Material And Methods

ARTIFICIAL HATCHING OF SILKWORM EGGS AFTER CHILLING

by

The parent sillworm, *milsuM bammahuM naiM 1 and PAK 2, belonging to Japanese and Chinese breeds respectively, were incubated and reared under optimum

Research Laboratory, Labore. The male and lemale insects of the two varietiests

The silkworm eggs of locally developed F1 hybrid were laid in Spring 1989 at 26oC, stored at a temperature of 22 to 26oC for a period of 61 days and refrigerated at a temperature of 4 to 6oC for 93 days. These were treated subsequently with Hcl solution of different concentrations at 48oC for a period of 6 minutes for their artificial hatching in autumn. Treatment with 20% HCl solution was found to be most suitable for obtaining maximum useful hatchability of 92.3% under local conditions.

of eggs was maintained at 80-85%. A control group of nine englayinoitauborant replications of three layings each was maintained at room temperature of 18 to 27 oc. The

The hybernating eggs laid by univoltine or bivoltine races of silkworm undergo a state of dormancy called diapause and do not hatch till the following spring after their exposure to a low temperature of around 5oC for several months. In Pakistan, due to unfavourable hot summer season, the spring silkworm rearing is based on hybernating eggs. As the hybernating eggs can produce only one generation in a year, therefore, it is not possible to utilize the eggs, laid in spring, for autumn rearing. This limitation has been overcome by developing methods for artificial hatching of eggs (Karishnaswami, 1973). The most successful and rather the only method practised at present is the acid treatment of eggs. Silkworm eggs can be converted to non-diapausing state by treatment with hot hydrochloric acid just before or shortly after the onset of diapause (Tazima, 1978). In Japan about 56% of the cocoon production comes from summer and autumn crops for which eggs are subjected to acid treatment to produce artificial hatching (Takami, 1967).

For immediate hatching, the time suitable for acid treatment is about 20 - 24 hours after oviposition, whereas far the postponement of hatching by more than 40 days, acid treatment is performed after cold storage of eggs (Tazima, 1978).

Mulberry, leaves which constitute the food of silkworm, grow continuously from early spring to late autumn in Pakistan and hence the silk output can be increased substantially by introducing post-spring and autumn rearing through artificial hatching of eggs of the current hybernating races. The possibility of artificial hatching of silkworm eggs immediately after oviposition for post-spring rearing has already been investigated by treating them after 21 hours of oviposition, with 14.5% HCl solution at 46oC for a period of 5.5 minutes to obtain 98% useful hatchibility under local conditions. (Muslim, 1977). A investigation was carried out to find suitable concentration of hydrochloric acid (HCl)

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solution to affect hatching of spring-laid eggs after their cold storage for autumn rearing.

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The parent silkworm eggs of local varieties PAK 1 and PAK 2, belonging to Japanese and Chinese breeds respectively, were incubated and reared under optimum conditions of temperature and relative humidity during spring 1989 in Sericulture Research Laboratory, Lahore. The male and female insects of the two varieties were separated at pupal stage. F1 hybrid eggs were produced by crosses between the female moths of the variety PAK 1 and male moths of the variety PAK 2. The eggs were laid by moths on April 15, 1989, at a temperature of 26oC. Autumn rearing is possible in most parts of Pakistan during the month of October after a long unfavourable hot summer period. The eggs were therefore, kept at the room temperature of 22 to 27oC for a period of 61 days (April 16, 1989 to June 15, 1989) before their cold storage. These eggs were then refrigerated for 93 days (June 16, 1989 to September 16, 1989) at a temperature of 4oC to 6oC till the time of acid treatment. The relative humidity during the refrigeration of eggs was maintained at 80-85%. A control group of nine egg layings in three replications of three layings each was maintained at room temperature of 18 to 27oC. The eggs of the control group were neither refrigerated nor treated with HCl.

The refrigerated eggs were taken out of the refrigerator on September 17, 1989 at 7 A.M. and kept at room temperature of 26 to 27oC for six hours. The eggs were then dipped in 2% formaline solution for ten minutes, washed with running tap-water and dried. Thirty six egg layings were selected and divided into four groups of nine each. Nine layings of each group were further sub-divided into three replications of three layings each. The duration and temperature were identical in all treatments e.g., 6 minutes and 48oC respectively for each group. The temperature of acid solution was maintained by using a hot water bath during the treatment.

The details of the experiment for application of different acid concentrations to the eggs are given below:

Group Nnumber			Replication number			concentration of HCl solution Percent					
No. 1 dated willde		1	No.								
		(i) 1 (1 (28 29 30 (%)	21.0		los catión No No solution				
	2 8	2 ((ii)	: 8.		20.0					
	3	3 ((iii) (i) (ii) (iii)	1.0	5.0 25.0 40.0	19.0					
	4	08 4		2.0		18.0					
		0.79 51	(iii)								
		5	(ii): (iii)	Control		No tr given	reatment was				

After acid treatment, the eggs were washed thoroughly with running tap-water, air-dried and incubated at room temperature of 26 to 27oC.

The hatching started on September 27 and continued till September 30, 1989. The hatching data was recorded daily in each treatment and replication by ocular observations and averaged for each group. The useful hatchibility was determined on the basis of the counts of the two major brushings and was expressed in percentage.

The details of the experiment for application of different acid concentral stluses.

The results of the experiment are given below in Table:1.

Table:1 Hatchability (%) of eggs refrigerated at 4 to 6oC and soaked in HCl solution of different concentrations at 48oC for 6 minutes.

Group No.	Replication No. solution		Concent- ration_ of HCl 27 September		Date-wise hatchability				Total Useful		
110.					(%) 28 29 30 (%)		hatch- ing (%)			hatch- ability	
1	. 2	3	4		5		6(1)		7	8 9	
1.		21	0.00					(i) 0 -			
1	(i)		85.0	5.0	1.0		- (1		91.0	90	.0
1	(ii)		60.0	25.0	-		- 1		85.0	85	
1	(iii)		30.0	40.0	5.0		- (i		65.0	60	
Av	Average:		55.0	23.3	2.0		-	46	80.3	78.	3
			18.0					u) (-			
2.	1	20									
	(i)		95.0	2.0	-		-		97.0	97.	0
	(ii)		70.0	20.0	-		-		90.0	90.	
2	(iii)		3.0	20.0	70.0)	2.0		95.0	90.	
Average:			56.0	14.0	23.3	3 W-010	0.6	nt, the	94.0	92.	3
3.		10					in perati	al moo	t Ja Lio	neduoin bi	
3	(i)	Septe	85.0	5.0	2.0	S 73d	92.0	00.0		e halchin	
181003	(ii)	epliqa	85.0	5.0	1.0	sa di	91.0	90.0			
3	(iii)		70.0	15.0	-	2.0	87.0	85.0			
Α	verage:		80.0	8.3	1.0	0.6	90.0	88.3			
		18									
4	(i)		90.0	5.0		_	95.0	95.0			
	(ii)		90.0	8.0			98.0	98.0			
	(iii)		30.0	50.0	5.0	5.0	90.0	80.0			
A	verage:	-	70.0	21.0	1.6	1.6	94.3	91.0			•

No hatching of eggs was observed in control group, which were neither refrigerated nor acid treated. It is evident from the above results that acid treatment with HCl solution greatly facilitated hatching of silkworm eggs in autumn in all four treatments. Hatching % was very high in them. However, the acid solution with concentration of 20 percent at 48oC when applied for 6 minutes after 93 days of refrigeration at a temperature of 4 to 6oC was found to give best results. The results are in agreement with those reported by Japanese investigators (Tazima, 1978). Therefore hatchibility obtained under the local conditions was considered satisfactory.

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