



# The Effect of Temperature to Incubation Period, Hatching Rate, Normality and Larvae Size of *Lutjanus johnii* Bloch, 1792

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**ABSTRACT:** This research analyzed the effect of temperature to the incubation period, hatching rate, larva normality and its size, volume of yolk and oil globule of *Lutjanus johnii* when it hatched so the optimal temperature for embryo growth can be determined. This research was an experimental research with 4 treatments of temperature (26°C, 28°C, 30°C and 32°C) and 3 repetitions. The results showed that incubation period needed at 26°C: 20.11±0.31 hours, 28°C: 17.08±0.425 hours, 30°C: 14.06±0.25 hours and 32°C: 13.08±0.16 hours. The highest hatching rate achieved at 30°C and 32°C temperature ( $P>0.05$ ) with 93.8±1.52% and 96.12±1.6% while the highest normality was at 28°C and 30°C temperature ( $P>0.05$ ) with 79.5±4.21% and 83.97±2.59%. The best size at hatch was at 26°C with 1684.35±8.06 μm, while the volume of yolk and oil globule at hatch did not show much difference. Based on the results acquired, incubation temperature at 30°C produced the best hatching rate, normality and larva length.

**Key words:** Temperature, Incubation Period, Hatching Rate, Normality, Larvae size, *Lutjanus johnii*

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## INTRODUCTION

International and domestic market in Indonesia on red snappers was overwhelming as it reached 199,088 metric tons in 2012 [1]. This condition creates opportunity for cultivating this commodity. Efforts on the hatchery of some red snappers such as *L. argentimaculatus* [2], *L. camphecanus* [3], *L. guttatus* [4], *L. peru* [5], *L. colorado* [6] has been done, but they are still experimental so the seed availability is still a major problem in its development [8].

*Lutjanus johnii* is one of red snappers' kinds which have been successfully bred at Maincentre for Mariculture Development (BBPBL) Lampung, Indonesia. However, the survival rate from seed mass productions is still very low. High mortality rate happens in embryotic and larval phases are the cause of fail attempts on its hatchery [9]. Embryotic and larval phases are phases which the fish are very susceptible to environmental change even in it is in a small scale [10]. One of the environmental factors which greatly affect the growth of the embryos and larvae is temperature [11] as it can affect its hatching rate, and survival rate of larvae [12], the size of larvae at hatch and yolk volume [13], incubation period [14] and abnormality to larvae produced [15].

Considering the susceptibility of embryotic phase and the great effects of the temperature, this research was aimed to determine optimal temperature needed for *Lutjanus johnii* embryos' growth based on its incubation period, hatching rate, larvae normality, larvae size at hatch and the volume of yolk and oil globule of *Lutjanus johnii* larvae at hatch.

## MATERIAL AND METHODS

This research was an experimental research using complete random design with 4 treatments of temperature (A: 26±0.5°C; B: 28±0.5°C; C: 30±0.5°C; D: 32±0.5°C) and 3 repetitions, which was conducted at Centre for Mariculture Development Lampung. Research container was baker glass with 1000 mL in capacity, located in the air-conditioned room with an aerator, an electric heater and a thermostat.

Eggs were from natural spawning of 5 male parents (weight 3.85±0.7 kg; length 48.06±2.5 cm) and 5 female parents (weight 4.1±0.21 kg; length 51.34±2.46 cm). Spawning was naturally done in a fiberglass round container with 15m<sup>3</sup> in volume. Eggs harvested then were selected by siphoning the eggs settled in the base of the container, while the floating eggs were used for the research. Selected eggs were then incubated in research container with 1000 eggs/L in density [16].

Larvae sample which had just hatched were taken 5 from each treatment [17], they were observed using a stereo microscope. Each sample was captured with AxioCam ERc 5s microscope camera and then measured by Axioo Vision Rel. 4.8. Total length measurement was conducted from the tip of the mouth to the end of the tail while yolk volume was calculated based on formula  $V: 0.1667 \cdot \pi \cdot L \cdot H^2$  and  $V : 4/3 \cdot \pi \cdot r^3$  for oil globule [18]. Incubation period and hatching rate were determined based on Gao et al. [19] research. Normality was the larvae percentage which possessed normal shape when they hatched [20] from the number of eggs which were hatched. The data then analyzed using MS Excel.

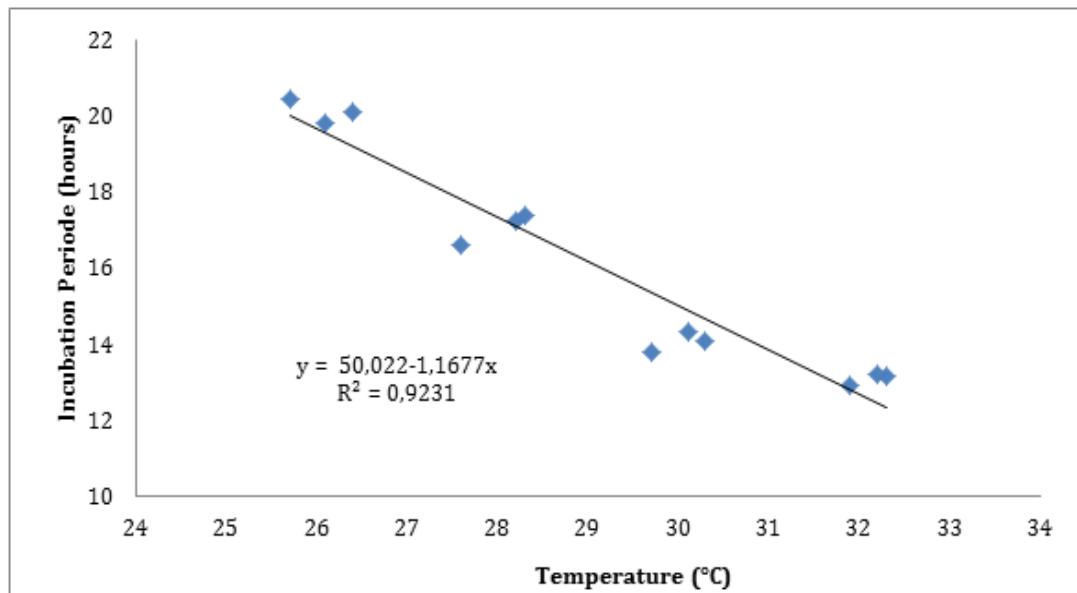
## RESULT

Incubation temperature gave significant effects to incubation period, hatching rate, normality of the larvae produced ( $P<0.05$ ). The data acquired on incubation period, hatching rate and normality of the larvae was shown in Table 1.

Incubation period from all treatments differed significantly ( $P<0.05$  and  $P<0.01$ ). The longest incubation period was at  $26^\circ\text{C}$  with  $20.11 \pm 0.31$  hours, while the fastest one was at  $32^\circ\text{C}$  with  $13.08 \pm 0.16$  hours. Incubation period on each treatment differed significantly and closely correlated with temperature on a linear pattern so the higher the temperature the shorter incubation period (Figure 1).

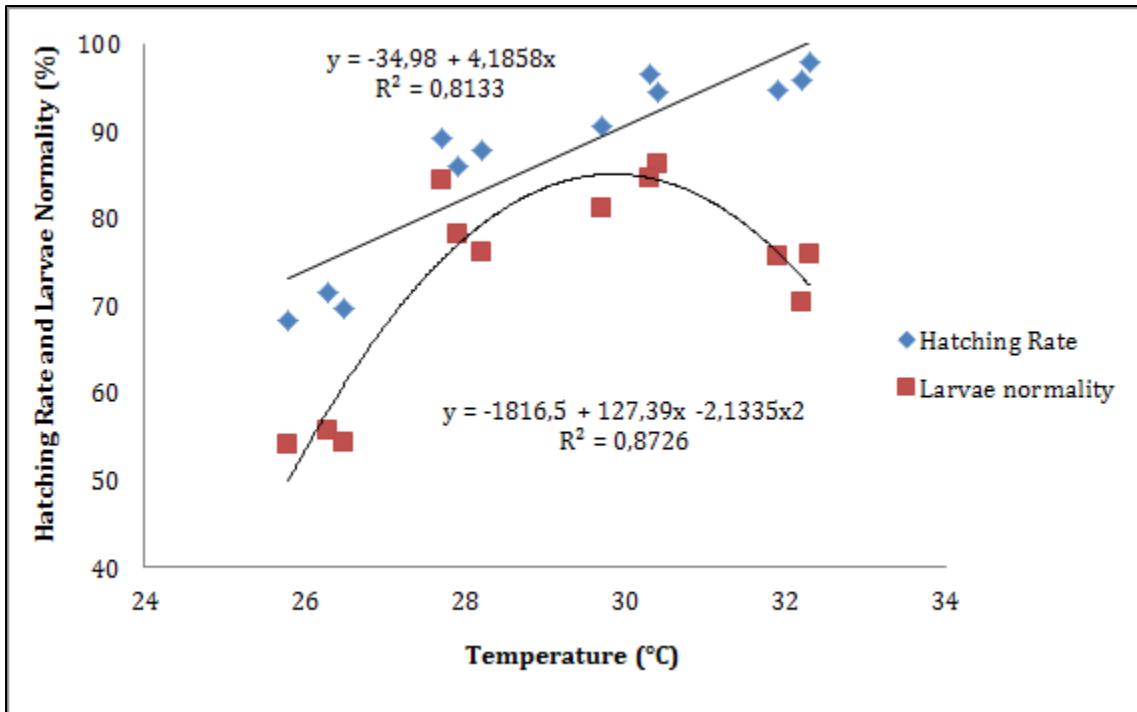
**Table 1.** Incubation period, hatching rate and normality of the larvae at hatch

Temperature (°C)	Incubation Period (Hour)	Hatching Rate (%)	Normality of the Larvae (%)
26	$20.11 \pm 0.31^d$	$69.78 \pm 1.61^a$	$54.61 \pm 0.88^a$
28	$17.08 \pm 0.425^c$	$87.59 \pm 1.52^b$	$79.5 \pm 4.21^c$
30	$14.06 \pm 0.25^b$	$93.8 \pm 2.99^c$	$83.97 \pm 2.59^c$
32	$13.08 \pm 0.16^a$	$96.12 \pm 1.63^c$	$73.86 \pm 3.14^b$



**Figure 1.** Regression of temperature's effect to incubation period of *Lutjanus johnii* embryos

Hatching rate on all incubation temperatures significantly different ( $P<0.05$ ), except at  $30^\circ\text{C}$  and  $32^\circ\text{C}$  ( $P>0.05$ ). The highest hatching rate achieved at  $30^\circ\text{C}$  and  $32^\circ\text{C}$  with  $93.8 \pm 2.99\%$  and  $96.12 \pm 1.63\%$  respectively while the lowest hatching rate was at  $26^\circ\text{C}$  with  $69.78 \pm 1.61\%$ . Larvae normality at hatch was also significantly different from all treatments ( $P>0.05$ ), except incubation period at  $28^\circ\text{C}$  and  $30^\circ\text{C}$  ( $P<0.05$ ). The highest normality was at  $28^\circ\text{C}$  and  $30^\circ\text{C}$  with  $79.5 \pm 4.21\%$  and  $83.97 \pm 2.59\%$  respectively and optimal temperature at  $29.86^\circ\text{C}$ , temperature at  $26^\circ\text{C}$  showed the lowest normality with  $54.61 \pm 0.88\%$  (Table 1). Temperature was closely correlated with the hatching rate and normality of larvae at hatch, where the hatching rate was linearly correlated with incubation temperature which was the higher the temperature the higher the hatching rate while normality of larvae at hatch followed quadratic pattern to incubation temperature where its value went up until the optimal temperature between  $28^\circ\text{C}$  and  $30^\circ\text{C}$  and then went down after that range of temperature (Figure 2).

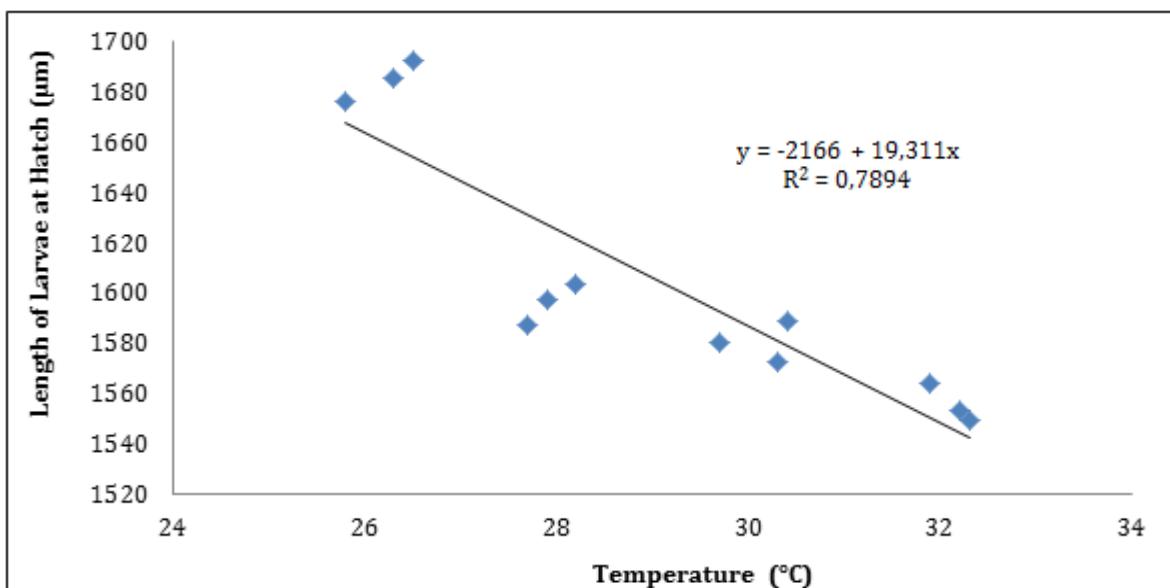


**Figure 2.** Regression between temperature, hatching rate and normality of the larvae

Temperature significantly affects the length of larvae at hatch ( $P<0.05$ ), but it did not significantly affect yolk volume and oil globule at hatch ( $P>0.05$ ) (Table 2.) Length of larvae at hatch was linear with the incubation temperature and closely correlated, which temperature at 26°C resulted the longest larvae  $1684.35\pm8.06$   $\mu\text{m}$  (Figure 3). However, temperature did not significantly affect yolk volume and oil globule at hatch ( $P>0.05$ ).

**Table 2.** Length of larvae, yolk and oil globule volume at hatch

Temperature (°C)	Length of Larvae ( $\mu\text{m}$ )	Yolk Volume ( $\text{mm}^3$ )	Oil Globule Volume ( $\text{mm}^3$ )
26	$1684.35\pm8.06^{\text{c}}$	$0.075\pm0.0095^{\text{a}}$	$0.0019\pm0.00006^{\text{a}}$
28	$1595.74\pm8.13^{\text{b}}$	$0.087\pm0.0075^{\text{a}}$	$0.002\pm0.00006^{\text{a}}$
30	$1580.66\pm8.14^{\text{b}}$	$0.077\pm0.007^{\text{a}}$	$0.002\pm0.00017^{\text{a}}$
32	$1555.57\pm7.8^{\text{a}}$	$0.076\pm0.012^{\text{a}}$	$0.002\pm0.00006^{\text{a}}$



**Figure 3.** Regression between temperature and length of larvae at hatch

## DISCUSSION

The velocity of incubation period was increased by the heightened incubation temperature, the difference of the periods on all treatments was very significant ( $P<0.05$ ). Linear effect of incubation period to temperature was also happened to some other kinds of fish such as *Lutjanus peru* [21] and *Epinephelus striatus* [22]. However, incubation period showed from all treatments were faster than *Lutjanus sebae* which has incubation period about 22.15 hours at 27-28°C [24]. The significance of incubation period difference from all treatments especially at 26°C:  $20.11\pm0.31$  hours and at 32°C :  $13.08\pm0.16$  hours showed that the higher temperature of incubation the faster embryotic growth so the last phase of embryos can be achieved faster, where at that phase embryos can secreted hatching enzyme proteolytic diffused from hatching gland cells [25].

This research showed increasing hatching rate along with rising temperature, but some other fish such as *Inimicus japonicus* has negative linear pattern where the hatching rate declines along with the rising temperature [26] or *Cyprinus carpio* which has increasing hatching rate at 30°C and decreasing at higher temperature [27]. Different pattern of hatching rate of various fish showed different ability to temperature adaptation of each kind of fish. Hatching rate will keep increasing if it is incubated at optimal temperature range and it will decrease if the temperature is over that temperature range. High hatching rate showed at 30°C and 32°C was because the temperatures were still at tolerated temperature range of red snappers which is at 33.5°C [28].

High normality of larvae at 28°C and 30°C showed that at those temperatures the embryos could adapt and grew well, while low normality at 26°C and 32°C showed disruption in the embryos' energy metabolism line that it affected their adaptation ability and embryotic growth [29]. Researches on *Plectropoma laevis* showed declining metabolism activity at high temperature [30]. Declining metabolism activity causes energy supply and distribution line in the embryos decreasing so the growth runs slow while more metabolism activity causes energy demand to rise [31], but more energy distribution is used for adaptation so the growth energy decreases [32]. Indicators which can be used are larvae's length, yolk volume and oil globule. Larvae at hatch produced at 32°C showed the shortest length with insignificant difference of yolk volume and oil globule on all treatments. Great length of larvae at 26°C incubation temperature was assumed to be closely related to the length of incubation period [32], where longer incubation period allowed for embryos to grow longer so they could hatch with greater length from embryos which hatch with shorter incubation period.

## CONCLUSION

Temperature is closely correlated with incubation period, hatching rate, normality and larvae size at hatch but insignificantly affects yolk volume and oil globule at hatch. Based on incubation period, hatching rate, larvae's normality and length results, optimal temperature for red snappers *Lutjanus johnii* Bloch, 1792 incubation is 30°C.

### Suggestion

Further research is suggested on incubation temperature at larval phase to determine optimal temperature for larvae rearing.

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