



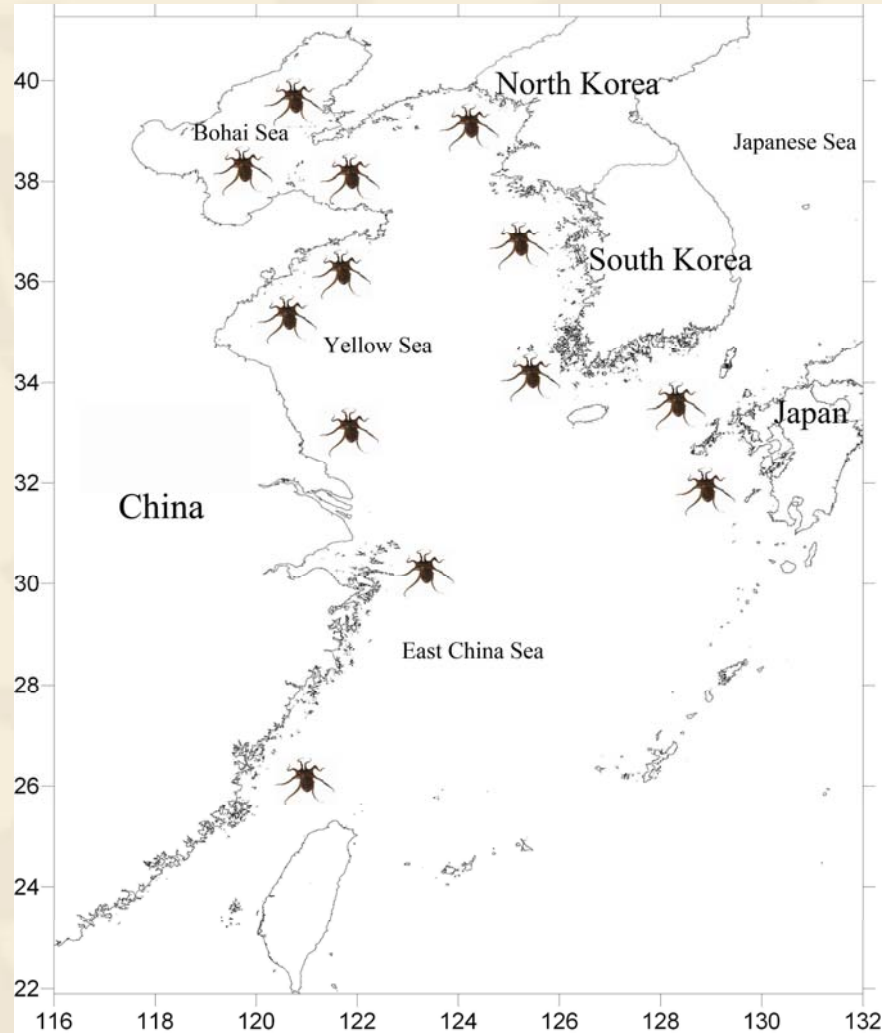
# **Studies on Reproduction and Population Genetic Diversity of *Octopus ocellatus***

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# Distribution of *Octopus ocellatus*



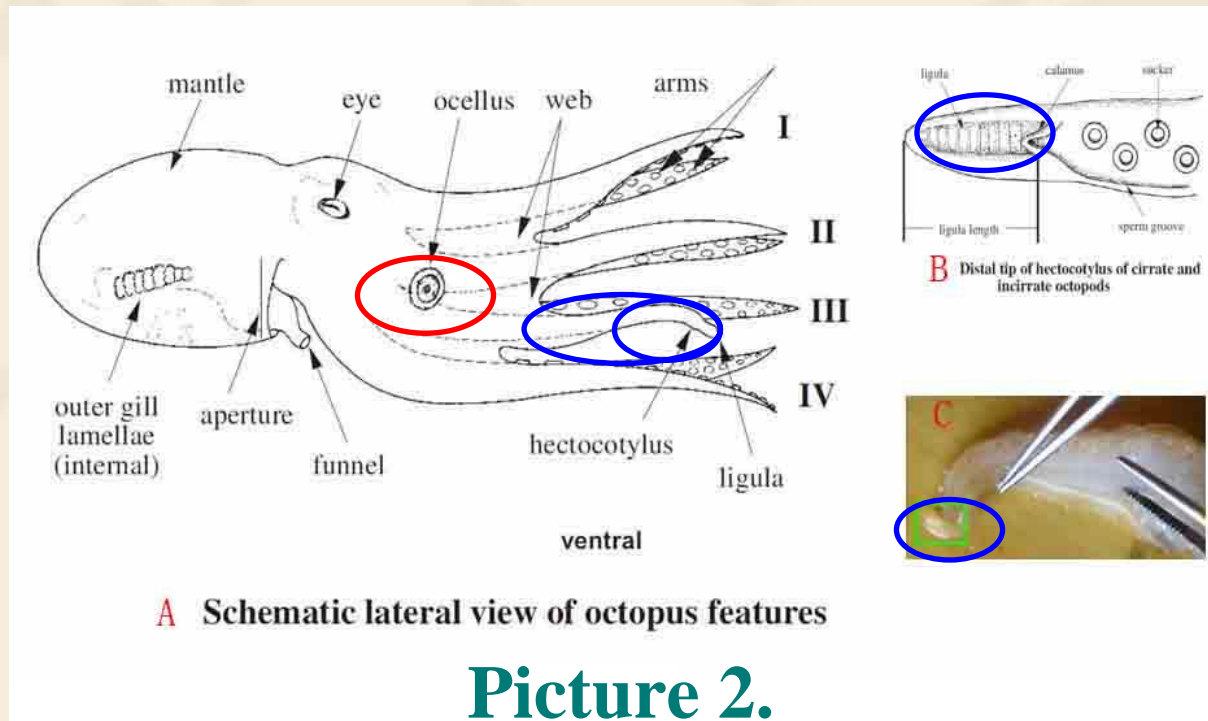
Picture 1.

# Background

- *Octopus ocellatus* is tasty, highly nutritive and rich in Arginine.
- mainly export to Korea and Japan.
- Life cycle is short, and growth very quickly.
- Be fit for culture and stock enhancement.
- resources reduced rapidly on account of intensive catching.

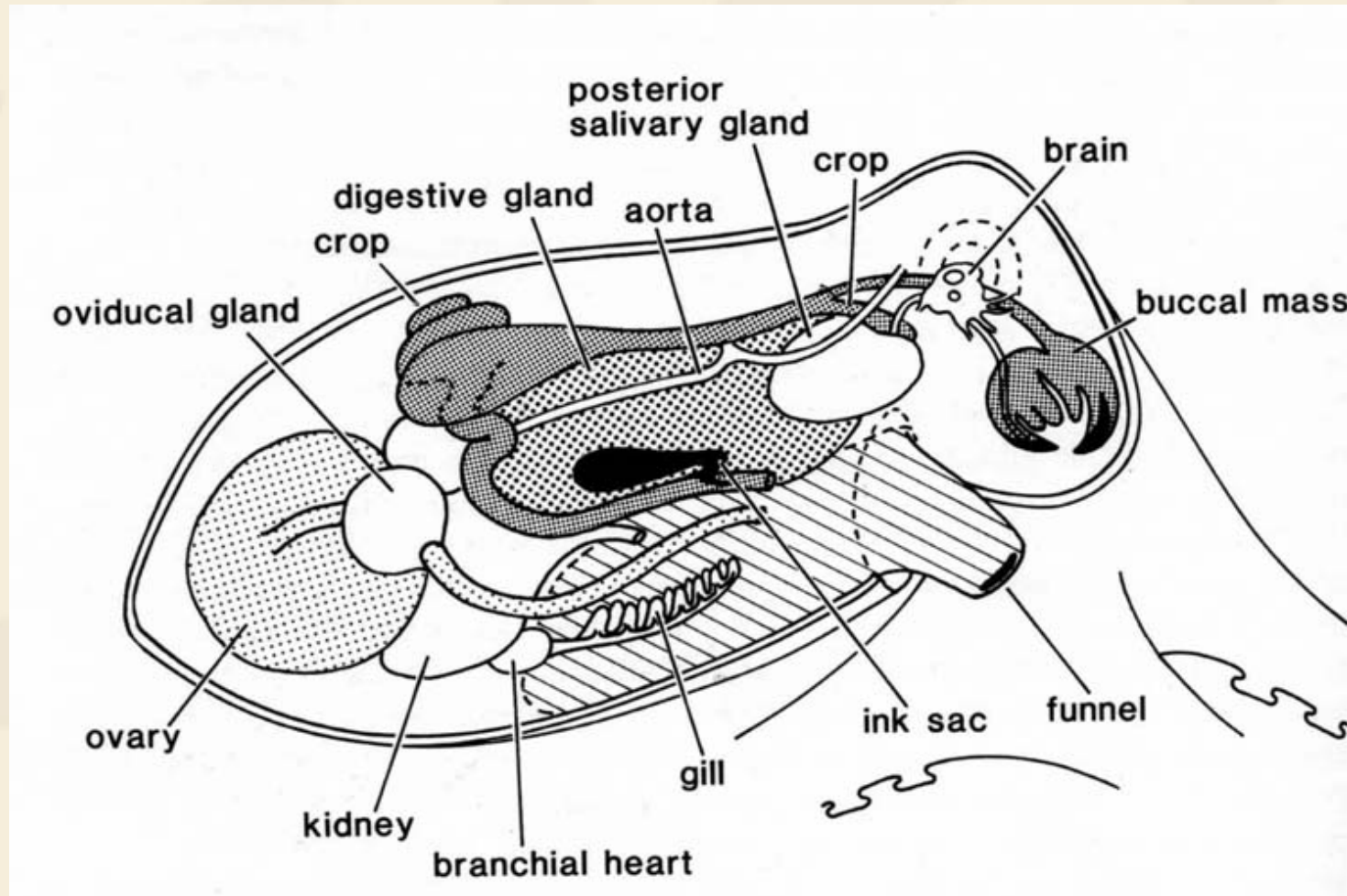
# Features of *Octopus ocellatus*

- **Hectocotylus:** The third right arm of the male, which had specialized into hectocotylus with taper ligula .
- **Ocellus:** Elliptic golden rings.



**Picture 2.**

# Generalized Anatomy of Female



Picture 3.

# Living Environment and Life Habits

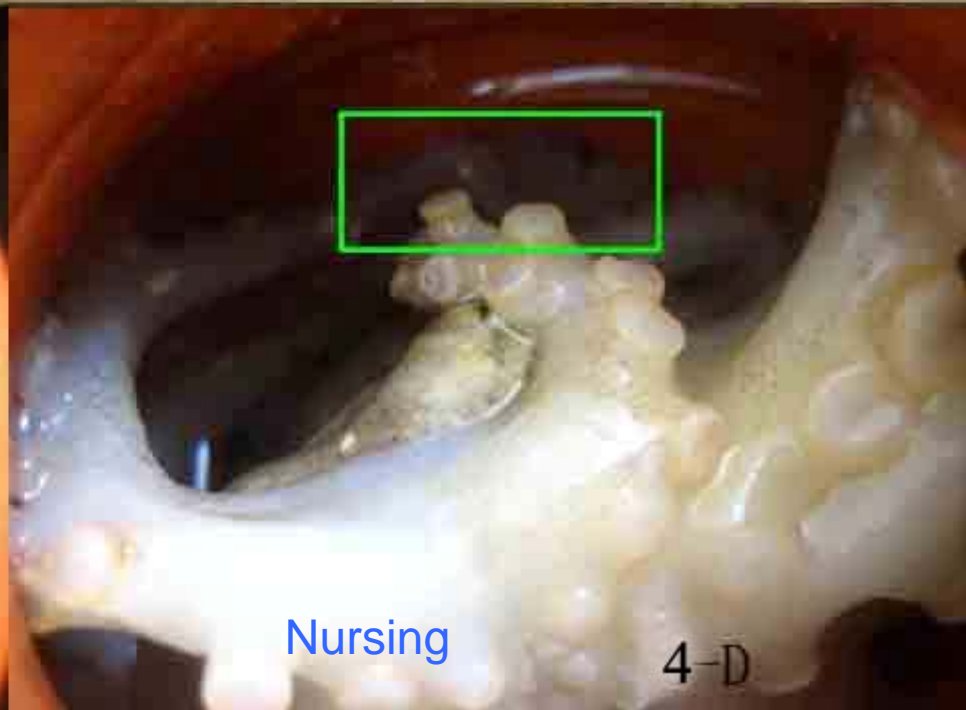
- ❖ They lived in the depth from 5-20m.
- ❖ The optimal temperature for *Octopus ocellatus* ranged from 13 to 26 °C.
- ❖ They were mainly benthic and gregarious in sea areas in the temperate zone by north, having a liking for troglodytism and preying clams and little crabs.
- ❖ When the temperature was below 10°C, they would reduce food intake, if it was below 6°C, they would neither move nor prey.

# Breeding Behaviour

- ✓ **Berried stage:** Along coastwise of shandong peninsula , the berried stage was from the last ten-day of April to the beginning of June. The optimal temperature was 11-14°C.
- ✓ **Mature:** The male could physiological mature for about 6 months, but female was used at least 10 months.
- ✓ **Mating season:** *Octopus ocellatus* breed once a year, and might have two breeding group.
- ✓ **Shell-dwelling:** In natural area, they were mating and breeding at 5-20m depth seabed, and shell-dwelling behavior was particularly evident in mating season.
- ✓ **Territory:** At mating season, the males had distinct territorial behavior, and frequently fought for the mating opportunity. (picture4-A、 B).

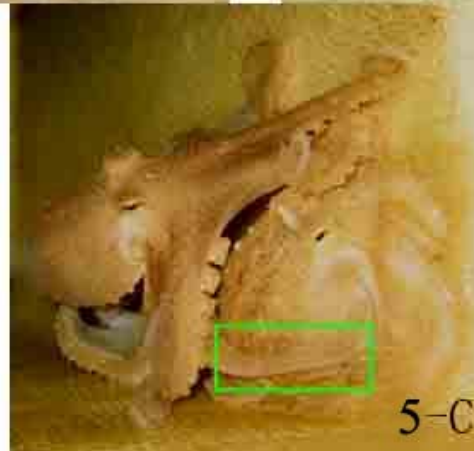
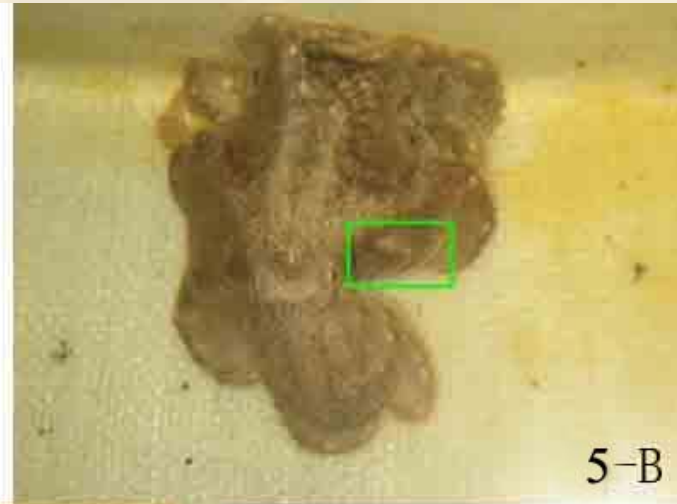
# Breeding Behaviour

- ✓ **Spawned eggs:** were appeared similar to cooked rice, winding together by the handle of the eggs like strings of grapes. (Picture4-C).
- ✓ **Food intake:** Before berried stage, food consuming was induced, and after breeding, it was no more food taking.
- ✓ **Nursing:** Female was taking care of eggs during the whole hatch stage (Picture4-D).
- ✓ **Rate of hatching:** was low, about 40-60%.
- ✓ **Mating positions:** 3 styles (Picture5).



# Breeding Behaviour

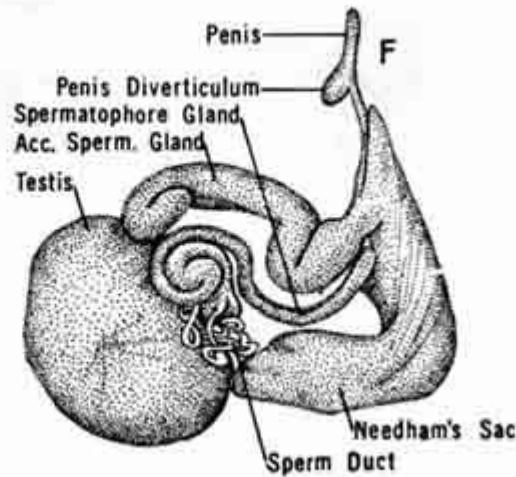
## -Mating Positions



# Reproductive System

## Anatomy-Male

- ▶ Male reproductive tract located in mantal cavity, composed of testis, sperm duct, Needham's sac, spermatophoric gland, penis *etc.*



**B**



**Picture 6.**

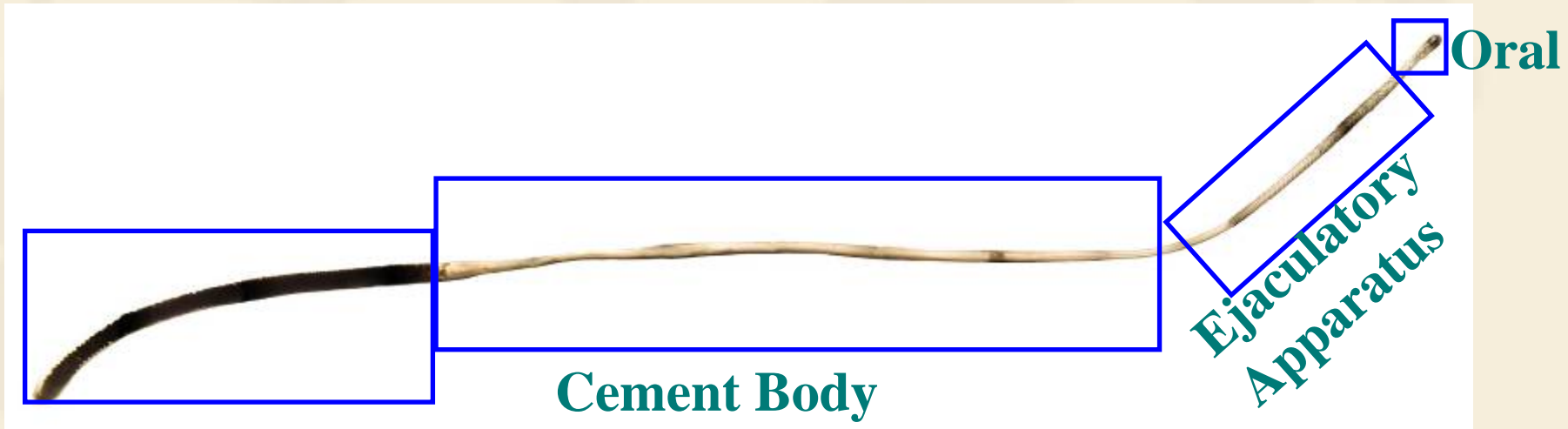
(Needham's sac)

# Reproductive System Anatomy-Male



Whole length of  
Spermatophore: 32-35mm

# Spermatophore

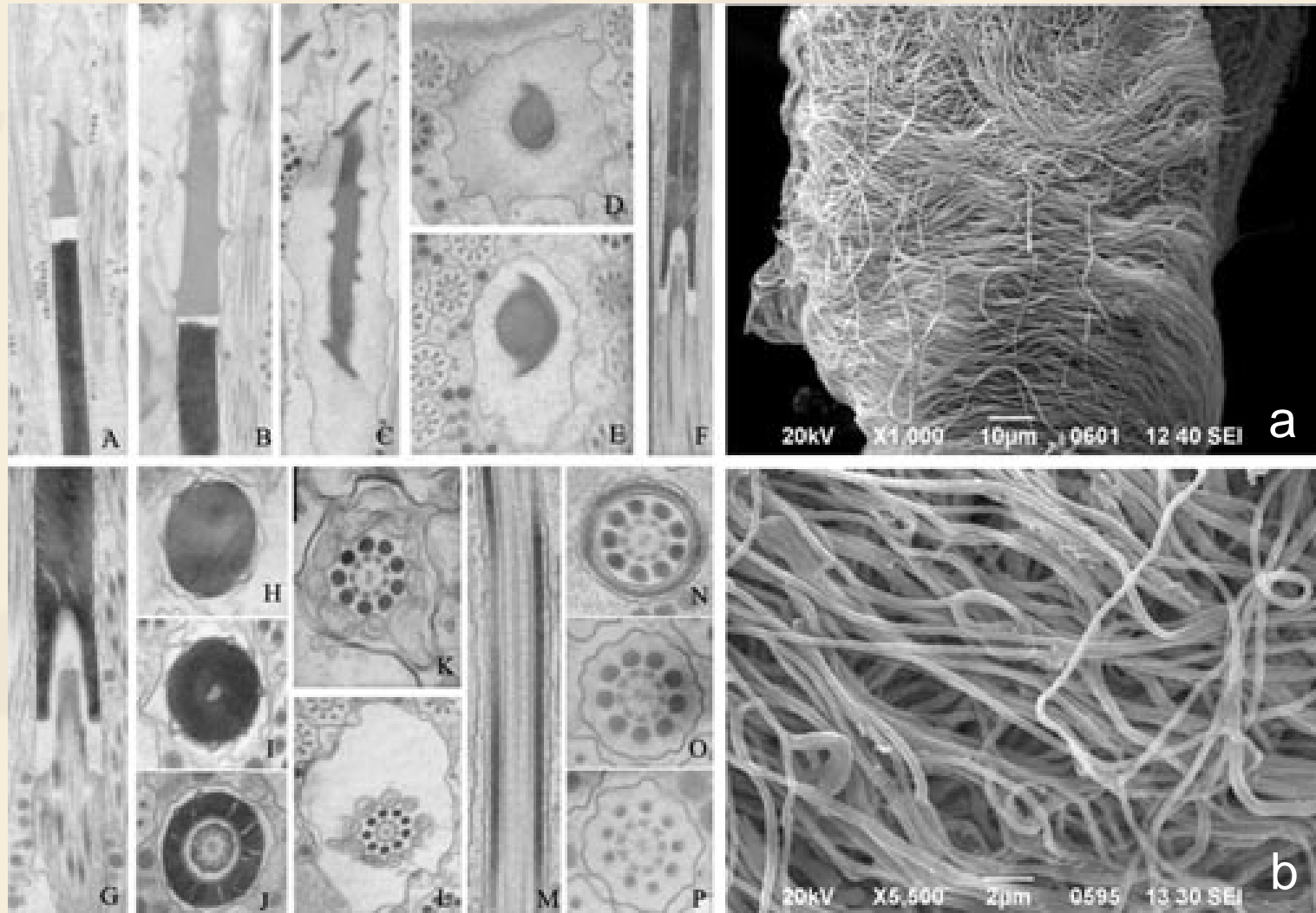


Sperm Mass/Cord

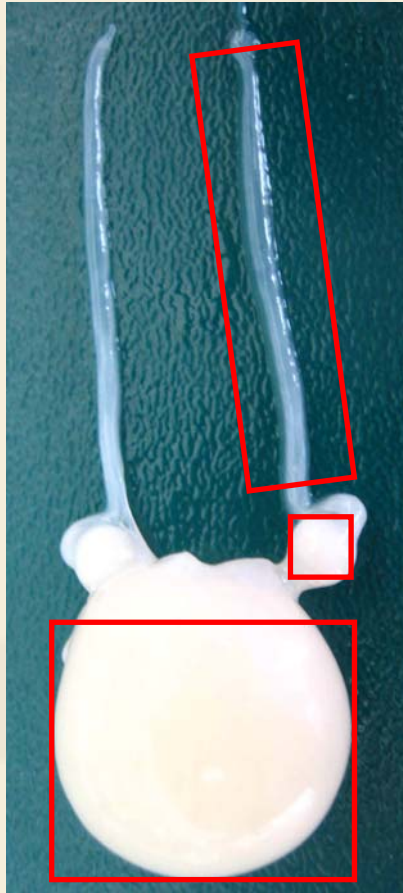
Sperm observed from microscope



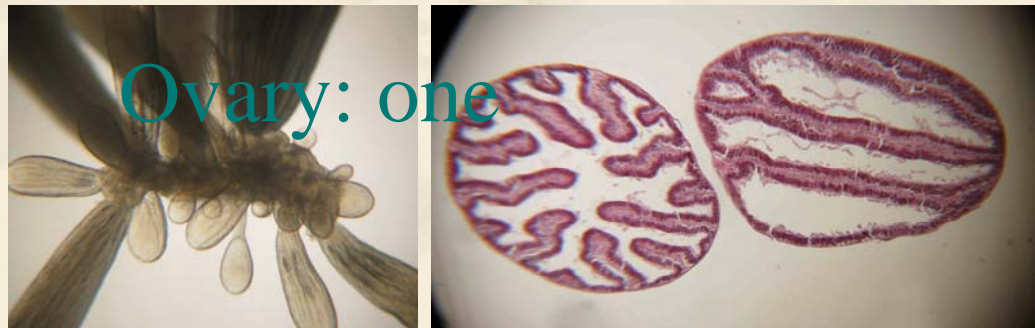
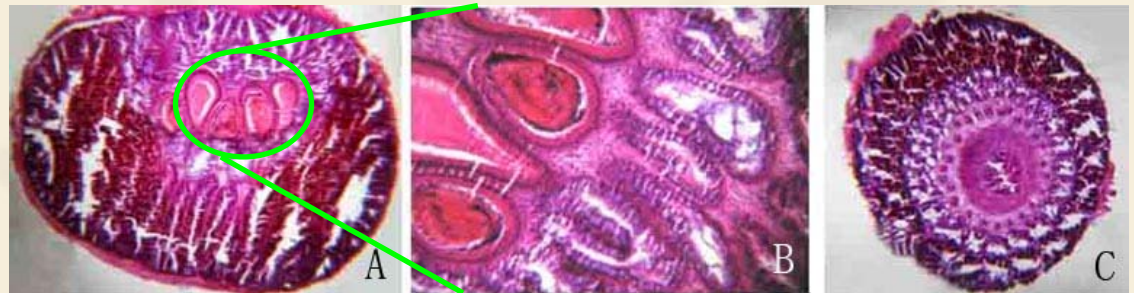
# Ultrastructure of Mature Sperm



# Reproductive System Anatomy-Female



Distal oviduct: double

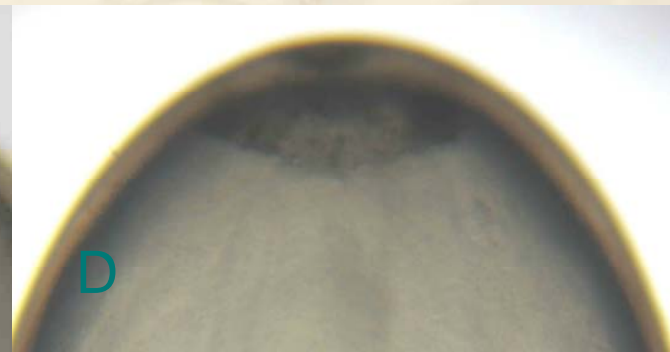
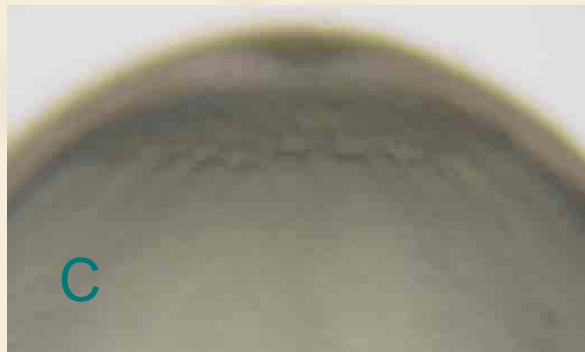
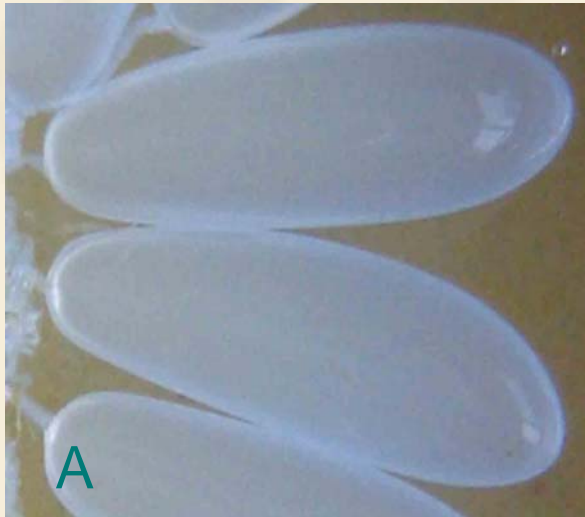


Ovary: one

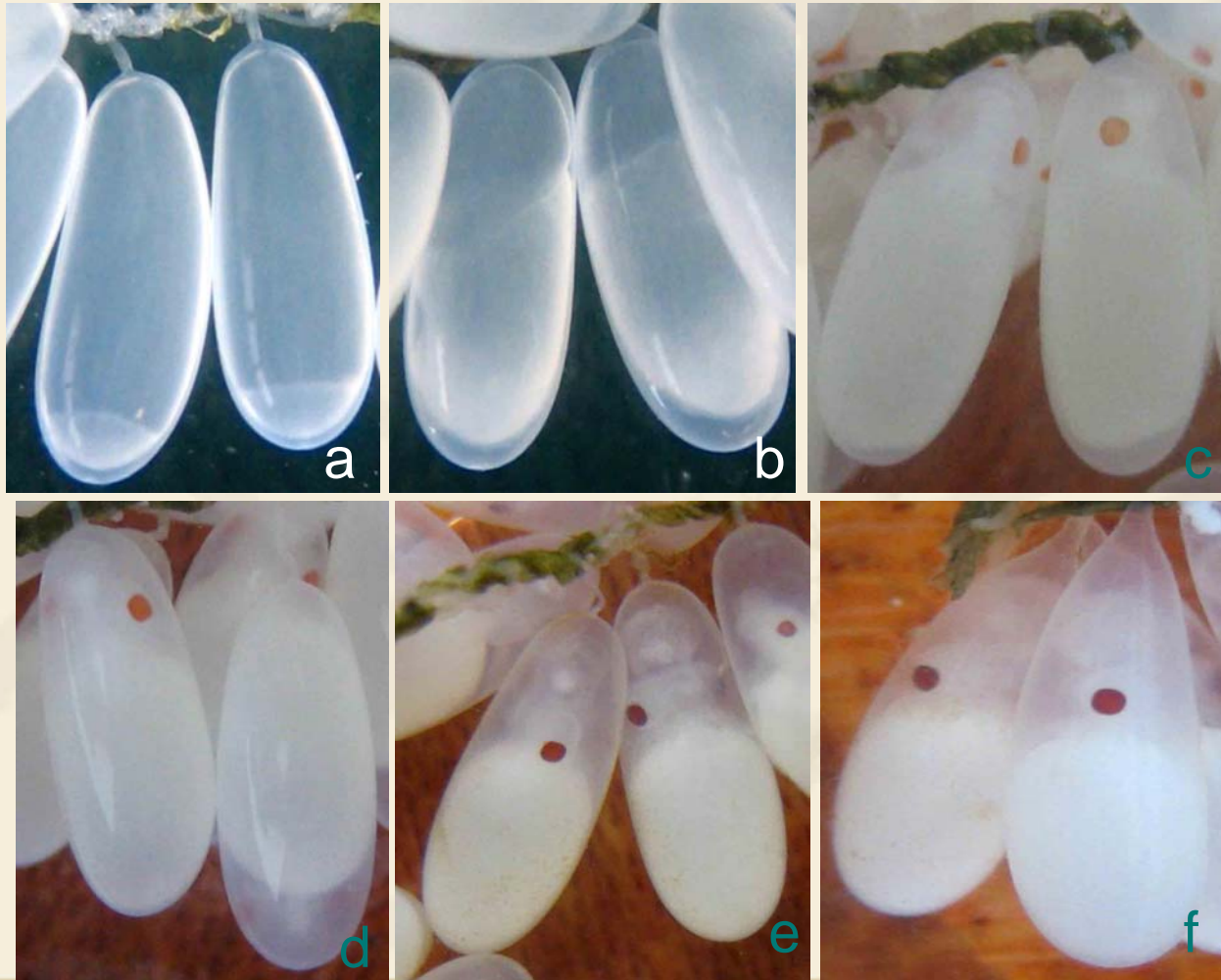
# Fecundity Ability

- ✓ Brood Amount: Range from 230 to 710, it was positive correlation with weight. The female of 130g could brood about 700 eggs.
- ✓ Average fecundity of each was about 512 eggs.
- ✓ Egg Diameter: Major axis of each mature egg was 5.25-5.75mm, and minor axis was 2.25-2.32mm.

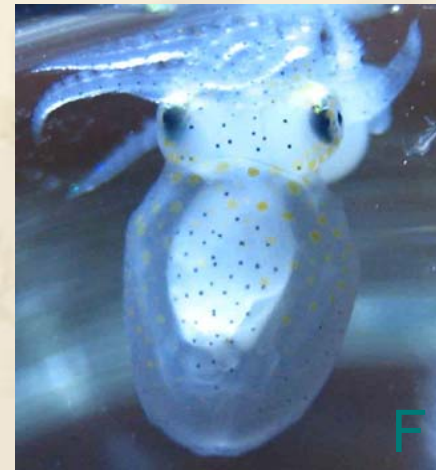
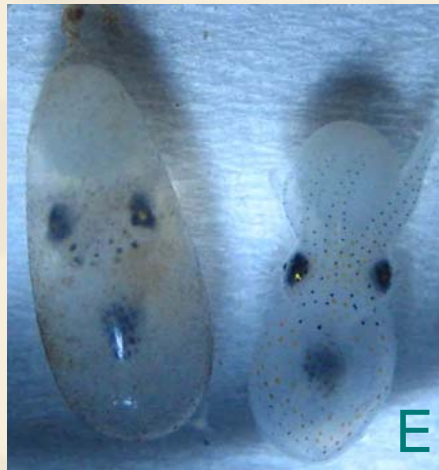
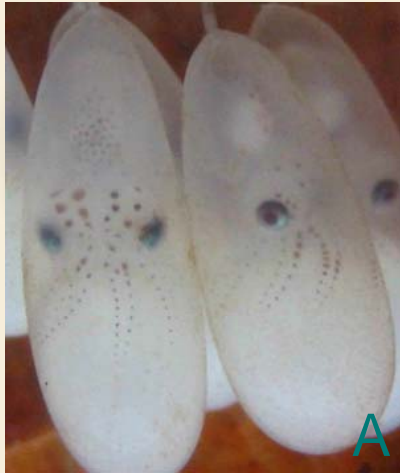
# Embryonic Development



# Embryonic Development



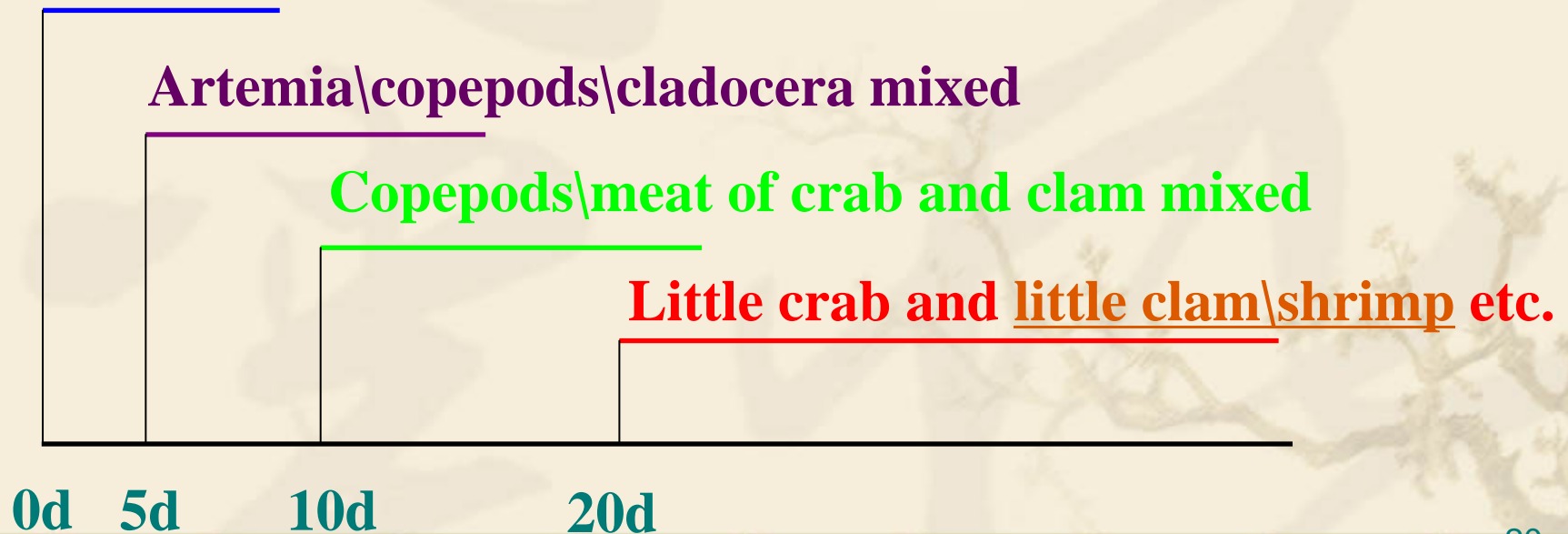
# Embryonic Development



# Behavior at Juvenile Stage

- **Proper salinity: 23-29(25-29)**, when it was below 20, death rate was markedly increased.
- **Nest: Shuck\whelk shell\clam shell**
- **Food: Brine shrimp nauplii\Copepods\cladocera\little shrimp\crab\clam**

## Brine shrimp nauplii





# Cannibalism during Juvenile Stage

- ✓ To the same size larvae, the small ones was higher than the big ones on daily cannibal rate.
- ✓ The period which was the most serious cannibalism was at average weight 0.03~0.08g, daily cannibal rate was much lower when the average weight came over 0.23g.

Group	Average Weight (g)	shelters	Number	Stat. after 15d		
				Number of Cannibalism	Rate of Cannibalism (%)	Daily Rate of Cannibalism (%)
①	0.03 ± 0.01	Y	10	3	30.00	2.00
②		N	10	8	80.00	5.33

# Factors of Affecting Cannibalism

- ✓ Density
- ✓ Food
- ✓ Nests
- ✓ Temperature
- ✓ Size difference

# Measures

- ✓ Suitable nests
- ✓ Proper food
- ✓ Lower culture density
- ✓ Temperature (below 24 °C)
- ✓ Reducing the size difference.

# Artificial Culture Techniques

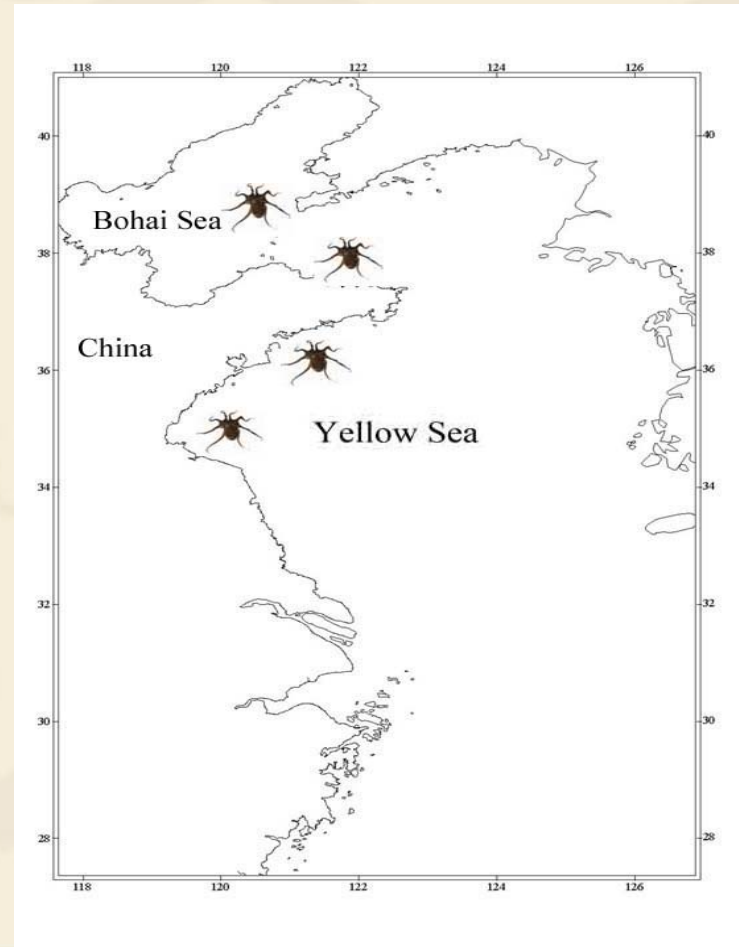
- **Temperature Control: 13-26 °C, when below 10 °C, food taking was induced.**
- **Building nest: PVC pipe\ small gallipot.**
- **Density: In the conditions of abundant of nests, 10-20/m<sup>2</sup>.**
- **food: Lively clam and little crab.**
- **Growth of speed: It can reach 60g for about 6 month.**
- **Control of cannibalism: No more than 10/ m<sup>2</sup> and abundant of food and nests.**



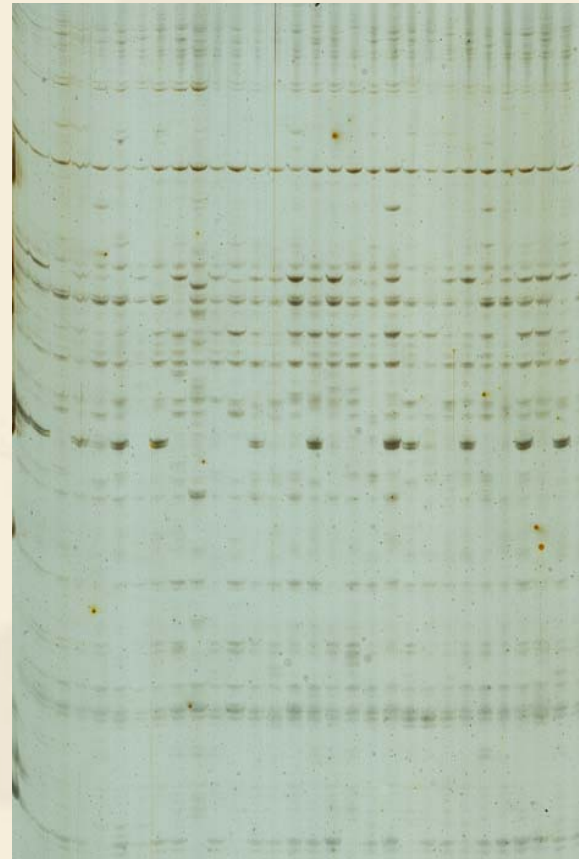
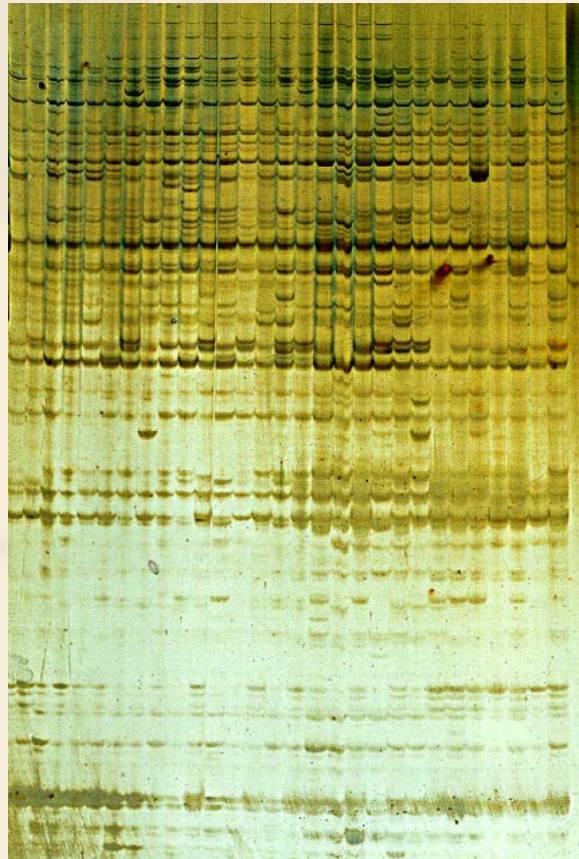
# Genetic Diversity on Four Different Geographical Group

❖ **Material:**  
DaLian\YanTai\  
QingDao\LianYunGang

❖ **Method: AFLP**



# Amplified Results of Primer Combination of E44M60 and E35M58



# Population Analysis of Genetic Diversity

Groups	Rate of Poly Loci P(%)	Nei's Index (H)	Shannon Index (I)	No. of Available Allele (Ne)
DaLian (DL)	67.93 %	0.2617	0.3863	1.4520
YanTai (YT)	62.87 %	0.2353	0.3497	1.4017
QingDao (QD)	62.03 %	0.2389	0.3530	1.4105
LianYunGang (LY)	64.56 %	0.2482	0.3675	1.4228

# Genetic Distance and Genetic Comparability

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	DL	QD	YT	LY
DL	****	0.9184	0.9515	0.9238
QD	0.0851	****	0.9358	0.9335
YT	0.0497	0.0663	****	0.9319
LY	0.0793	0.0689	0.0705	****

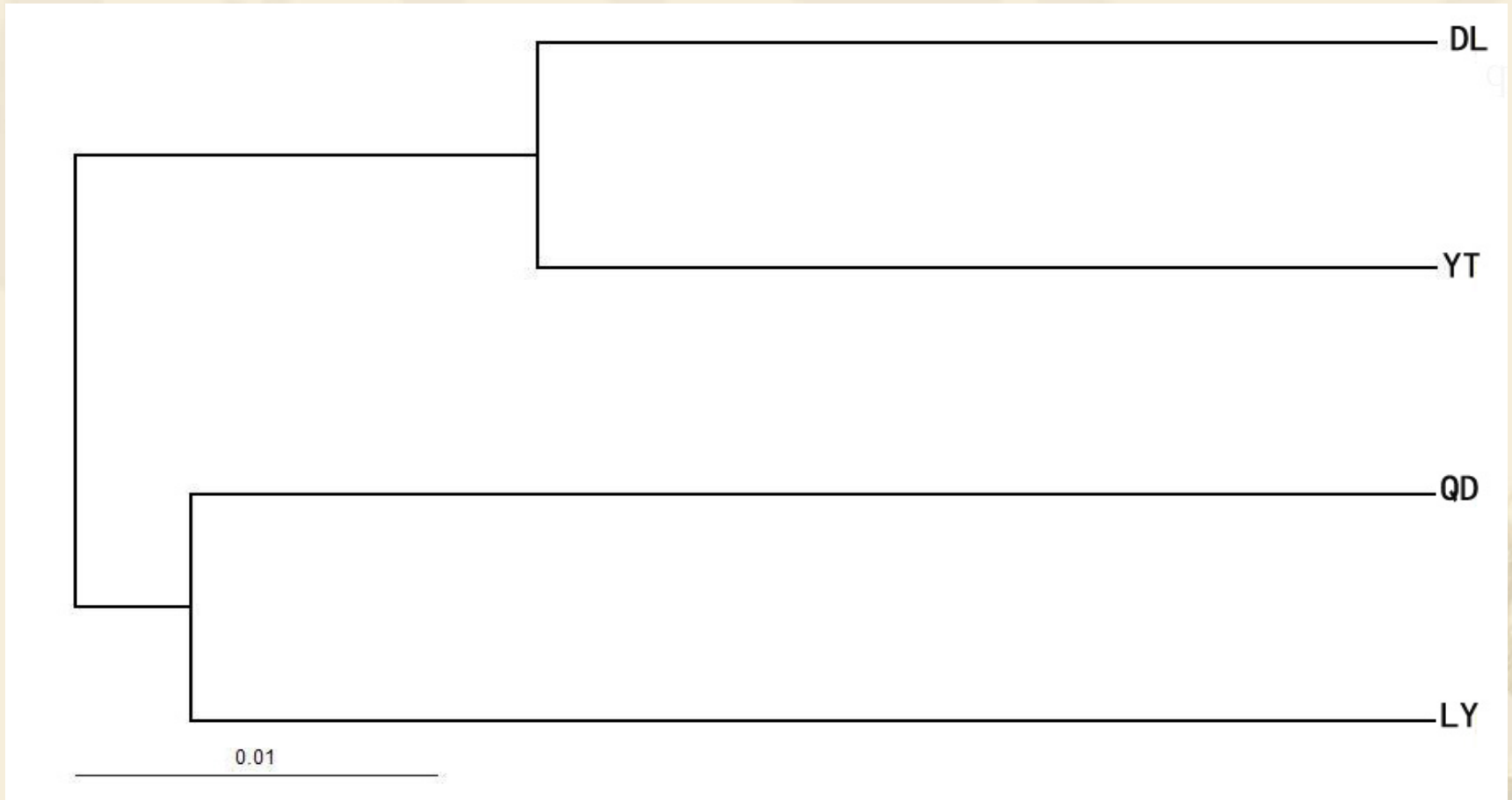
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Genetic Comparability (above diagonal) and  
Genetic Distance (below diagonal)

# Population Genetic Differentiation (AMOVA Analysis)

Source of Variance	df	Sum of Quares	Variance Components	Percentage of Variance (%)
Among Populations	3	427.108	3.79345 $V_a$	<b>11.72</b>
Within Populations	116	3313.638	28.56580 $V_b$	88.28
Total Variation	119	3740.742	32.35926	

# Population Clustering



# Prospect

- ❖ To develop artificial feed .
- ❖ Large-scale pond aquaculture technology.
- ❖ overwintering technique.
- ❖ Research on stock enhancement *Octopus ocellatus*, to complement the amount of natural resources.
- ❖ To develop molecular markers.



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所战略

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Thanks a lot

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