

## EFISIENSI DAN ENERGETIKA PENETASAN KISTA ARTEMIA (*Artemia salina*) PADA SALINITAS MEDIA YANG BERBEDA

### EFFICIENCY AND ENERGETICS OF ARTEMIA (*Artemia salina*) CYSTS HATCHING IN DIFFERENT OSMOLARITY MEDIA

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#### Abstrak

Tingkat penetasan kista *Artemia salina* dalam proses penetasan bervariasi karena banyak faktor. Osmolaritas medium merupakan salah satu faktor yang menentukan proses penetasan kista *A. salina*. Tujuan dari penelitian ini adalah untuk menilai proses penetasan kista *A. salina* dalam berbagai kondisi osmotik pada efisiensi dan laju penetasan energetika. Desain eksperimen yang digunakan adalah rancangan acak lengkap 2 x 5 faktorial. Faktor pertama adalah penambahan klorin (non-dekapsulasi dan dekapsulasi) dan faktor kedua adalah osmolaritas medium (640,27; 787,02; 901,76; 1080,51 dan 1227,25 mOsm.L<sup>-1</sup>H<sub>2</sub>O). Hasil penelitian menunjukkan bahwa penetasan kista *A. salina* pada berbagai kondisi osmotik secara signifikan berbeda dengan tingkat penetasan. Tidak ada perbedaan yang signifikan dalam efisiensi energetika penetasan yang diperoleh dari masing-masing kelompok eksperimen. Media dengan tingkat osmolaritas iso-osmotik menjadi *hyperosmotic* (901,76-1227,25 mOsm.L<sup>-1</sup>H<sub>2</sub>O) memberikan tingkat penetasan yang tinggi. Energetika efisiensi penetasan kista *A. salina* berkisar antara 640,27 sampai 1227,25 mOsm.L<sup>-1</sup>H<sub>2</sub>O pada media *hyposmotic*, *isosmotic* dan *hyperosmotic*.

**Kata kunci:** artemia, dekapsulasi, energetika, penetasan, salinitas

#### Abstract

Hatching rate of *Artemia salina* cysts in the hatching process is variable due to many factors. Osmolarity of the medium is one of the factors determining hatching process of *A. salina* cysts. The purpose of this study is to assess the hatching process of *A. salina* cysts in a various osmotic condition on the hatching energetics efficiency and rate. The experimental design used was a 2 x 5 factorial completely randomized design. The first factor is an addition of chlorine (non-decapsulation and decapsulation) and the second factor is osmolarity of medium (640.27; 787.02; 901.76; 1080.51 and 1227.25 mOsm.L<sup>-1</sup>H<sub>2</sub>O). The results showed that hatching of *A. salina* cyst on various osmotic condition has significantly differed the hatching rate. No significant differences in the hatching energetics efficiency were obtained from each experimental groups. Media with osmolarity level of iso-osmotic to hyperosmotic (901.76-1227.25 mOsm.L<sup>-1</sup>H<sub>2</sub>O) provides a high hatching rate. Energetics efficiency of artemia cysts hatching is ranged from 640.27 to 1227.25 mOsm.L<sup>-1</sup>H<sub>2</sub>O in both hyposmotic, isosmotic and hyperosmotic media.

**Keywords:** artemia, decapsulation, energetics, hatching, salinity

#### Introduction

*Artemia salina* is one of the natural food that has high nutrient content and most widely used in shrimp aquaculture, particularly in a hatchery. As a natural food, *A. salina* can be used in the stadia of Nauplius and adults (Pangkey, 2011). The problem on the artemia cultivation is the limitation of the supply in the market and the low hatching rate of the cyst (60%) Low hatching rate of *A. salina* cysts is this is due to the high energy needed for hatching. The high energy of hatching associated with high energy needs for

osmoregulation process. To overcome the problem with high energy for osmoregulation, the cyst hatching process might be performed in an isosmotic condition.

Salinity is closely linked to the osmotic pressure of ions and water. Osmolarity of media as one of the factors for optimum hatching rate and energetics efficiency of *A. salina* is unknown. Therefore, this experiment was conducted in order to assess the efficiency and energetics hatching artemia cysts on various media osmolarity (Lantu, 2010).

**Material and Methods**

Research conducted at the Institute of Seed and Brackish Water Aquaculture and Marine Unit Brackish Water Fish Seed, Sluke Rembang. *A. salina* cysts are hatched at a density of 150,000 cysts/L. Media water used to hatching artemia using sea water from around Satker PIAP Sluke, Rembang. *A. salina* hatching container using gallons of mineral water with a capacity of up to 15 liters of 5 pieces.

The method used in this study is an experimental method with a laboratory scale. The experimental design used was completely randomized design with a 2x5 factorial design. The first factor is the addition of chlorine (non-decapsulation and decapsulation) and the second-factor osmolarity media (640.27; 787.02; 901.76; 1080.51 and 1227.25 mOsm.L<sup>-1</sup>H<sub>2</sub>O). The variables measured were the average level of osmotic work, hatchability cyst, hatching energetics efficiency and enzyme activity of Ca-Chorionase. This study uses ten treatment and four replications.

The rate of observed data is Osmotic Work (TKO) is calculated based on the difference in the value of artemia with the osmolarity of blood osmolarity test medium. The measurement technique and blood osmolarity media *A. salina* by using Automatic Micro Osmometer Roebing, while the employment rate osmotic value criterion according to (Anggoro & Nakamura, 1996):

$$TKO = (P_{osm\ blood} - P_{osm\ media})$$

where :

TKO = Level of Osmotic (mOsm.L<sup>-1</sup>H<sub>2</sub>O)  
 P<sub>osm blood</sub> = blood osmotic pressure (mOsm.L<sup>-1</sup>H<sub>2</sub>O)

$$HR = \frac{N}{C} \times 100\%$$

P<sub>osm media</sub> = osmotic pressure of the media treatment (mOsm.L<sup>-1</sup>H<sub>2</sub>O)

Hatching rate artemia cysts is done by calculating the *A. salina*. Based on the number of cysts *A. salina* hatched by the number of Nauplius generated. The method can be used to determine the degree of hatching according to Khumaidi (2012),

where :

HR = hatching rate  
 N = number of cysts hatched  
 C = the total number of cysts were hatched

$$E = \frac{Ne}{Te} \times 100$$

Efficiency energetics of *A. salina* cyst hatching is determined by calculating the magnitude of the use of energy yolk-eggs for embryonic development and cyst hatching process, using the following formula

(Anggoro and Muryati, 2007):

where :

E = efficiency energetic of cyst hatching  
 Ne = calorific value after the initial larval cysts hatch  
 Te = initial calorific value : cysts in phase 2 cells

Ca-chorionase enzyme activity is determined by following Johnson (2003), by taking a sample of fluid vitelline cyst *A. salina* 5000 gr mass at 10 hours of incubation, homogenized and consolidated with a reagent calcium phosphate, and then examined with CP -2004 spectrophotometer. The technique of taking liquids vitelline follow the way Anggoro and Nakamura (2002), with the help of Fisher microtonal-blender – 3000 and CP - 35 micropipettes.

Water quality parameters measured media such as temperature, pH, DO and salinity. Measured electrolyte content consisting of Na<sup>+</sup>, Cl<sup>-</sup>, Ca<sup>2+</sup>, Mg<sup>2+</sup>, and K<sup>+</sup>.

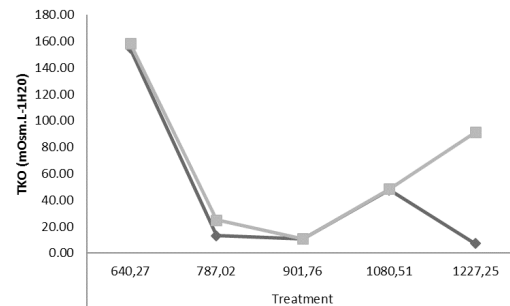


Figure 1. Osmotic level of *A. salina* cysts hatching

Remarks : ◆ Non-Decapsulation;  
 ■ Decapsulation

**Results and Discussion**

TKO

Hatching rate cyst of *A. salina* of each treatment can be seen in Figure 1 .

Based on the Figure 1, highest values of osmotic rate of non-decapsulated *A. salina* first instar were existed on the media with osmolarity of 640.27; 1080.51; 787.02; 901.76 and 1227.25, with the value of 154.71; 47.54; 12.99; 10.76 and 7.28 mOsm.L<sup>-1</sup>H<sub>2</sub>O. On the other hand, the decapsulated artemia instar obtained highest osmotic rate at the media with the osmolarity of 640.27; 1227.25; 1080.51; 787.02 and 901.76 with the value of 158.64; 91.29; 48.44; 24.93 and 10.82 mOsm.L<sup>-1</sup> H<sub>2</sub>O, consequently.

The analysis of variances indicated that the osmotic rate of the artemia cysts was significantly different with the addition of chlorine and various osmotic condition (p<0.05).

This results proves that the energy needs for osmoregulation in isosmotic condition is relatively small compared to the hyperosmotic and hypoosmotic conditions. Treatment A in the present study show that osmolarity value of *A. salina* cyst is higher than the osmolarity of the medium. This suggests that the environmental conditions is hypotonic. In a hypotonic environment, body fluid of aquatic animal is hyperosmotic to the environment. In a hypertonic environment, aquatic animal body fluids is hypoosmotic to the medium, therefore water from the body fluids tends to move to the outside by osmosis (Hamka *et al.*, 2013).

**Hatching Rate (HR)**

The average value *A. salina* cyst hatching during the study can be seen in Figure 2.

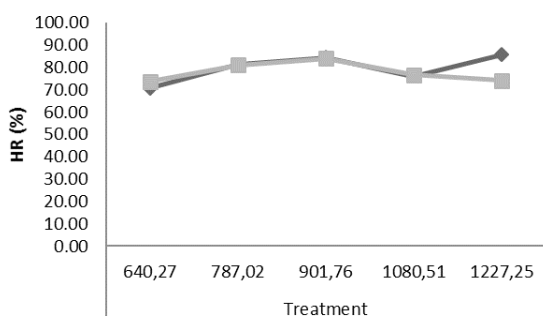


Figure 2. Average value of hatching rate of *A. salina* cyst

Remarks : ◆ Non-Decapsulation; ■ Decapsulation

Based on the figure 2, average values of hatchability of non-decapsulated cyst were obtained in the media with osmolarity of 1227.25; 901.76; 787.02; 1080.51 And 640.27 mOsm.L<sup>-1</sup>h<sub>2</sub>o, i.E. 85.78; 84.43; 81.31; 76.08 And 70.80 %, respectively. On the contrary, hatchability of the decapsulated artemia were high at the media with osmolarity of 901.76; 787.02;1080.51; 1227.25 And 640.27 mOsm.L<sup>-1</sup>h<sub>2</sub>o with the value of 84.05; 81.03; 76.75; 74.18 And 73.78%, consecutively. Analysis of variance indicated that hatching rate of artemia cysts were significantly different (p<0.05) in the present of chlorine and different osmotic conditions of media.

When the osmolarity level of the medium is too high, hatching process of the artemia cysts will be disturbed. In the very high osmotic condition, artemia cysts could not have a chance to obtain enough water for the metabolism process and vice versa (Jubaedah *et al.*, 2006). According to Drinkwater and Crowe (1991), hatching of artemia cysts occurs when there is a differences in osmotic pressure between outside and inside the cysts.

**Energetic efficiency hatching artemia cysts**

The average value of energetic efficiency of *A. salina* cysts hatching in the present study can be seen in figure 3.

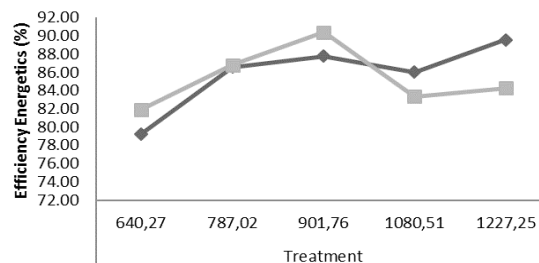


Figure 3. Average value of energetics efficiency of artemia cysts hatching

Remarks : ◆ Non-Decapsulation; ■ Decapsulation

Based on the Figure 3, energetic efficiency of non-decapsulated artemia cysts hatching at the medium osmolarity of 1227.25; 901.76; 787.02; 1080.51 and 640.27 mOsm.L<sup>-1</sup>H<sub>2</sub>O is 89.54; 87.76; 86.58; 86.00 and 79.24 cal/g, while the average value. Analysis of variance showed that the presence of chlorine and osmolarity of medium give no significant effect on the energetic efficiency of *A. salina* cysts hatching (p>0.05).

Highest value was obtained on the TKO of treatment A. The high value of TKO require energy to make ballance to the environment which further lowering the hatching rate efficiency.

**Ca-Chorionase enzyme activity**

Ca-Chorionase enzyme activity is demonstrated at Table 1.

Table 1. Ca-Chorionase enzyme activity on *A. salina* cyst hatching

Media Osmolarity Treatment mOsm.L <sup>-1</sup> H <sub>2</sub> O	Enzyme activity of Ca - Chorionase (µmol Pi.ml <sup>-1</sup> )	
	Non-Decapsulation	Decapsulation
640,27	1898.45	1949.87
787,02	2114.81	2113.02
901,76	2250.18	2256.01
1080,51	2117.65	2119.98
1227,25	2401.47	2408.97

Results of the experiment demonstrated that Ca-chorionase enzyme activity is decreased in hypoosmotic environment. the lower osmotic condition inhibit the hatching process due to the low enzyme

activity. This results is consistent with the result demonstrated by Anggoro and Muryati (2007), which indicated that the environment with too high or too low salinity will thwared hatching process of artemia cysts and weakening the Ca-chorionase enzyme activity, therefore the embrio have a difficulty in breaking the eggshell (chorion).

The mechanism of hatching occurs for two reason. First is due to the embrionic activity and movement, and second is due to the Ca-chorionase enzyme activity. Ca-chorionase enzyme activity plays a role in the process of embrittlement and softening of the eggshell layer to assist the embrio to release from the chorion in the right time (Isriansyah, 2011).

### Water Quality and Content of Electrolyte

Results of water quality and content of electrolyte ions during the study for the temperature were ranges between 26 - 28 °C, pH ranges were from 7.2 to 7.4 and the dissolved oxygen content were ranging from 6.9-7,3 mg/L. The value of the electrolyte ion content in the cyst hatching of *A. salina* with non-decapsulation in osmolarity 640.27; 787.02; 901.76; 1080.51 and 1227.25 mOsm.L<sup>-1</sup>H<sub>2</sub>O is a chloride (Cl<sup>-</sup>) 12.30 to 12.32; 15.12; 17.92; 20.72 and 23.51; sodium (Na<sup>+</sup>) 5.28; 6.48; 7.69; 8.89 and 10.09; calcium (Ca<sup>2+</sup>) 1.72; 2.11; 2.30; 2.66; and 3.28; magnesium (Mg<sup>2+</sup>) 1.73; 2.12; 2.31; 2.67 and 3.27; potassium (K<sup>+</sup>) 1.63; 1.68; 1.82; 2.10 and 3.10.

The electrolyte ion content in hatching process of decasulated *A. salina* cysts in the medium with osmolarity of 640.27; 787.02; 901.76; 1080.51; and 1227.25 mOsm.L<sup>-1</sup>H<sub>2</sub>O is 12.30-12.32; 15.12; 17.92; 20.72; 23.50-23.52 for chloride (Cl<sup>-</sup>), 5.27-5.28; 6.48-6.49; 7.68-7.69; 8.88-8.92; 10.08-10.09 for sodium (Na<sup>+</sup>), 1.72; 2.10-2.11; 2.29-2.30; 2.65-2.66; 3.27-3.28 for calcium (Ca<sup>2+</sup>), 1.72-1.73; 2.11; 2.29-2.30; 2.64-2.65; 3.27 for magnesium (Mg<sup>2+</sup>), and 1.62-1.63; 1.68-1.69; 1.82-1.83; 2.11; 3.09-3.10 for potassium (K<sup>+</sup>). Salinity can be expressed as the total concentration of salts (ionic electrolytes) that is ionized in the water and influence the osmoregulation process.

### Conclusion

Media with osmolarity level of isoosmotic to hyperosmotic (901.76-1227.25 mOsm.L<sup>-1</sup>H<sub>2</sub>O) are able to provide high osmotic rate with low energy for *A. salina* cysts hatching proccess. The energetic efficiency of *A. salina* cysts hatching is the same in the range of 640.27-1227.25 mOsm.L-1H<sub>2</sub>O in the media with osmolarity of hypoosmotic, isoosmotic and hyperosmotic.

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