treatment. Decapsulation was performed at 20, 24, 28, 30 and 32°C temperatures. Temperature and salinity of water were checked using a Handheld Salinity, Conductivity & Temperature meter (YIS 30, USA).

During the decapsulation of *Artemia* cyst, continuous aeration was offered in each beaker for proper hatching of embryo. For this porpose, aerator (SB-348A air pump) was used. pH of decapsulation solution was maintained at 8.0 to 8.5.

Hatching rate of the decapsulated cyst was observed after 36 hours of aeration. Hatching rate was calculated using following formula (Sorgeloos and Skujlasekarapandian 1984):

$$\text{Hatching rate (\%)} = \frac{\text{Number of hatched embryo} \times 100}{\text{No. of hatched embryo / No. of unhatched embryo}}$$

The hatching rate data obtained from the study were analyzed statistically using statistical software (SPSS, version 16.0, SPSS Inc., Chicago, USA). One way ANOVA and multiple camparison test (Tukey HSD post hoc test) was used to compare different hatching rate at different temperatures. All data were presented with mean ± standard error (SE).

RESULTS AND DISCUSSION

The effect of temperature on decapsulation of *Artemia* cyst at 20 ppt salinity is presented in Fig. 1. Highest hatching rate (72.59 ± 5.03%) was observed in 32ºC temperature at 20 ppt salinity. Lowest hatching rate (57.03 ± 3.67%) was found in 30°C temperature at 20 ppt salinity. But there is no significance difference ($p > 0.05$) among 20, 24, 28, 30 and 32°C temperature.

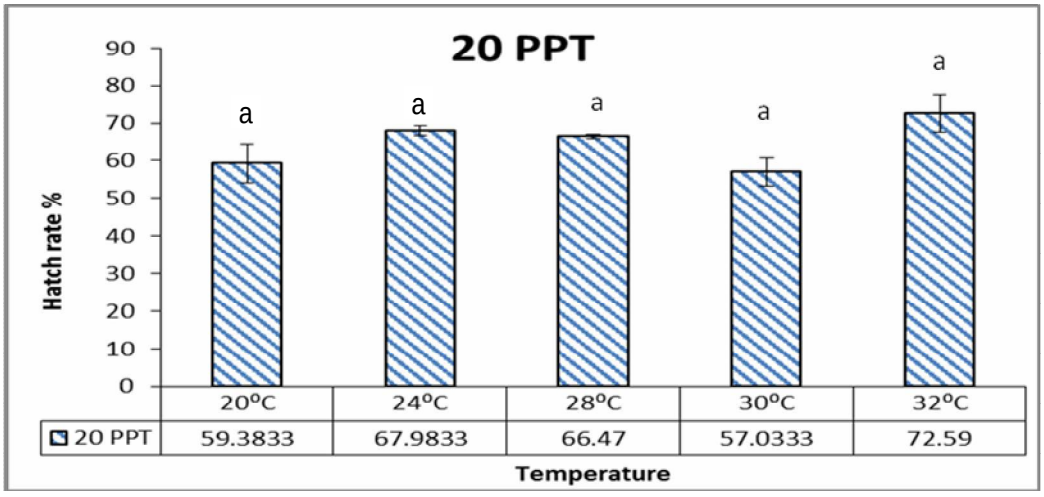

Fig. 1. Effect of temperature on decapsulation of *Artemia* cyst at 20 ppt salinity. Values are mean ± SEM. Bar with similar letter designated no significantly difference ($p > 0.05$, analyzed by one way ANOVA and Tukey´s test).

Ahmed *et al.* (1997) performed an experiment on the effect of decapsulation on viability and hatching of *Artemia* cysts at different salinities. They found highest hatching rate (75.7 ± 2.5 %) and lowest (70.0 ± 1.7 %) at 20 ppt

salinity after 36 hrs of incubation. So the highest hatching rate is similar to this study but lowest hatching rate could be due to the low temperature (20°C).

Significantly highest hatching rate was found 75.84 ± 3.67% in 28°C temperature and lowest hatching rate 47.97 ± 8.06% in 32°C temperature at 25 ppt when compare with among 20, 24, 28, 30 and 32°C temperature (Fig. 2). There was significant difference among 24, 28 and 32°C and no significant differences were observed among 20, 30 and 32°C.

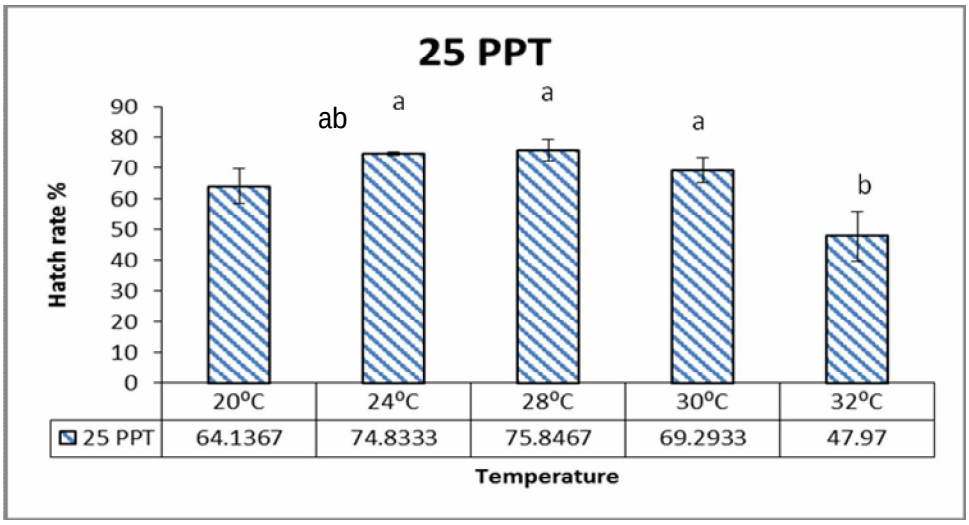

Fig. 2. Effect of temperature on decapsulation of *Artemia* cyst at 25 ppt salinity. Data as mean ± sem. Bar with different letters represent significantly difference ($p < 0.05$, analyzed by one way ANOVA followed by Tukey´s test).

Rajkumar and Babu (2015) recorded 72% hatching rate at 25 ppt salinity. They also observed different hatching rate at different temperature. Highest hatching rate (88%) was obtained at 29°C and lowest hatching rate (28 %) at 20°C temperature.

Maximum hatching rate 74.45 ± 0.83% was found in 28°C temperature at 28 ppt salinity. Lowest hatching rate 46.21 ± 8.18% in 32°C temperature at 28 ppt salinity (Fig. 3). There was no significant difference among 20, 24, 28 and 30°C ($p > 0.05$).

Highest hatching rate 89.38 ± 0.96% was observed in 24°C temperature at 30 ppt salinity and lowest hatching rate 39.58 ± 10.12% was found at 32°C temperature in 30 ppt salinity (Fig. 4). Significantly lowest hatching rate was found in 32°C when compared with 20, 24 and 28°C. There was no significant difference among 20, 24, 28 and 30°C ($p > 0.05$).

An experiment was conducted by Rajkumar and Babu (2015) on the influence of salinity and temperature on the growth of *Artemia.* They observed 84 and 87% hatching rate of *Artemia* cyst at 28 and 30 ppt salinities respectively. But higher hatching rate (89.38 ± 0.96%) was found in this study at 24°C temperature. Wansonga and Olendi (2017) found 84.11% hatching rate at 28 ppt salinity in 28°C temperature. Highest (58.7 ± 1.5 %) and lowest (45.7±3.5%) hatching rate were calculated at 30 ppt salinity after 36 hours of incubation (Ahmed *et al.* 1997).

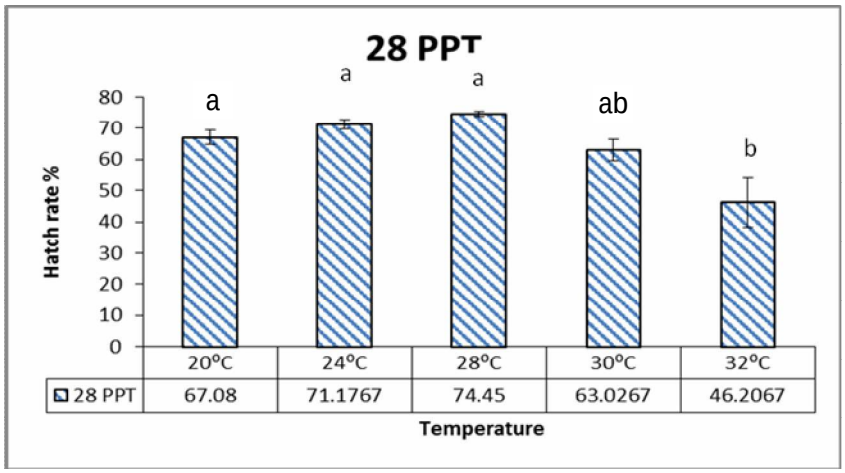

Fig. 3. Effect of temperature on decapsulation of *Artemia* cyst at 28 ppt salinity. Data are recorded as mean ± SEM. Bar with dissimilar letters designated significantly difference ($p < 0.05$, analyzed by one way ANOVA and Tukey´s test).

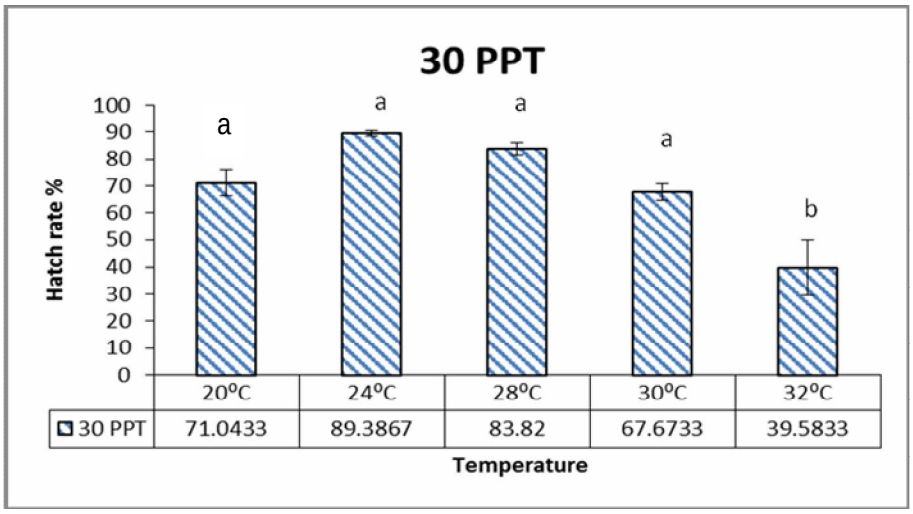

Fig. 4. Effect of temperature on decapsulation of *Artemia* cyst at 30 ppt salinity. Data are recorded as mean ± SEM. Bar with different letters specified significantly difference ($p < 0.05$, analyzed by one way ANOVA and Tukey´s test).

Significantly highest hatching rate 76.46 ± 4.63% was found in 24°C temperature at 32 ppt salinity and lowest hatching rate 47.36 ± 7.04% was found in 32°C temperature at 32 ppt salinity (Fig. 5). There was no significant difference among 20, 24, 28 and 30°C ($p > 0.05$).

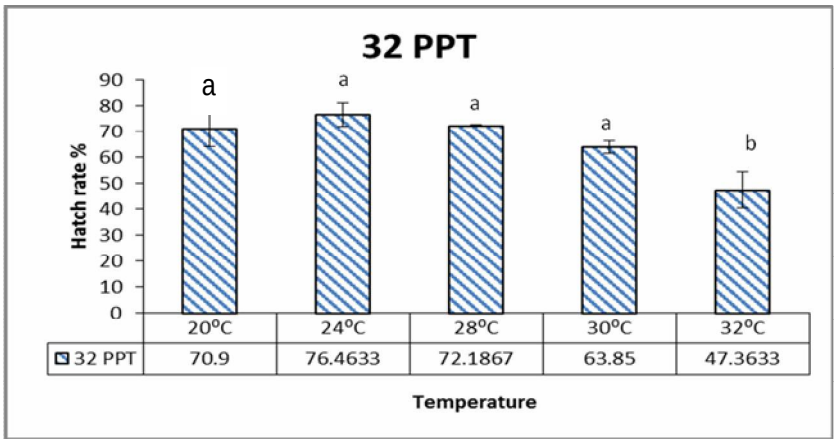

Fig. 5. Effect of temperature on decapsulation of *Artemia* cyst at 32 ppt salinity. Data are recorded as mean ±SEM. Bar with different letter show significantly difference ($p < 0.05$, analyzed by one way ANOVA and Tukey´s test).

Here, maximum hatching rate 70.58±9.5% was observed in 24°C temperature at 35 ppt salinity and lowest hatching rate 25.86 ± 8.10% was found in 32°C temperature at 20 ppt salinity. Significantly lowest hatching rate was found in 32°C at 35 ppt (Fig. 6). There was no significant difference among 20, 24, 28 and 30°C ($p > 0.05$).

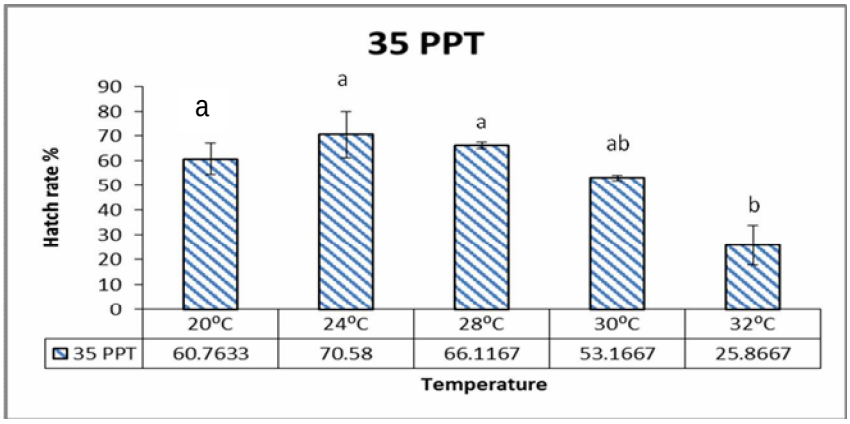

Fig. 6. Effect of temperature on decapsulation of *Artemia* cyst at 35 ppt salinity. Data are recorded as mean ± SEM. Bar with diverse letters directed significantly difference ($p < 0.05$, analyzed by one way ANOVA and Tukey´s test).

Significantly maximum hatching rate 69.17 ± 0.70% was found in 28°C temperature at 40 ppt and lowest hatching rate 20.57 ± 2.71% was found in 32°C temperature at 40 ppt salinity (Fig. 7). There was no significant difference between 30 and 32°C and among 20, 24, 28 and 32°C ($p < 0.05$). But significantly highest hatching rate was observed in 20, 24, 28 compared with 30 and 32°C at 40 ppt.

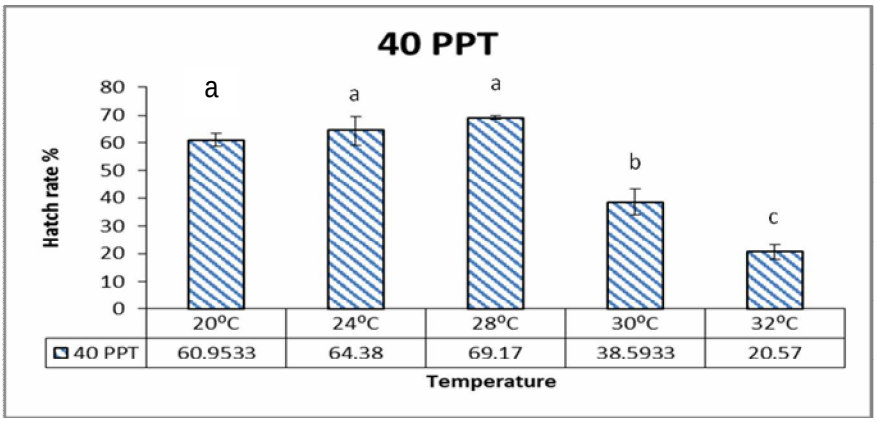

Fig. 7. Effect of temperature on decapsulation of *Artemia* cyst at 40 ppt salinity. Data are recorded as mean ± sem. Bar with different letters pointed out significantly difference ($p < 0.05$, analyzed by one way ANOVA and Tukey´s Test).

Sorgeloos and Skujlasekarapandian (1984) suggested 35 ppt salinity of sea water is ideal for *Artemia* cyst decapsulation in laboratory condition. But Rajkumar and Babu (2015) found 75 and 58% hatching rate at 32 and 35 ppt salinity respectively. Ahmed *et al.* (1997) recorded maximum (47.7 ± 2.1%) hatching rate and minimun (40.3 ± 1.2%) at 40 ppt salinity for processed and preserved cyst whereas Wansonga and Olendi (2017) observed 89.88% hatching rate at 40 ppt salinity. But this result did not match properly with the previous literature.

Generally, *Artemia* cyst decapsulation depends on many factors like temperature, salinity, pH, cyst quality, aeration etc. Moreover, fixed temperature, cyst density and container's design also play important role in maximal production of *Artemia* cyst (Lavens and Sorgeloos 1996, Sharahi and Zarei 2016). The variation of *Artemia* decapsulation rate in different literature could be the effect of different above mentioned factors.

It can be concluded from the above discussion and the findings of this study that the decapsulation rate of *Artemia* cyst greatly depends on salinity and temperature. This result, 30 ppt salinity and 24°C temperature could be the best

combination of higher rate of decapsulation of *Artemia* cyst. Moreover, decapsulation rate decreases above 35 ppt salinity and 30°C temperature.

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