

hybrid between *Pavo cristatus* and *P. muticus muticus*. Growing tail-feathers from both sexes provided material for study.

Figure 1 shows the karyotype of a female peafowl. The first and second largest pairs consist of submetacentric chromosomes clearly distinguishable from each other by size difference. No. 3 is represented by an acrocentric pair and No. 4 by a subtelocentric pair. They are the same length as the long arm of No. 1. A Z-chromosome was identified as a submetacentric similar in size to Nos. 3 and 4, remaining unpaired in female cells. The chromosomes Nos. 5, 6 and 7 are of the same length, being about  $\frac{3}{4}$  the length of Nos. 3 and 4. No. 5 is an acrocentric pair with a definite short arm. Nos. 6 and 7 are submetacentric and metacentric chromosomes, respectively. The chromosome identified as a W-chromosome in female cells is one of the small acrocentrics approximately  $\frac{2}{3}$  the size of chromosomes Nos. 5-7. The eighth pair consists of small acrocentrics slightly shorter than the W-chromosome. There are at least 30 pairs of microchromosomes giving a possible chromosome number of 78 in total. They form a continuous series in size without any visible morphological difference.

Ostrich (*Struthio camelus camelus*) (Figure 2): Developing wing-feathers of a young ostrich of unknown sex were used for study.

Karyotype analysis revealed that the largest 6 pairs were recognizable individually on account of their characteristic configurations. The first 2 pairs of chromosomes

consist of submetacentrics, the No. 2 chromosomes being slightly shorter than No. 1. The chromosomes of No. 3 are acrocentric, being similar in length to the long arm of No. 1 chromosomes. Chromosomes 4, 5 and 6 are acrocentric, having slightly decreasing size in order, but they are not always distinguishable from one another. In addition, there are a number of small acrocentrics. The diploid number of the chromosomes available for counting in this species seems to be 80. It seems probable that the bird under study may be a male, because of the fact that there was no heteromorphic pair amongst the larger chromosomes. More detailed studies are in progress<sup>6</sup>.

*Résumé.* Description d'une technique de culture à partir de la moëlle des plumes, cette technique ayant permis l'analyse du caryotype du Paon et de l'Autruche.

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<sup>6</sup> Our cordial thanks are due to Director S. NAKAGAWA of the Maruyama Zoo and Director H. H. TAKAHASHI of the Obihiro Zoo, for their kind cooperation in collecting specimens for study. Further, we are grateful to Mr. M. ITOH for his friendly assistance

## A Refined Test for X-Ray Induced Dominant Lethals in *Drosophila*

Samples of newly inseminated *Drosophila* eggs can be obtained by the egg collection method of WÜRGLER et al.<sup>1</sup> The eggs deposited by the females on a moist black paper are transferred to a small plexiglass plate (1 mm thick) with the aid of a small brush. Under a dissecting microscope they are arranged side by side in the centre of the plate and kept wet by adding a small drop of water. The sample can thus be irradiated the moment it has reached a desired age. Egg collection and irradiation are carried out in a climatized room with a temperature of 25°C and 96% relative humidity.

After irradiation the eggs are transferred to a strip of agar (2% agar, 5 mm thick) lying on a glass plate (7.5 × 1.5 cm, one end cut to a point). The number of irradiated eggs (EI) is counted under a dissecting microscope by placing them in groups of 10 beginning about 1.5 cm from the tip of the agar strip. The glass plate with the agar strip and the eggs is then brought into a special culture tube. The latter consists of a glass tube (2 cm in diameter) of about 9 cm length whose upper end can be closed by a foam rubber stopper while the lower end fits onto a hollow plastic bowl (2 cm high). The bowl contains 2.5 ml of dry yeast medium (200 ml water, 5 g dry yeast and 30 g sugar are boiled; upon cooling, 4 ml of a Nipazol solution (4.28 g Nipazol-M-sodium in 200 ml 70% alcohol) and 4 ml of a Terramycin solution (0.0022 g in 200 ml 70% alcohol) are added). 1 drop of a concentrated aqueous suspension of living yeast is added to the surface of the medium and partially covered with a small piece of blotting paper.

The culture tubes containing the eggs are kept in a room with a temperature of 25°C and 60% relative humidity. After about 21 h the surviving larvae hatch. Attracted by the yeast<sup>2</sup> and following a humidity gra-

dient<sup>3</sup> they crawl into the medium. 48 h after egg collection practically all surviving larvae have reached the medium. Now the glass plate with the agar strip is removed from the culture tube, and surviving larvae found on the agar are brought back into the tube. The dead embryos and the empty egg shells lying on the agar strip are gently dried in a stream of heated air and covered with paraffin oil. Since the egg shells are now transparent the dead embryos can be classified under a dissecting microscope and counted according to the following syndromes: WE, white mottled eggs; KU, eggs with a typical spherical concentration of opaque material in the central part of the egg; BR, embryos with a brownish colour; LL, white embryos with larval structures visible; HL, larvae which opened the egg shell but did not hatch; TL, larvae which died immediately after hatching. In addition living hatched larvae (NL) which occasionally have not been transferred back from the agar strip into the tubes are noted.

The tubes with the surviving larvae are brought into a room with a temperature of 25°C and 96% relative humidity. The high humidity in the vials causes most of the individuals to pupate outside the medium (about 4 cm above the surface). On the eighth day after irradiation the pupae are carefully removed from the culture tube with a wet brush and brought into small empty tubes, individually for the collection of virgin females or

<sup>1</sup> F. E. WÜRGLER, H. ULRICH and H. W. SPRING, *Experientia* 24, 1082 (1968).

<sup>2</sup> G. BENZ, *Revue suisse Zool.* 62, 305 (1955).

<sup>3</sup> G. BENZ, *Experientia* 12, 297 (1956).

Mortality and hatching time of *Drosophila melanogaster* in culture tubes with different populations densities

Total No. of			% mortality		Average No./tube			$T_{50}$
eggs	larvae	flies	embryonic	postembryonic	eggs	larvae	flies	
			Non-irradiated controls					h
90	86	85	4.44	1.16	5	4.8	4.7	222
140	126	122	10.00	3.17	10	9.0	8.7	224
280	263	254	6.07	3.42	20	18.8	18.1	224
600	556	520	7.33	6.47	40	37.1	34.7	235*
240	236	180	1.67	23.73*	60	59.0	45.0*	257*
320	315	157	1.56	50.16*	80	78.8	39.2*	310*
			400 R					
170	83	65	51.18	21.69	10	4.9	3.8	230
240	130	99	45.83	23.85	20	10.8	8.2	229
480	255	206	46.87	19.22	40	21.2	17.2	232
720	399	320	44.58	19.80	80	44.3	35.6	242
			1200 R					
900	87	39	90.33	55.17	50	4.8	2.2	229
1400	157	76	88.78	51.59	200	22.4	10.8	236
2000	233	125	88.35	46.35	400	46.6	25.0	236
4000	394	173	90.15	56.09	800	78.8	34.6	245

In all tests pupal mortality was negligibly small. Asterisks indicate unfavourable rearing conditions resulting from overpopulation of the culture tubes.  $T_{50}$ , time at which 50% of the surviving flies have hatched.

in groups of 5 for other experiments. The number of pupae damaged during this procedure (ZP) is usually very small. Some days later, the flies hatched in the tubes are classified according to their phenotype (G1, G2, ... G10; flies which cannot be classified with certainty are taken together as GU) and the dead pupae according to their lethal syndrome (tentative classification<sup>4</sup>: FP, individuals dead before or immediately after pupal molt; SP, pupae dead with imaginal structures visible; HP, pupal case open and fly partially hatched). These counts and the counts of dead embryos permit the calculation of the number of dead larvae (SL) which cannot be counted directly.

The data obtained in such a dominant lethal test can be evaluated on a high speed electronic computer with a Fortran programme (Prozwert-10). For this purpose the number of irradiated eggs (EI), the number of dead individuals (WE, BR, LL, HL, TL, FP, SP, HP), the number of surviving flies (G1, G2, ... G10, GU) and the number of 'accidents' (NL, ZP) are punched on data cards. In the output the lethality as well as the survival data for single syndromes or meaningful combinations of syndromes, and/or for single genotypes and any combination of genotypes can be obtained selectively. Every output list consists of (1) the experimental values and (2) the values obtained after correction of the data for the appropriate spontaneous mortality found in non-irradiated controls<sup>5</sup>.

An extension of this programme (Prozwert-model-punch) punches the calculated survival data on new data cards which can then be used for fitting various models of survival curves to the experimental data<sup>6</sup>.

ULRICH<sup>6</sup> has shown that in large vials the rate of postembryonic mortality depends on the population density. Therefore with the small culture tubes described in this paper a series of tests were carried out (Table). Population densities ranging from about 5–80 larvae/tube were realized by placing into the tubes various numbers of non-irradiated eggs or eggs X-rayed at an average age of  $15 \pm 1.5$  min after deposition<sup>7</sup>. Beside the mortality data the hatching time of the flies has also been recorded. From plots of these data on probability paper the  $T_{50}$  values, i.e. the time at which 50% of the sur-

viving flies have hatched, have been estimated graphically. As can be seen from the Table, unfavourable rearing conditions (increased developmental time and/or increased postembryonic mortality) are not correlated with the number of larvae initially present in the tubes, but with the number of larvae which reach the pupal stages. In experiments not resulting in a high percentage of dead pupae the number of surviving flies can be taken for the estimation of the critical number of eggs/tube. In consequence for routine tests the number of eggs/tube has to be limited so that not more than 25–30 pupae/tube will be produced.

This dominant lethal test has been adopted successfully for experiments in which stages of spermatogenesis or oogenesis<sup>8</sup> were irradiated<sup>9</sup>.

*Zusammenfassung.* Die nach Röntgenbestrahlung frisch abgelegter besamter *Drosophila*-Eier registrierbare Letalität kann nach verschiedenen Letalsyndromen aufgliedert werden. Zur Auswertung stehen Computerprogramme zur Verfügung. Spezielle Tests zeigten, dass mit der beschriebenen Zuchtanordnung bestrahlte und unbestrahlte Populationen, von denen 25–30 Puppen (bzw. Fliegen) pro Zuchtglas überleben, ohne Beeinflussung durch ungünstige Zuchtbedingungen auf dominante Letalfaktoren geprüft werden können.

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<sup>4</sup> P. MOLLET, Diplomthesis ETH, unpublished.

<sup>5</sup> F. E. WÜRGLER, Int. J. Radiat. Biol. 14, 193 (1968).

<sup>6</sup> H. ULRICH, Revue suisse Zool. 67, 287 (1960).

<sup>7</sup> U. PETERMANN, Diplomthesis ETH, unpublished.

<sup>8</sup> U. LEUTHOLD and F. E. WÜRGLER, in preparation.

<sup>9</sup> Work supported by Schweizerischer Nationalfonds zur Förderung der wissenschaftlichen Forschung and Jubiläumsfonds 1930 der ETH.