HATCHABILITY OF VANAME SHRIMP EGG (*Litopenaeus vannamei*) WITH DIFFERENT ETHYLENE DIAMINE TETRA ACETATE (EDTA) IN HATCHING MEDIA

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Abstract. Hatchery is one of the efforts to support the fry production process. However, until now, fry produced in the hatchery have not been able to meet the existing needs due to low egg hatchability and high mortality. The solution used in this problem is to treat seawater using ethylene diamine tetraacetate (EDTA) on hatching media. EDTA is a chelating agent that can help bind heavy metals in seawater and reduce their toxicity. This study aimed to determine the most effective dose of EDTA in increasing the hatchability of vannamei shrimp eggs. The water treatment medium used was a round hatchery with a volume of 3,000 L of water. Based on the study's results, the optimal dose was four ppm with an average fecundity of 174,000 eggs/head, 92% FR, 90% HR, and 85% SR. At a dose of 0 ppm, the average fecundity was 128,000 eggs/head, 59% FR, 61% HR, and 60% SR. At a dose of 2 ppm, the average fecundity was 129,067 eggs/head, 83% FR, 70% HR, and 78% SR. At a dose of 3 ppm, the average fecundity was 133,467 eggs/head, 91% FR, 85% HR, and 80% SR. The results showed that the concentration of EDTA had an effect ($P<0.05$) on the hatchability of vannamei shrimp eggs.

Key words: *Ethylene Diamine Tetra Acetate, Hatching rate, Udang Vaname (Litopenaeus vannamei)*

Running title:

INTRODUCTION

Vaname shrimp is one of Indonesia's leading commodities that continues to increase production, both to meet the needs of the domestic market and the export market. Vaname shrimp farming in Indonesia is currently the mainstay of the aquaculture sector and is a priority for aquaculture development to improve the national economy. In the 2019 period, the achievement of shrimp production was 517,397 tons and is targeted to increase by 250% in 2024 to 1,290,000 tons with a production value of 36.22 trillion in 2019 increasing to 90.30 trillion in 2024 (KKP, 2021). The increase in vaname shrimp production has increased the demand for shrimp seeds. This is a good prospect for hatchery entrepreneurs to develop shrimp hatcheries on a large scale to produce quality seeds.

Hatcheries are one of the efforts to support the production process of quality vaname shrimp fry. However, until now, the fingerlings produced in hatcheries have not been able to meet the existing needs. This is due to the high mortality rate in hatchery centers due to low egg hatchability caused by several factors, such as decreased water quality (Purwono et al., 2012). Therefore, the solution to this problem is to treat the water by adding EDTA to the hatchery media. This is because EDTA has a cheaper price and can also bind heavy metals such as iron (Fe) and copper (Cu) in water that can be toxic to shrimp eggs, so that the quality of water in the egg hatching tanks can be maintained (Afrianto & Abdul, 2014).

Ethylene diaminetetraacetate (EDTA) in egg hatching media is generally done in very low doses, because too high doses can hurt egg development. Several studies have been conducted by Afrianto and Abdul (2014) with a dose of 5 ppm, resulting in a hatching rate of 79%. Based on this, this final project was conducted to determine ethylene diaminetraacetate (EDTA) use with low doses of 0, 2, 3, and 4 ppm in vaname shrimp egg hatching media.

MATERIALS AND METHODS

Study area

The research was conducted in September-December 2023 at CV Krakatau Haura Baraka Hatchery in Kalianda, Lampung Province.

Procedures

Tools and materials in this study are maturation tubs, spawning tubs, hatching tubs, holding tubs, parent sera, naupli sera, aeration, stirrer, beaker glass, sera, microscope, filter bag, pH paper, thermometer, refractometer, male and female parents, EDTA, squid, and sea worms. Observation parameters include fecundity, fertilization rate, hatching rate, survival rate, and water quality, including pH, temperature, and salinity.

The vaname shrimp broodstock selection process is divided into two stages. The first stage is potential. Potential is looking for female broodstock that have matured gonads by looking at the intensity of brownish-yellow gonad color on the back of the female broodstock. Male broodstock that are gonadally mature are characterized by the appearance of white sperm bags located near the foot of the road and potentially carried out in the morning at 08.00 WIB.

The second stage is sampling. Sampling is looking for fertilized female broodstock by looking at the presence or absence of sperm attached to the telikum of gonadally mature female shrimp broodstock. The sampling process is carried out by sliding the female parent, as the characteristics of the mated female parent will be seen in sperm attached to the female ovum genitals. Sampling is done at 09.00, 10.00, and 11.00 am.

The mature fertilized female parent is moved into a hatching tub and given water treatment with the addition of EDTA at different doses according to 4 treatments. Each hatching tub has a volume of water as much as 3000 L. Furthermore, the parent performs the egg release process. After the female parent laid the eggs, the female parent was returned to the maturation tub. This process was carried out at 22.00 WIB.

Data analysis

The method used in this research was the experimental method and a complete randomized design (CRD) method, consisting of 4 treatments, and each treatment was repeated as many as 3 times. The treatments were;

P1: 0 ppm EDTA dose
P2: dose of EDTA 2 ppm
P3: dose of EDTA 3 ppm
P4: dose of EDTA 4 ppm

RESULTS AND DISCUSSION

Fecundity of Vaname Shrimp Broodstock

Based on the data obtained, it is known that fecundity is increasing along with the increasing content of EDTA added to the hatching medium, which is consecutively 128,000, 129,067, 133,467, and 174,000 grains/head.

Table 1. Duncan Test Results of EDTA Treatment on Fecundity

No	Treatment	Average
1	0 ppm	128.000±4.000 [a]
2	2 ppm	129.067±3.717 [a]
3	3 ppm	133.467±5.605 [a]
4	4 ppm	174.000±6.000 [b]

Different superscript letters indicate significantly different results ($P<0.05$)

In the treatment of EDTA, 0, 2, and 3 ppm get a lower average fecundity than four ppm. This is thought to be because the higher the dose of EDTA, the better the fecundity of the reared vaname shrimp. The function of EDTA is to help maintain water quality by binding heavy metals in the maintenance media. According to the results of research by Fuady (2013), the content of heavy metals in the maintenance media can affect the fecundity of vaname shrimp eggs. Fecundity of vaname shrimp eggs shows a decrease in fecundity resulting from vaname shrimp reared in media contaminated with heavy metals.

Fertilization Rate (Degree of Fertilization)

Based on the data obtained, it is known that the degree of fertilization is increasing with increasing doses of EDTA addition to the hatchery media. Consequently, the FR values produced were 59%, 83%, 91%, and 92% for adding EDTA 0, 2, 3, and 4 ppm. This is thought to be due to the addition of EDTA to the hatching media, which can form complexes with heavy metals such as iron, calcium, and magnesium. In hatcheries, EDTA is usually added to the water medium to encourage the development and survival of eggs and larvae. This is by the results of research by (Fuady, 2013) that heavy

metal exposure in the rearing medium can affect the fecundity of vaname shrimp eggs, the use of EDTA in the rearing medium can help reduce heavy metal exposure and improve water quality, which in turn will affect the Fertilization rate (FR).

The results of Duncan's test on treating EDTA against FR are in Table 2.

No	Treatment	Average
1	0 ppm	59±1,15[a]
2	2 ppm	83±5,77[b]
3	3 ppm	91±6,43[b]
4	4 ppm	92±6,93[b]

Different superscript letters indicate significantly different results (P<0.05)

Hatching Rate (Degree of Hatching)

Observation of the degree of hatching or Hatching Rate (HR) of vaname shrimp aims to determine the hatchability of vaname shrimp eggs. The degree of hatching rate obtained successively for adding EDTA 0, 2, 3, and 4 ppm is 61%, 70%, 85%, and 90%. EDTA treatment at zero ppm produces a lower average HR Giving EDTA 2, 3, and 4 ppm is effective in increasing shrimp eggs' hatchability compared with no EDTA. The higher the dose of EDTA, the better the effect on the HR of the reared vaname shrimp. This is because heavy metals in the waters are known to have various toxic effects at different concentrations depending on the species and stage of growth (growth rate slows down), development (cell formation and cell division slows down), health (respiration), and larval survival which is much more susceptible to death (mortality). More susceptible to mortality. This is why heavy metals, even at low or high concentrations, are considered a significant problem for hatchery production (McDougall, 2019).

Table 3. Duncan Test Results of EDTA Treatment on HR

No	Treatment	Average
1	0 ppm	61±0,57[a]
2	2 ppm	70±7,23[a]
3	3 ppm	85±8,18[b]
4	4 ppm	90±7,54[b]

Different superscript letters indicate significantly different results (P<0.05)

Survival Rate (Survival Rate)

Observation of the Survival Rate Naupli (SR) of vaname shrimp aims to determine the survival of vaname shrimp naupli. Survival treated with EDTA gets different results. The average survival naupli obtained successively for adding EDTA 0, 2, 3, and 4 ppm is 60%, 78%, 80%, and 85%. In the zero ppm EDTA treatment, the lowest average SR is obtained; the higher the dose of EDTA used, the better the effect on the SR of the reared vaname shrimp naupli. This is because EDTA can help bind heavy metals in the maintenance media to increase the survival rate of vaname shrimp naupli. This is based on McDugal's (2019) statement that EDTA helps bind heavy metals to improve larval survival by reducing the concentration of chromium, copper, zinc, cadmium, and lead in marine waters.

Table 4. Duncan Test Results of EDTA Treatment on SR

No	Treatment	Average
1	0 ppm	60±4,35[a]
2	2 ppm	78±2,30[b]
3	3 ppm	80±2,51[b]

4	4 ppm	85±5,77[b]

Different superscript letters indicate significantly different results (P<0.05)

Water Quality

The results of water quality measurements in both maturation tanks, hatching tanks, and holding tanks show the optimal range of values by SNI (BSN, 2014) to support the production of naupli CV Karakatau Haura Baraka. This is supported by the high percentage of the highest degree of hatching, which is 90%. Hatchability and larval development are closely related to water quality, such as temperature and salinity. The embryonic phase

The embryonic phase until hatching into naupli is a phase that is very vulnerable to environmental changes, even if the changes occur in a narrow range of time (Pratiwi, 2020). The following results of water quality observations can be seen in Table 5.

5. Water Quality Parameters

Parameters	Unit	Maturation Tub	Hatching Tub	Holding Tub	Literature Source
pH	-	7-8	7-8	7-8	7-8 (SNI., 2014)
Temperature	⁰ C	28-29	29-30	29-30	28-33 (SNI,2014)
Salinity	ppt	30-31	30-31	30-31	30-34 (SNI, 2014)

Discussion

The provision of EDTA in the hatching media of vaname shrimp eggs can effectively increase the hatchability of vaname shrimp eggs by binding heavy metals in the hatching media. The optimal dose of all treatments was four ppm with an average fecundity of 174,000 eggs/head, FR 92%, hatching degree 90%, and SR 85%.

The suggestion to be given from this research is that further research needs to be carried out on the provision of EDTA with different doses in the hatching medium for shrimp hatchery activities so that there will be more references related to the use of EDTA doses in seawater treatment and as a reference for hatcheries in order to carry out optimal water treatment to increase the seed products produced.

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