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Mass Rearing and Life History Studies of Drosophila Under Laboratory Conditions

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Wild Drosophila flies can be collected on fermenting sugary, flavourous fruits or vegetables [1-2]. Marked difference have been observed with respect to insecticide resistance between laboratory reared flies and those collected freshly from the field [3-4]. As the present workers were embarking upon genotoxicity as well as resistance studies, mass rearing of resistant and susceptible strains of D. melanogaster, obtained from a Swiss laboratory was of utmost importance. Six strains of Drosophila melanogaster, including one local and five other strains obtained from Swiss Toxicological Institute, Zurich i. e. (a) "Basic", homozygous for basic chromosome having particular character of narrow shaped orange eyes; (b) Berlin-K, a strain being maintained since early 20th century as a reference Wild Strain without having any pesticide exposure, collected from Berlin, Germany; (c) Hikone-R, strain resistant to several pesticides including DDT, chlordane, parathion and other early pesticides; (d) Kansanaes, resistant to inorganic chemicals like mercury compounds and other toxic mineral chemicals; (e) Ug pren, strain susceptible to DDT with vestigial wing marker (Dr. F. E. Wurgler personal communications). Whereas, the local strain was the same as used by Khan et al. [1]. All the said strains were reared on wheat bran diet [5] and kept in double wall wooden boxes of 51x 66 x 41cm with two chambers in each (Fig. 1). Each box was partitioned into two chambers for keeping larvae and adult flies of the same strain in the same box, but separately. The temperature was maintained by airconditioning around $27^{\circ}C \pm 1^{\circ}C$ with $65 \pm 5\%$ RH in summer, while in winter for this purpose, electric bulbs of 15 watts fitted in each chamber.

To control bacterial and fungal growth, different combinations of selected fungicides/antibiotics/bactericides and anti-mold agents were tried. Additionally, streptomycine (0.1%) @ 5ml/litre of food together with grisovin, sodium metabisulfite, citric acid, benzoic acid, propionic acid or methyl propylpara hydroxybenzoate assisted in combating the problem. These additives at the rate of 0.1 mg/litre, 2.5 ml/litre and 0.05 mg/litre of food were added into the food respectively, with continuous stirring, just before pouring the food into the bottles. To observe efficacy of each additive regarding microbial growth control during insect culture development, twenty



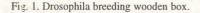


TABLE 1. LIFE HISTORY OF *DROSOPHILA MELANOGASTER* AT $26^{\circ} \pm 1^{\circ}$ C on Wheat Bran Media.

1.	Embryonic development	= 1	1 day
2.	Ist larval instar		1 day
3.	2nd larval instar	a.01 = 0.02	1 day
4.	3rd larval instar	1000 H <u>a</u> 0001 ni	2 days
5.	Prepupa	a. x = 11.00	4-5 days
6.	Pupa		4 days
		Total	: 9 days

TABLE 2. SELECTION OF BEST TEMPERATURE RANGE FOR DROSOPHILA REARING ON WHEAT BRAN DIET.

S.No.	Temperature °C	Average generation ime (in days)	Remarks
1.	$20^{\circ} \pm 2$	$14 \text{ SD} \pm 0.63$	1997 - 1998 - 1998 - 1998 - 1998 - 1998 - 1998 - 1998 - 1998 - 1998 - 1998 - 1998 - 1998 - 1998 - 1998 - 1998 -
2.	$25^{\circ} \pm 1$	11 SD ± 0.63	-
3.	$27^{\circ} \pm 1$	9 SD ± 0.37	
4.	$30^{\circ} \pm 1$	8.5 SD ± 0.71	sterility*
5.	$35^{\circ} \pm 1$	9 SD ± 0.32	30% deaths**

*Sterility was determined as eggs obtained from healthy adults were kept in batches of 10 in wheat bran diet at desired temperature; on emergence 10 pairs in three replicates were kept on $26^{\circ} \pm 1^{\circ}$ C. Number of flies appeared in F1 in both types are compared and 50% or less production in under test set were considered as sterile.

**Three replicates with 10 eggs in each were kept at desired temperature, Larval and adult mortality was observed at every 24 hours and pupae failed to emerge were counted as dead. Control batch was also kept on $26^{\circ} \pm 1^{\circ}$ C for comparison. feeding bottles were prepared in aforesaid way and kept along with 25 pairs of *Drosophila* sp. at the designed temperature and RH%. Simultaneously, a blank control was also kept under the same conditions for comparison.

Adult flies were most conveniently transferred, periodically for egg-laying, from older to newer bottles by positioning, the former upside-down over the latter and shaking/ tapping. Life history, on described parameters, was also observed and presented in Table 1.

Mass production of *Drosophila* flies was the basic need. As rearing media, the recipe used was by Aijaz *et al.* [5].

TABLE 3. EFFICACY OF DIFFERENT PRESERVATIVES IN MOLDSAND FUNGUS DEVELOPMENT CONTROL IN WHEAT BRAN MEDIA*

FOR DROSOPHILA							
S.	Preservatives Quantity used Perc		cent**				
No.		Control					
1.	Sodium metabisulphite	0.1mg/litre food	40				
2.	Grisovin tablets powder	0.1mg/litre food	30				
3.	Citric acid	2.5ml/litre food .	Nil				
4.	Benzoic acid	0.1mg/litre food	40				
5.	Propionic acid	2.5ml/litre food	60				
6.	Propyl para hydroxy benzoate	0.05mg/litre food >	95				
7.	Propyl para hydroxy	0.05mg+5ml/litre >	98				
	benzoate+propionic acid	food					
8.	Untreated (Control)	and the second sec	Nil				

*Wheat bran media was prepared as per recipe developed by Aijaz *et al.* [5] i. e. Agar : Yeast : Sugar : Bran

8 gm : 8 ml : 50 ml : 132 ml and preservatives in 1000 ml water. Aijaz et al. used 0.1% streptomycin 5 ml + propyl parahydroxy benzoate 0.3 gm/litre food.

** $C = 100 - [T^{-1} . X \text{ in . 100}]$

Where C = % control., T = Total No. bottles., X in = Mean No. of bottles infested with microbial growth.

Flies were reared at 26-28°C temperature and between 60 - 72% RH%. The changes in the life-time duration with the change in temperature was also reported by Demerec and Kaufmann [6]. As evident from Table 2, it was found that $35^{\circ} \pm 1^{\circ}$ C was quite improper for *Drosophila* rearing and $30^{\circ} \pm 1^{\circ}$ C exerts sterility effects on the flies. This finding was also in accordance with Demeren and Kaufman [6]. In the present study it was found that the lowest temperature, $20^{\circ} \pm 2^{\circ}$ C made adverse effects on life cycle duration and biological activities.

As shown in Table-3 amongst various fungicide and antimold agents, propylhydroxy benzoate in combination with propionic acid was found to be a good rearing media preservative.

Key words: Drosophila, Mass rearing, Life history.

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