
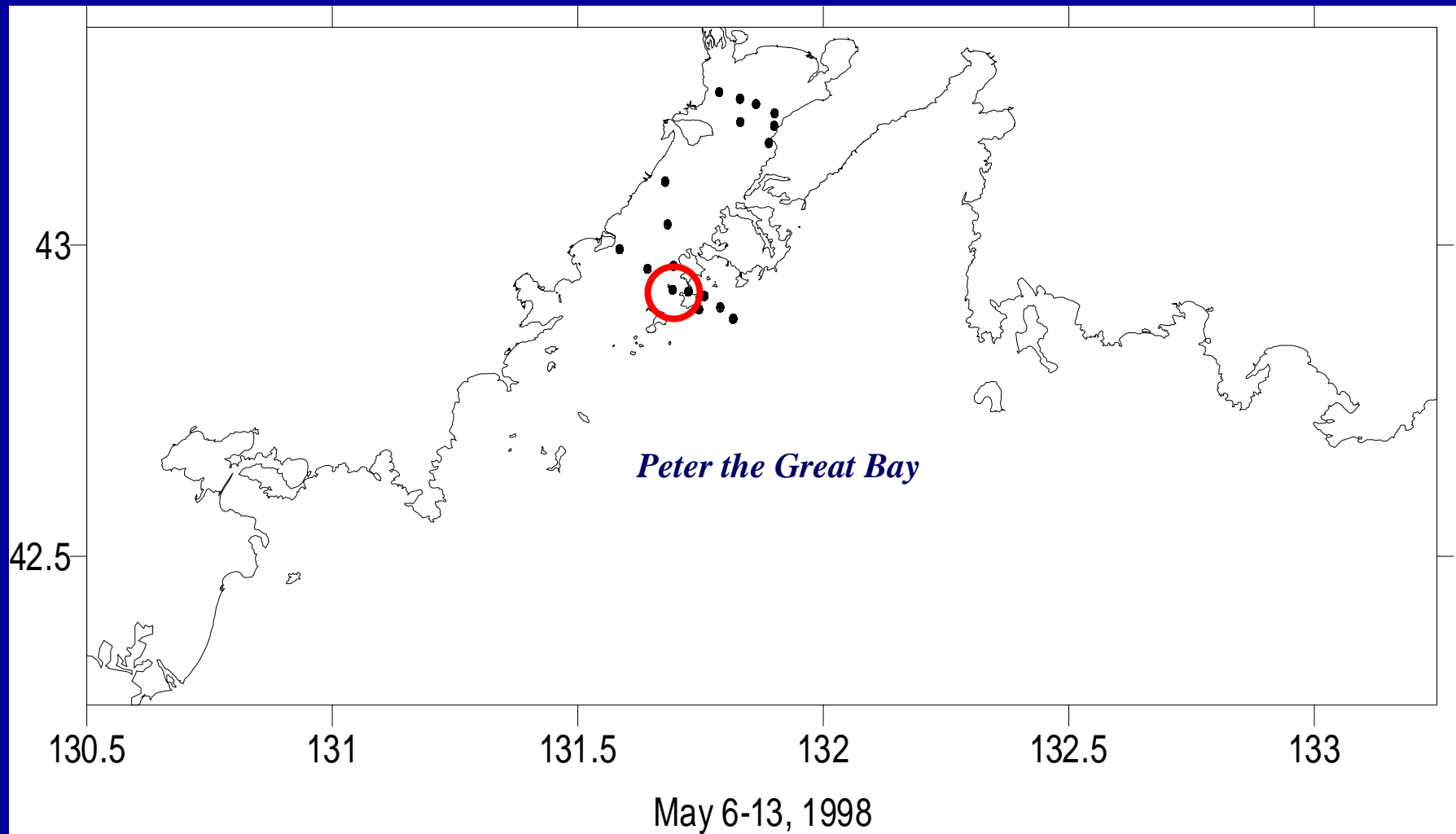


# *In situ* experiments to investigate the japanese anchovy eggs development

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1. To trace daily rhythm of anchovy spawning
  2. To evaluate the per hour anchovy eggs mortality on certain stages
  3. To determine the time of the anchovy eggs development
  4. To estimate the total mortality of anchovy during its embryogenesis

## Area of surveys (June, July of 2004)



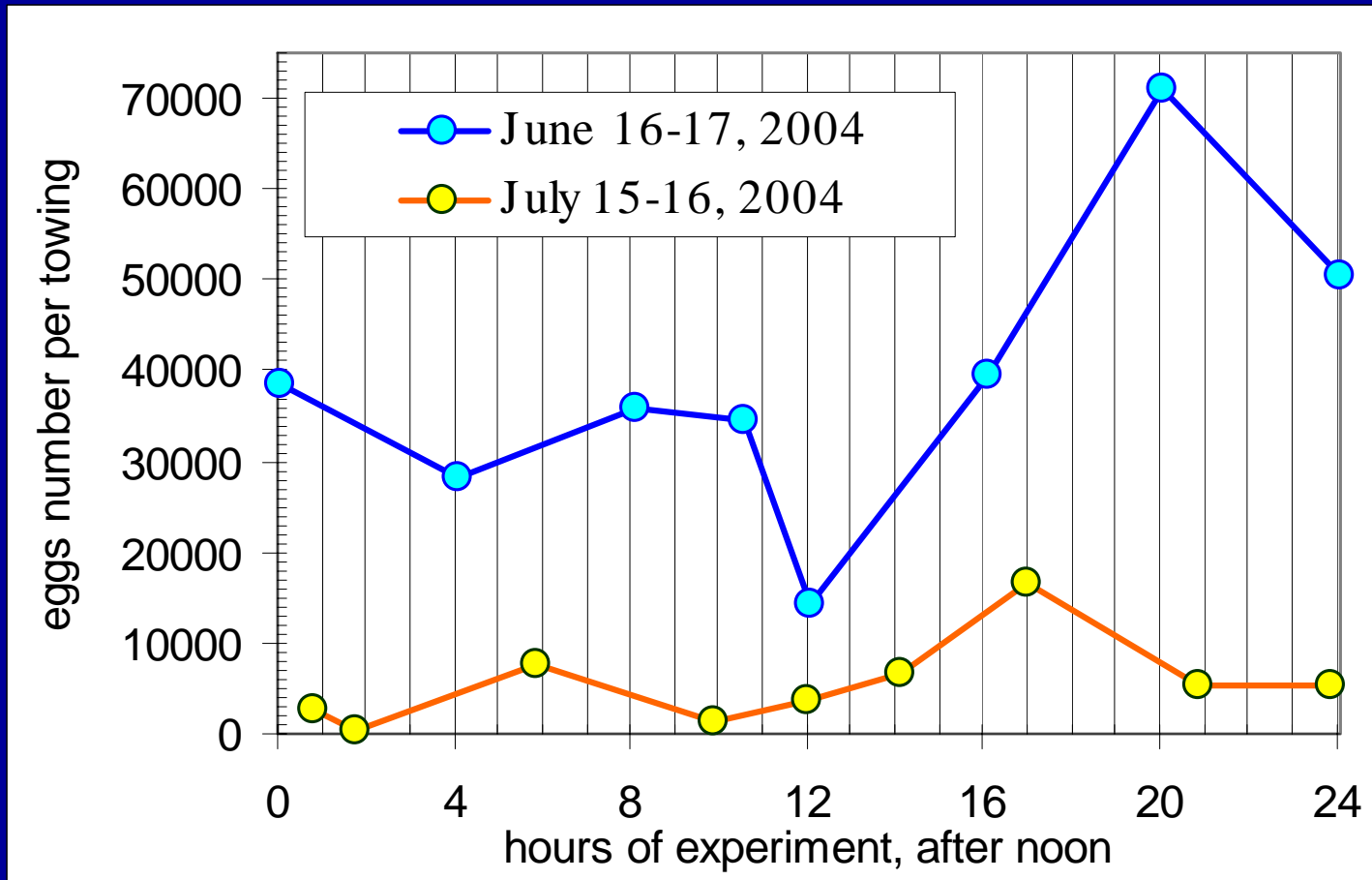
○ - the 24-hour ichthyoplankton stations

# Sampling



The samples of fish eggs were collected with 1-2 hours interval by 10-minutes horizontal towing the IKS-80 planktonic net ( $S = 0.5 \text{ m}^2$ ) in surface layer with the velocity about 2.5 kt.

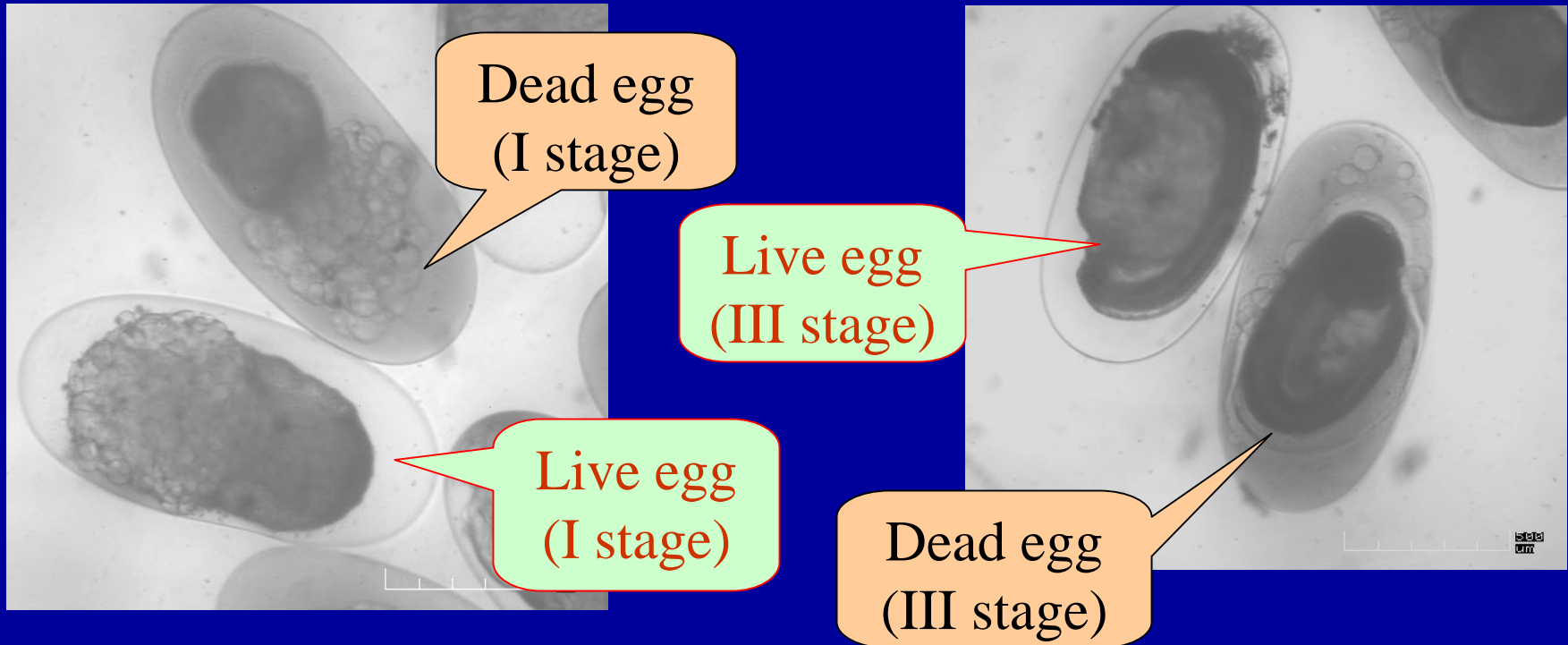
## Change of the total number of anchovy eggs per towing



The fluctuations look as accidental ones; so impossible to understand the time of mass spawning.

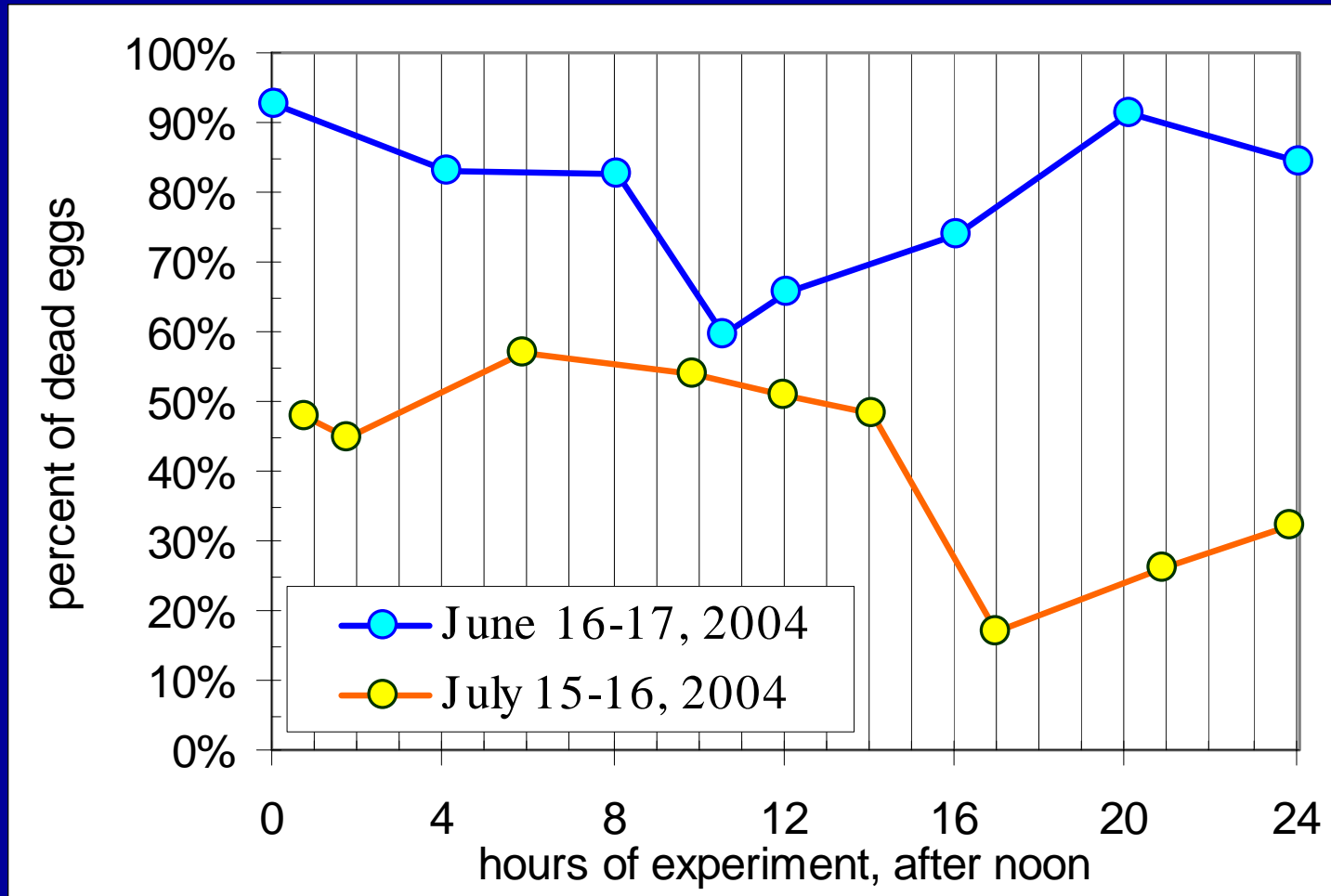
We suppose that a total number of eggs in certain case is determined by accidental reasons, as patchiness of ichthyoplankton.

## Live and dead eggs



Internal structure of the dead eggs is destructed: yolk membranes are broken, yolk granules are fallen out, egg membrane is deformed, germinal disk or developing embryo is deformed, shifted off the normal position and clenched, the whole internal space of the egg is whitened and looks opaque under microscope.

## Mean share of dead eggs is 60 %

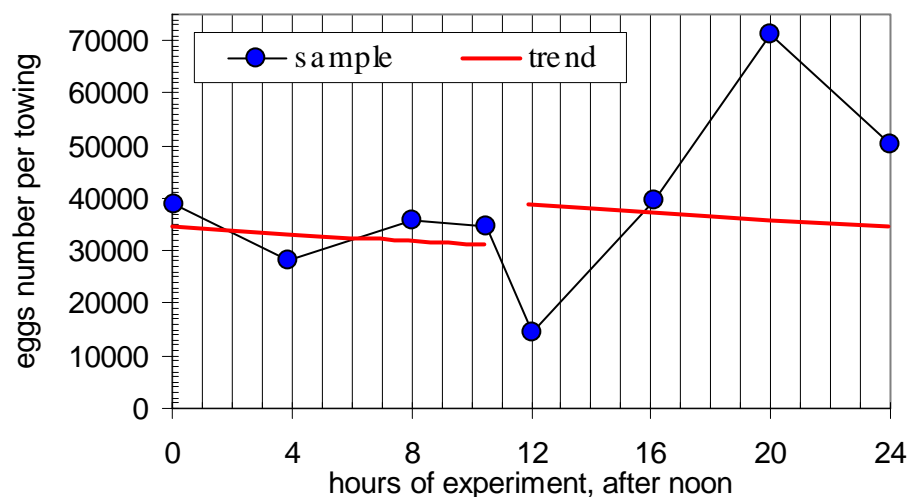


In aquarial experiments, the dead eggs sink down immediately. In natural conditions they remain for a time in surface layer because of water stratification and turbulence.

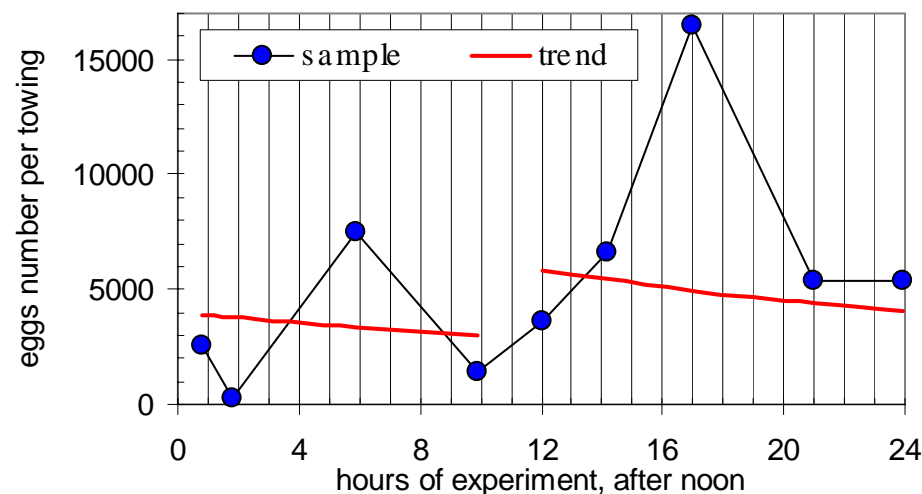
However, daily dynamics of their share could be traced with the minimum at night-morning.

# Total number of eggs had insignificant negative trend after the time of mass spawning in midnight

June 16-17, 2004



July 15-16, 2004



$$N = N_{\text{midnight}} * 0.99^t$$

$$N = N_{\text{midnight}} * 0.97^t$$

The trend could be presented as exponential function  $N = N_{\text{midnight}} * k^t$ , where  $t$  – is the time after midnight.

For the total number of eggs, the  $k$  is close to 1, that means a slight elimination of the eggs (mainly dead ones) because of sinking, grazing, hatching or other reasons.

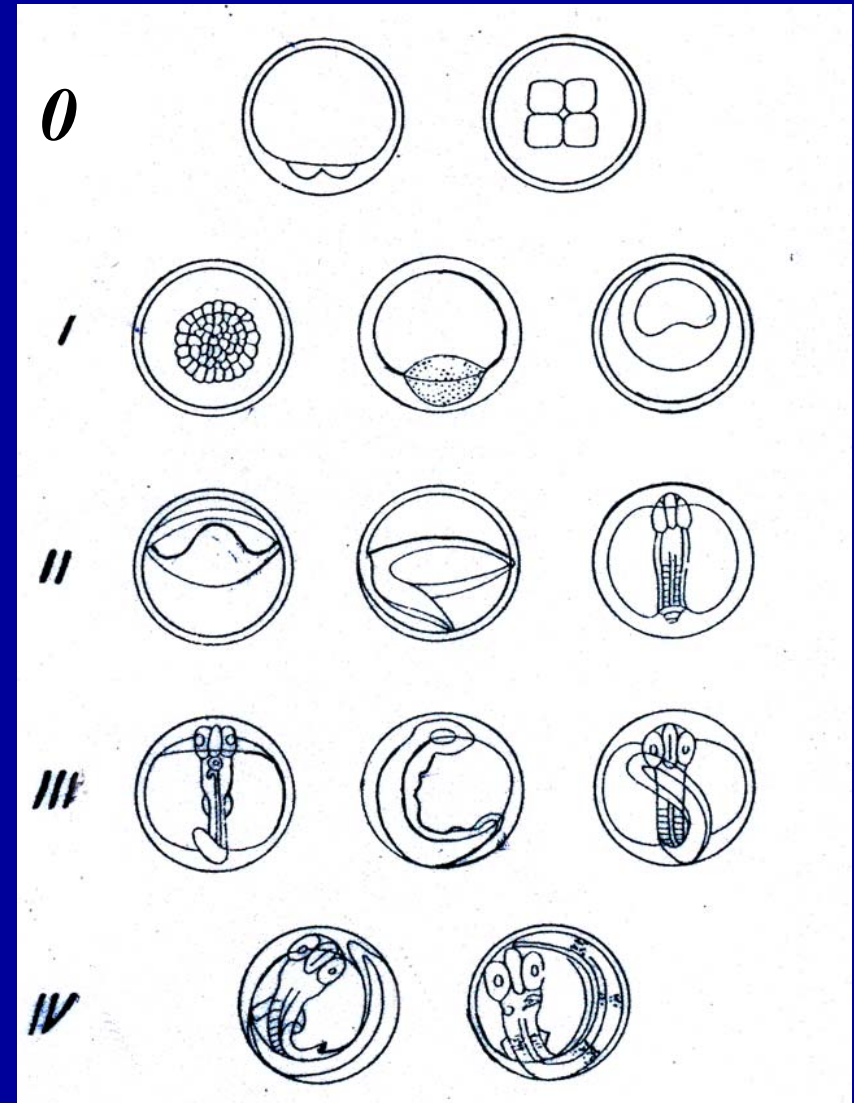
# Were the dead eggs damaged in the process of sampling?

To check this hypothesis, let us consider the daily changes of the “age structure” of live and dead eggs, taking into account the stages of their development.

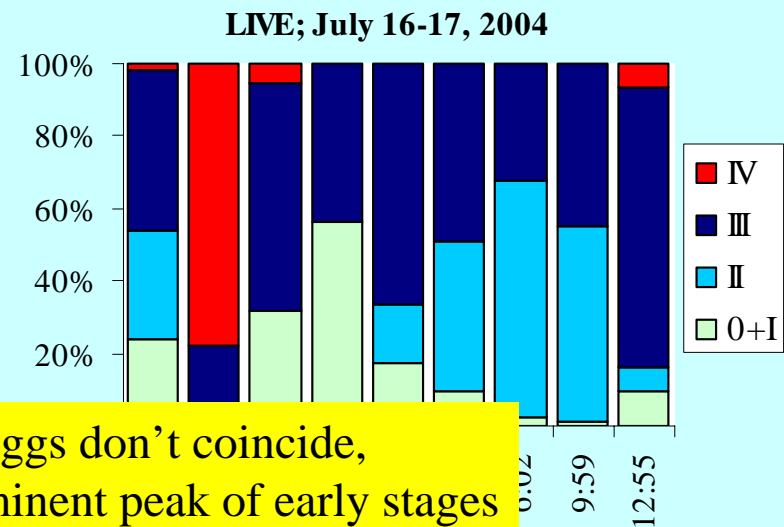
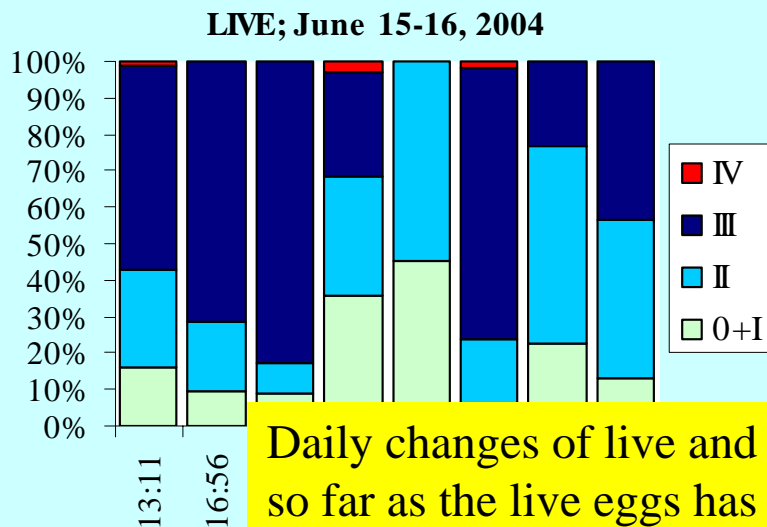
According to Rass (1949), the following 5 stages of development could be distinguished during the embryogenesis.

Step-by-step, the embryo changes from a couple of cells to a small fish (larvae).

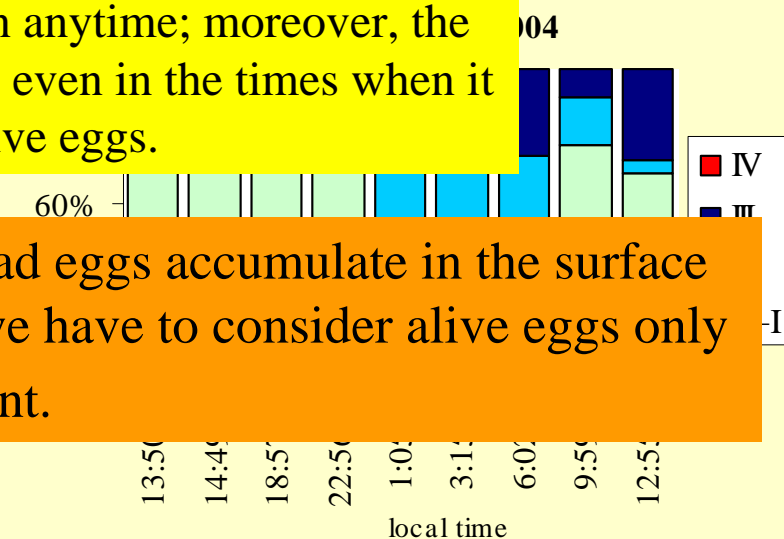
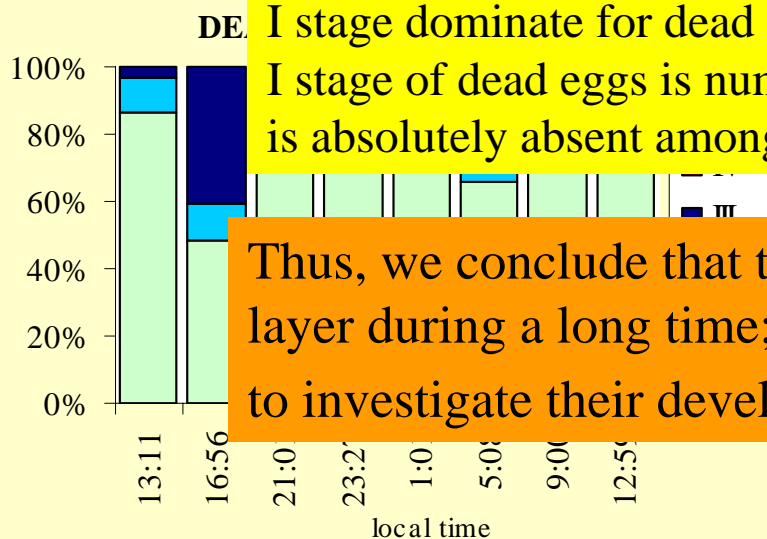
Two main states of an embryo could be distinguished: the early embryogenesis (stages 0 and I) when the embryo has no organs; and the late embryogenesis (stages II-IV) with developing organs.



# Daily changes of the structure of live and dead eggs

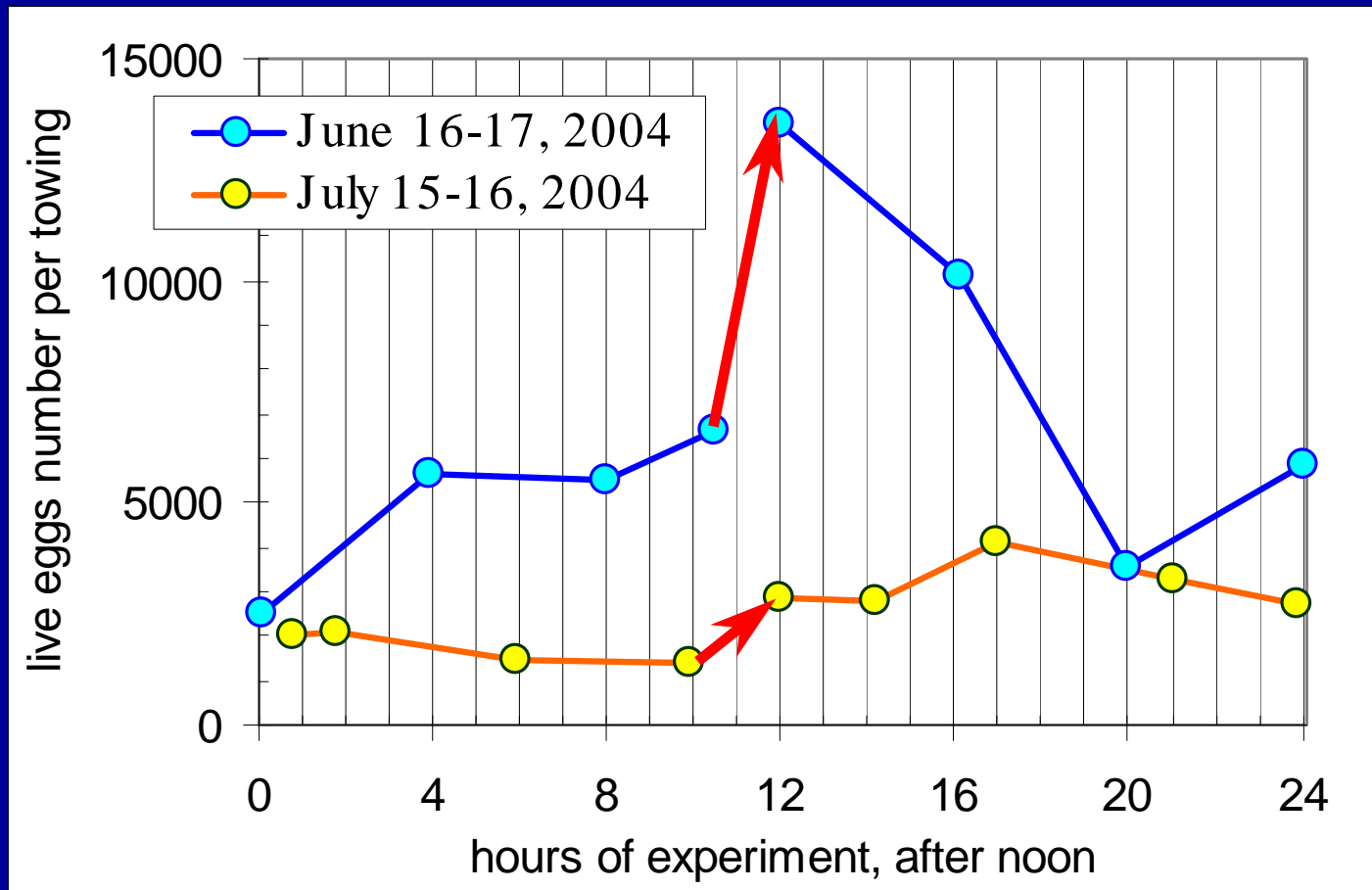


Daily changes of live and dead eggs don't coincide, so far as the live eggs has a prominent peak of early stages at midnight, corresponding to the time of mass spawning, and the III stage dominates in daytime, but the eggs of the I stage dominate for dead eggs in anytime; moreover, the I stage of dead eggs is numerous even in the times when it is absolutely absent among the live eggs.



Thus, we conclude that the dead eggs accumulate in the surface layer during a long time; so, we have to consider alive eggs only to investigate their development.

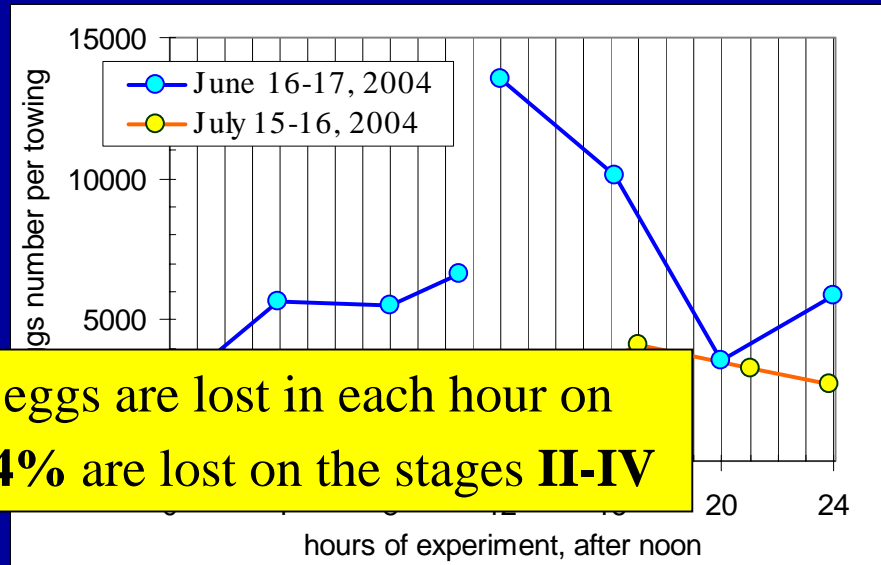
The number of live eggs decreased considerably after the time of mass spawning in midnight



For any period between observations  $i$  and  $i+1$ , the eggs survival could be presented by exponential function  $N_{i+1} = N_i * s^{\Delta t}$ , where  $s$  – coefficient of survival;  $\Delta t$  – time interval. The eggs per hour mortality is:  $m = 1-s$ .

The coefficients of survival/mortality had a significant fluctuations that could be explained by changes of the stages ratio

| June 16-17, 2004 |                         | July 15-16, 2004 |                         |
|------------------|-------------------------|------------------|-------------------------|
| time interval    | coefficient of survival | time interval    | coefficient of survival |
| 0--4             | 1.22                    | 0--2             | 1.03                    |
| 4--8             | 0.99                    | 2--6             | 0.84                    |
| 8--10            |                         |                  |                         |
| 10--12           |                         |                  |                         |
| 12--16           |                         |                  |                         |
| 16--20           |                         |                  |                         |
| 20--24           | 1.13                    | 21--24           | 0.94                    |



So, in average **16-20%** of live eggs are lost in each hour on the stages **0 and I**, but only **0-4%** are lost on the stages **II-IV**

For any certain period, the total survival of the eggs is composed from the partial survival of each stage fraction, or  $s = p_I s_I + p_{II} s_{II} + p_{III} s_{III} + p_{IV} s_{IV}$ , where  $p$  – percentage of each stage.

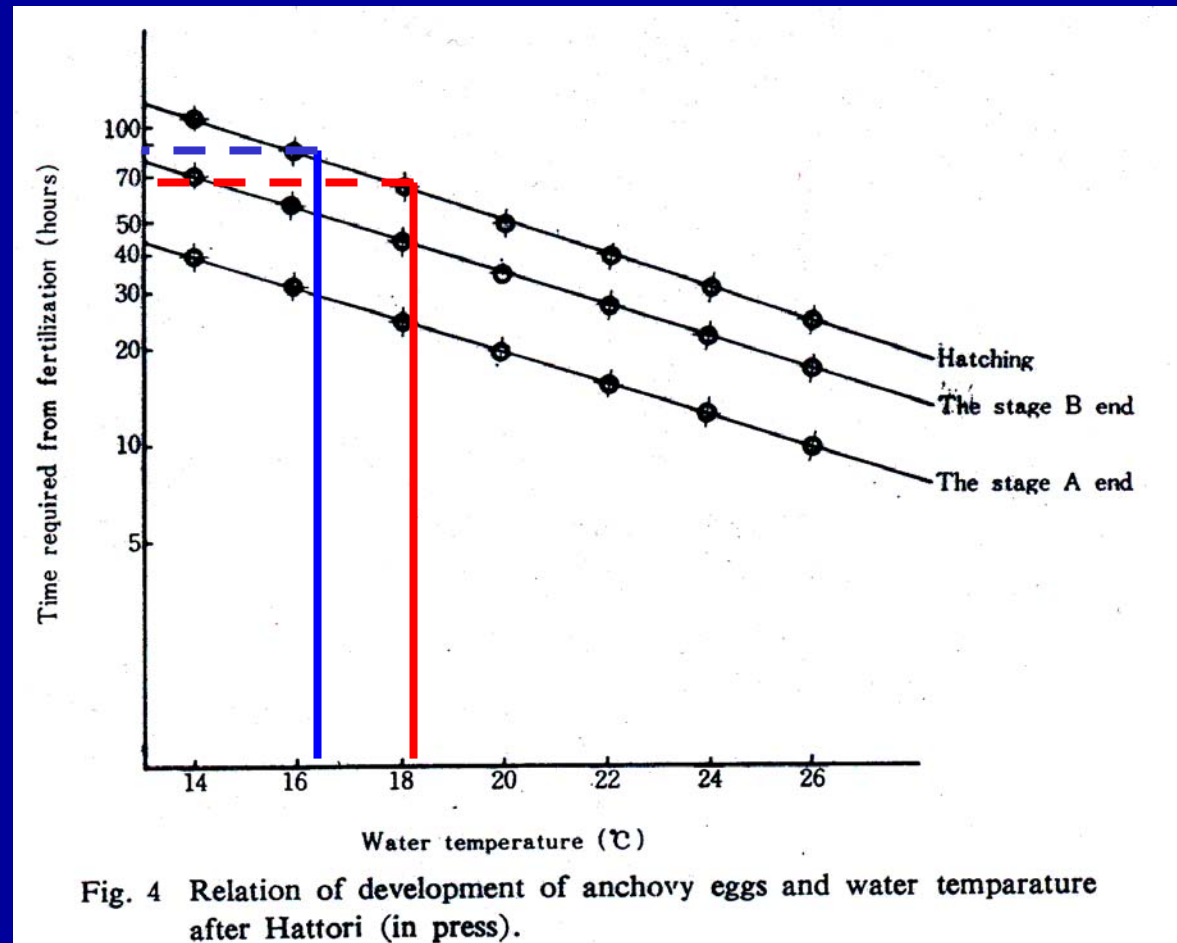
Using this formula for all intervals, we have estimated the coefficients of survival for each stage of embryogenesis. They are the following:

|                  | stage 0+I | stages II-IV |
|------------------|-----------|--------------|
| June 16-17, 2004 | 0.80      | 1.00         |
| July 15-16, 2004 | 0.84      | 0.96         |

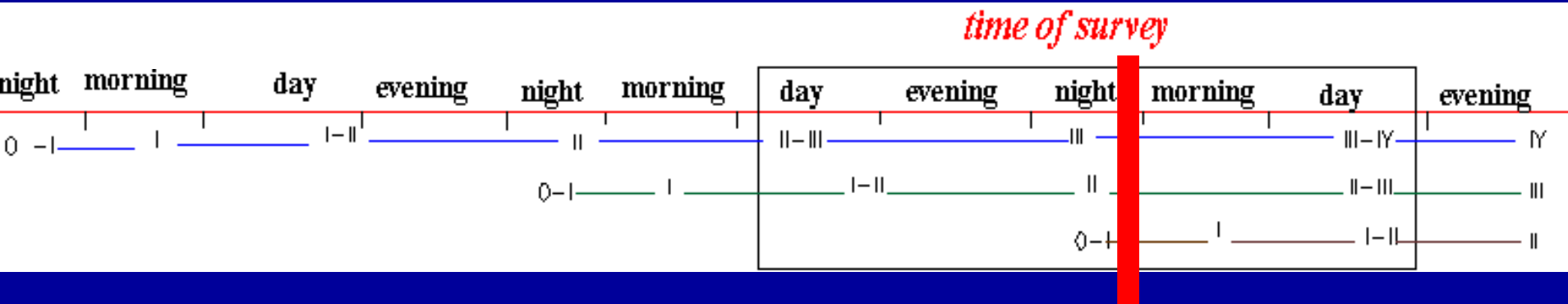
The final survival  $S = s_{0-I}^{t_1} * s_{II-IV}^{t_2}$ , where  $t_1, t_2$  – are the duration of the stages 0-I and II-IV.

In aquarial experiments, the duration depends on water temperature:

In the time of our experiments, the mean SST was 16.4 °C in June and 18.1 °C in July. According to aquarial experiments, the total duration of embryogenesis in these conditions is 70-80 hours in June and 60-70 hours in July.

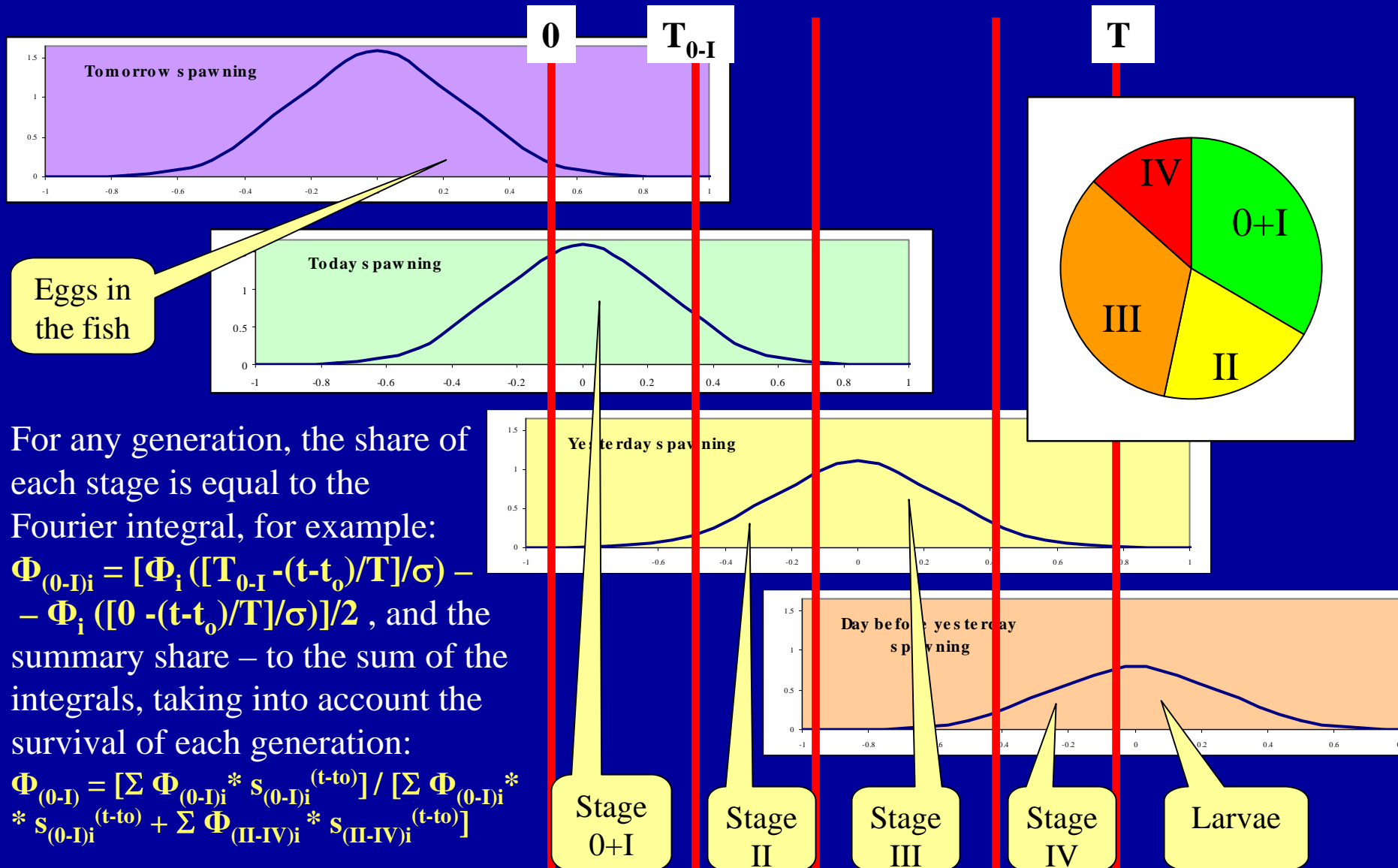


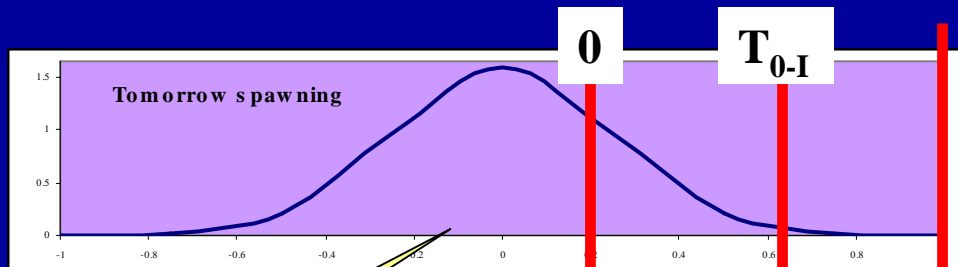
In natural conditions, the duration of embryogenesis could not be defined visually, so far as it is longer than the time interval between portions of new spawned eggs (> 1 day) – that's why several generations of the eggs are presented in samples in any time



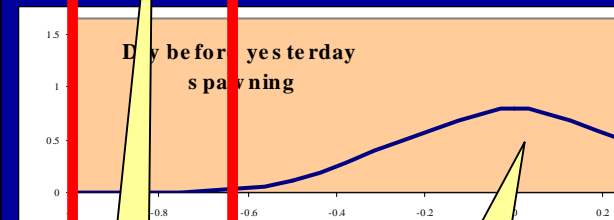
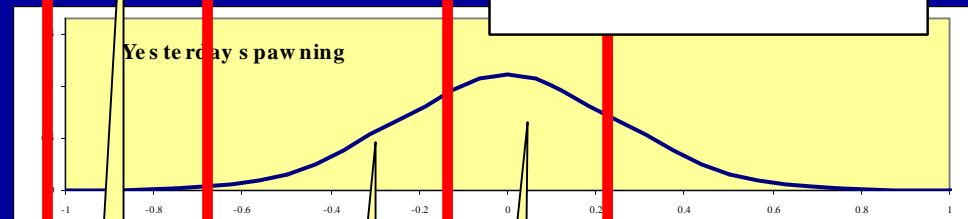
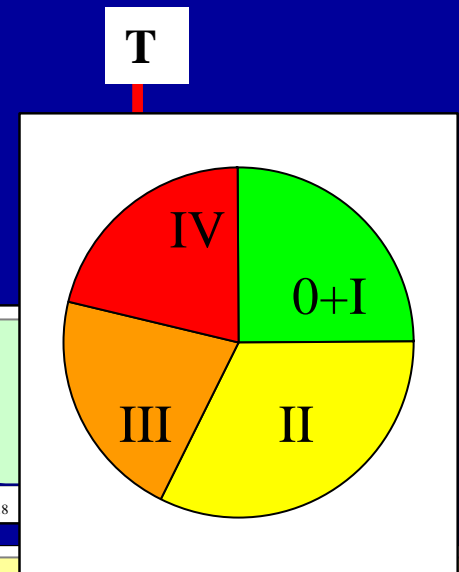
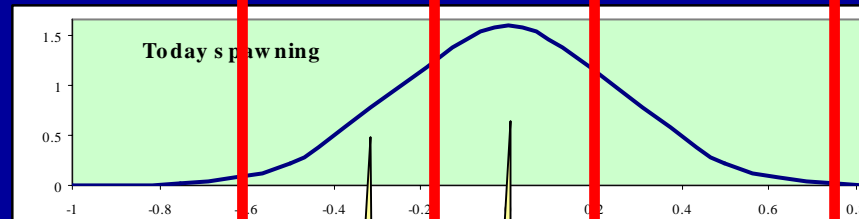
Each generation is presented with the certain stage dominating at the moment, so all stages are in the sample in any time. However, their ratio changes regularly that shows the regularity of the eggs development within a day.

We try to model the each generation development as Gauss distribution function movement along the time axis





Eggs in the fish



Stage  
0+I

Stage  
II

Stage  
III

Stage  
IV

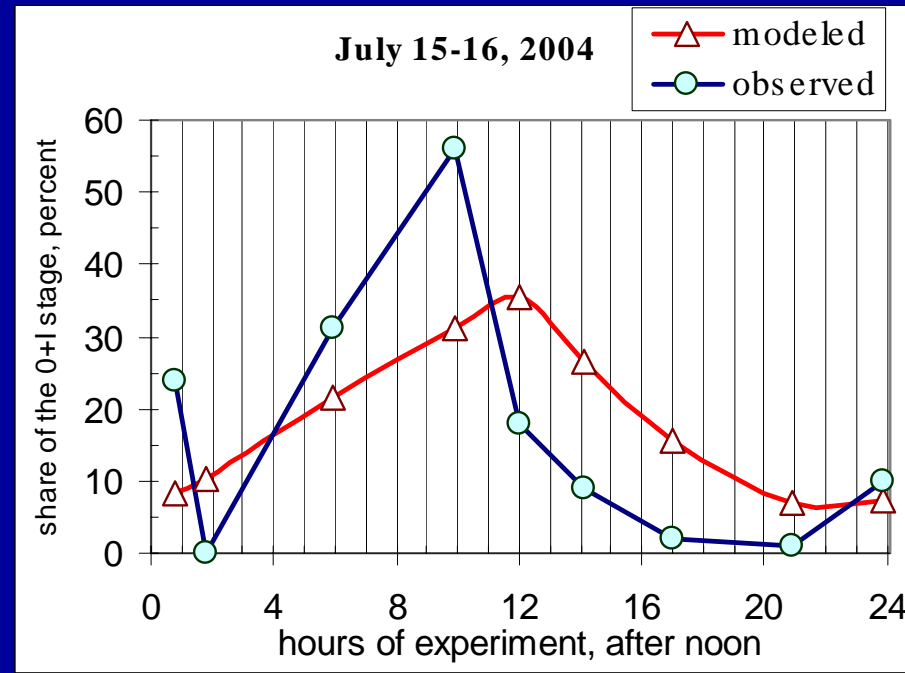
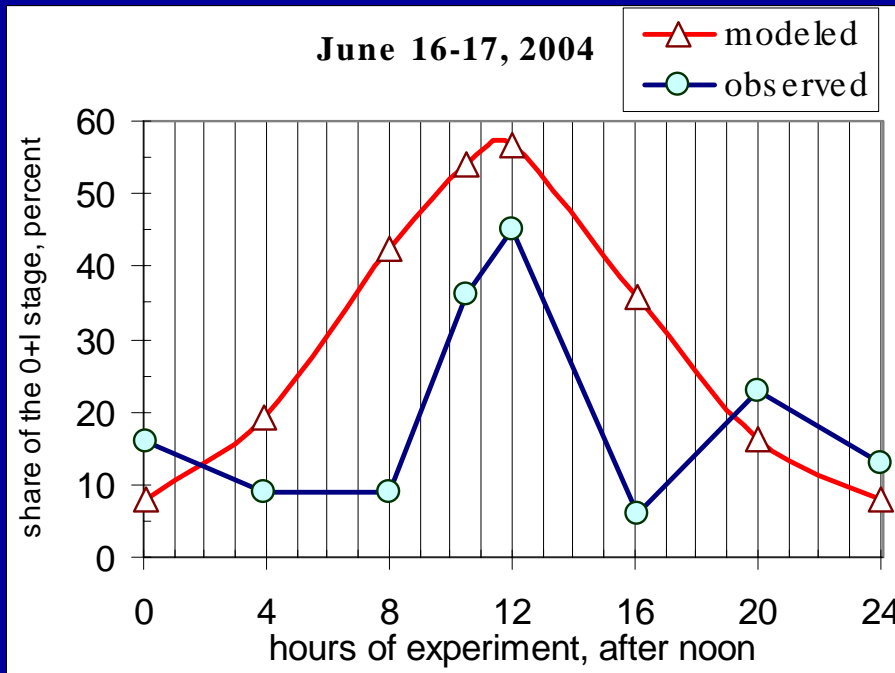
Larvae

The model parameters are:  
 $\sigma^2$  – dispersion;  
**T** – total duration of embryogenesis;  
**T<sub>0-I</sub>** etc. – relative duration of each stage.

The model was tuned to the data of observations, using the last squares method.

# Results of modeling in compare with observations

Modeled and observed share of the 0+I stage



The model  
parameters:

$$\sigma = 0.07; \quad T_{0-I} = 0.12$$

$$T = 76 \text{ hours}$$

$$t_1 = 9.1 \text{ h}; \quad t_2 = 66.9 \text{ h}$$

$$\sigma = 0.10; \quad T_{0-I} = 0.07$$

$$T = 66 \text{ hours}$$

$$t_1 = 4.6 \text{ h}; \quad t_2 = 61.4 \text{ h}$$

The total survival of the anchovy eggs  $S = s_{0-I}^{t1} * s_{II-IV}^{t2}$  :

The survival on the stage 0-1 is **0.13** in June and **0.45** in July

The total survival is **0.13** in June and **0.04** in July

Because of low survival, the eggs on stage IV were not numerous both in June and July. The losses occur mostly during early embryogenesis, that's why dead eggs on stage IV were absent in our samples.

# Conclusions

1. Mass spawning of japanese anchovy occurs in midnight, 95% of the eggs are spawned within 10-12 hours (the  $2\sigma$  period)
2. The eggs mortality is higher on the early stages of embryogenesis (0-I): 16-20% per hour, than on the late stages (II-IV): 0-4% per hour
3. The total time of the eggs development at SST 16-18°C is 66-76 hours; it obviously has a negative dependence on water temperature
4. The total mortality of anchovy during its embryogenesis is estimated as 87-96%; it occurs mainly in early embryogenesis; any dependence of the mortality on water temperature is not revealed.

## Future plans

- to evaluate the eggs mortality dependence on environmental conditions (water temperature, salinity, water column stability, wave turbulence) and other possible factors, as grazing.

*Thank you for your patience!*

