# Ecological factors affecting on eggs development and life span of meracidia of Fasciola gigantica

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### **Abstract**

This study was carried out during the period from February to August 2008 at the laboratories of Biology dep. ,College of Education , University of Karbala. The study involved the effect of temperature, sodium chloride, and pH on the period and the percentage of eggs development. The study involved also the effect of previous factors on the life span of meracidia . The percentage of developed eggs and the period of eggs development decreased significantly when the temperature increased. The optimal pH value for eggs developing was pH 7 while the developing declines significantly when pH less and more than pH 7. The study showed also that eggs developing declines significantly when sodium chloride increased.

Concerning of the life span of meracidia, the study revealed a significant decrease in the life span with temperature and sodium chloride increasing. The study showed also that the best pH value for the lifespan of meracidia was 7, while it declines significantly when pH less and more than 7.

#### الخلاصة

أجريت الدراسة الحالية خلال المدة من شباط إلى آب 2008 في مختبرات قسم علوم الحياة / كلية التربية / جامعة كربلاء والتي تضمنت تأثير درجة الحرارة وتركيز كلوريد الصوديوم و الاس الهايدروجيني في فقس بيوض و حيوية مهدبات طفيلي Fasciola gigantica ، إذ أظهرت الدراسة انخفاضاً معنوياً في المدة الزمنية لفقس البيوض و نسبة فقسها بارتفاع درجات الحرارة ، ثم لوحظ بان الاس الهايدروجيني الأمثل لفقس البيوض كان PH7 بينما ارتفعت المدة الزمنية لفقس البيوض وانخفضت النسبة المئوية للفقس وبصورة معنوية عند أعلى وأدنى منه ، وبينت النتائج ارتفاعًا معنويًا في المدة الزمنية الفقس بيوض و سربة فقس البيوض وانخفضت وانخفاض في نسبة الفقس بزيادة تركيز كلوريد الصوديوم .

أما بالنسبة للمهدبات فقد لوحظ انخفاض معنوي في مدة حياتها بارتفاع درجات الحرارة و زيادة تركيز كلوريد الصوديوم ، وكان الاس الهايدروجيني الأمثل للمهدبات PH7 إذ سجلت فيه أطول مدة حياة بينما قصرت مدة الحياة وبصورة معنوية عند أدنى وأعلى من هذا الاس.

\*البحث مستل من رسالة ماجستير للباحث الأول

#### **Introduction**

Eggs of Fasciola gigantica are considered as the largest among digeneae eggs. They are oval in shape, have an operculum in one end, and  $(150-197) \mu$  length and (90-140) width (1) . Eggs of Fasciola hatch in the range of (10 - 35) C° (2). They hatch in (10-11) days at 35  $C^{\circ}$ , (12-24) days at  $25C^{\circ}$  and 33 days at 17 C°. (4) mentioned that eggs developing stopped above 37 C° and bellow 10 C°. It was found also that concentration of pH and sodium chloride affect the developing of eggs (4). pH 8 delates the developing of eggs (5). Experiment carried out by (6) showed that the light has an important role in the activation and hatching of eggs. The light activate hatching enzymes found in cement layer of the egg operculum (7). It was noted that the different in osmotic pressure between inside and outside of eggs lead to open the operculum of eggs (8)

Eggs of *F. gigantica* hatch into ciliated larva called meracidia which are broad anteriorly and tapering posteriorly to a blunt end. The cuticle is ciliated, and there is an anterior papilliform protrusion and a pair of darkly staining eye spots visible near the anterior end of the body (9). Meracidium liberates from the egg into water then swim until it get appropriate intermediate host otherwise it would die if she couldn't find the appropriate snail within 24 hours as a result of exhausting its storaged energy (10) . Meracidium attracted to the appropriate snail by chemotaxis produced by the snail (11) and (12). Meracidia have more epidemiological importance than other parasite life cycle stages especially for trematodes which use snails as intermediate hosts. Among the ecological factors that affect the mortality of *F. gigantica* meracidia , are water , temperature and pH (13)

## Materials and methods

Eggs of *Fasciola gigantica* (picture 1) were obtained from gall bladders of slaughtered cattles in the abattoir of Karbala city. The eggs were washed several times by distilled water. They were identified according to (14), then three experiments were carried out :-

First experiment : 105 eggs were distributed to seven (10 ml sized) flasks with river water in every flask 15 eggs, the seven flasks were kept then at different temperature degrees (10, 15, 20, 25, 30, 35, 37) C°, respectively.

Second experiment : 120 eggs were distributed to eight (10 ml sized) flasks with different pH water (5, 5.5, 6, 6.5, 7, 7.5, 8, 8.5), respectively, in every flask 15 eggs, all at 25 C°.

Third experiment : 105 eggs were distributed to 7 (10 ml sized) flasks with different percentages of sodium chloride water (distilled water 0.00001%, river water 0.05%, 0.2%, 0.4%, 0.6%, 1%, 1.5%), respectively, in every flask 10 eggs the seven flasks were kept at 25 C°.

## Meracidia

Eggs of *F. gigantica* were incubated at  $25C^{\circ}$  for 17 days in dark then they were exposed to strong light to stimulate meracidia liberation (15) fresh meracidia (picture 2) were used in three experiments as following :-

First experiment : 60 meracidia were distributed to six (10 ml sized) flasks with river water in every flask 10 meracidia , the seven flasks were kept then at different temperature degrees (10, 15, 20, 25, 30, 35)  $C^{\circ}$ , respectively .

Second experiment : 80 meracidia were distributed to eight (10 ml sized) flasks with different pH water (5, 5.5, 6, 6.5, 7, 7.5, 8, 8.5), respectively then the flasks were kept at 25 C°.

Third experiment : 60 meracidia were distributed to six (10 ml sized) flasks with different percentages of sodium chloride water (distilled water 0.00001%, river water 0.05, 0.2 %, 0.4 %, 0.6 %, 1%, 1.5%), respectively in every flask 10 meracidia all at  $25C^{\circ}$ .

Statistical analysis was performed using an (ANOVA) by excel.



Picture 1 eggs of *Fasciola gigantica* 100X



Pictures 2 meracidium of *F. gigantica* 400X

## **Results**

Table 1 show that rates of eggs hatching period and percents of hatching were inversely related ( $p \le 0.05$ ) to the temperature degree . Shortest hatching period was 9.12 days and less percent of hatching was 53.3% at 35 C°, while the longest rate of hatching period was 44.5 days and highest percent of hatching was 86.6% at 15 C° , but the developing of eggs stopped completely at 10 C° and 37 C° .

Concerning the effect of pH on eggs development, the table show that the shortest rate of egg hatching period was 17.5 days and highest hatching percentage was 86.6 % at pH7, while the longest period was 30.5days at pH 8 and less percent of hatching was 33.3 days at pH 5.5 while developing stopped completely at pH 5 and pH 8.5. Table 1 show also that rates of eggs hatching period directly related and the percent of hatching inversely related ( $p \le 0.05$ ) to the sodium chloride of water. Shorter hatching period was 17.1 days and highest percent of hatching was 86.6% with distilled water . but the developing of eggs stopped completely at 1.5 %.

Table 2 show the effect of temperature , pH , and sodium chloride on the life span of meracidia . Lifespan of meracidia was inversely related ( $p \le 0.05$ ) to the temperature degree, 20.3 , 25.1 , 18.45 , 8.80 , 4.1 , and 2.05 hours at  $10C^{\circ}$  ,  $15C^{\circ}$  ,  $20C^{\circ}$  ,  $25C^{\circ}$  ,  $30C^{\circ}$  , and  $35C^{\circ}$  respectively . Concerning the effect of pH , the table show that the longest life span was 16.9 hours at pH7, while it declined bellow and above pH7 (P<0.05) . lifespan of meracidia also inversely related ( $p \le 0.05$ ) to sodium chloride, 19.7 , 18.1 , 7.3 , 4.7 , 2.05 , and 0.95 hours at 0.00001 , 0.05 % , 0.2% , 0.4 % , 0.6 % , 1% , and 1.5 % respectively.

	Temperature degree C°										Į	Н			Sodium chloride %								
	10	15	20	25	30	35	37	5	5.5	6	6.5	7	7.5	8	8.5	D.W. 0.00001	0.05	0.2	0.4	0.6	1	1.5	
Period of hatching /day	0	44.5	26.1	17.8	11.1	9.1	0	0	32	27.2	22.4	17.5	25.2	30.5	0	17.1	18	22.18	24.7	28.1	30.2	0	
Percent of hatching %	0	86.86	86	73.3	66.6	53.3	0	0	33.3	46.6	60	86.6	66.6	53.3	0	86.6	86.6	73.3	66.6	53.3	33.3	0	
	$2.9 = LSD_{0.05}$								$3.02 = LSD_{0.05}$								$2.37 = LSD_{0.05}$						

Table 1 Effect of temperature, pH, and sodium chloride on the period and the percent of F. gigantica eggs hatching

Table 2 Effect of temperature, pH, and sodium chloride on the life span of F. gigantica meracidia

		Temp	eratur	e degr	ee C°	_	pH								Sodium chloride %							
	10	15	20	25	30	35	5	5.5	6	6.5	7	7.5	8	8.5	D.W. 0.00001	0.05	0.2	0.4	0.6	1	1.5	
Lifespan of meracidium /hours	29.3	25.1	18.4	8.8	4.1	2.05	0	2	4.1	10.2	16.4	8.3	3.7	0	19.7	18.1	7.2	4.7	2.05	0.95	0	
		1	.62 = I	$LSD_{0.0}$	5			$1.69 = LSD_{0.05}$								$1.41 = LSD_{0.05}$						

#### **Discussion**

This study showed that the period of *F.gigantica* eggs hatching was inversely related to the temperature degree . The periods of hatching were 44.5, 26.16, 17.84, 11.14, and 9.12 days at  $15C^{\circ}$ ,  $20C^{\circ}$ ,  $25C^{\circ}$ ,  $30C^{\circ}$ , and  $35C^{\circ}$  respectively .These results are nearly agreed with (17), 12 days at  $28C^{\circ}$  and (18), 14 days at  $26C^{\circ}$ . The development of eggs stopped completely at  $10C^{\circ}$  and 37  $C^{\circ}$ , this result agree with (4), this may attributed to the inhibition of the activity of the egg enzymes at high and low temperature degrees .

The study proved also that the optimal pH was 7 for the shorter period and high percent of eggs hatching while the period of hatching was longer and the percent of hatching was less bellow and above pH7, this may attributed to the inhibition of enzymes of the egg at high and low pH.

Concerning the effect of sodium chloride on eggs hatching, our study showed that the period of hatching directly related to the sodium chloride of water, while the percent of hatching inversely related to the sodium chloride of water. The optimal sodium chloride for eggs hatching was with distilled water 0.00001%, this may attributed to the effect of osmotic pressure out side the egg on the operculum of the egg and this may cause delaying opening it, in this regard (8) found that the defference in osmotic pressure between inside and outside of the eggs cause opening the operculum of the egg and exiting the meracidium.

This study showed also that the life span of meracidia inversely related to the temperature degree .This may attributed to the raising of the meracidium activity at high temperature causing rapid exhausting of storaged energy (19) .

Concerning the effect of pH , This study showed that the optimal pH was 7 for longest lifespan 16.40 hours, while the life span of meracidia declined gradually bellow and above pH 7 . This result nearly agreed with (4) she said that the optimal pH was 7.2 , this may attributed to the effect of pH on the activity of meracidia enzymes .

The study showed also that the life span of meracidia was inversely related to the sodium chloride of water, this result agree with (20) he mentioned that sodium chloride shortens the lifespan of the meracidium. This may attributed to the large energy exhausted by the meracidium to resist the high difference in osmotic pressure between outside and inside the meracidium.

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