other initial media.

One hundred eye-antennal discs isolated from Oregon-R larvae yielded about 4 x  $10^5$  single cells, whereas one hundred wing discs from the same strain produced about 4.5 x  $10^5$  cells. One hundred eye-antennal discs from Bar larvae yielded about 3 x  $10^5$  cells by the same procedure.

When single cell suspensions each containing  $10^5$  cells in 0.3 ml medium were introduced into tightly covered micro-beakers, rotated on a gyratory shaker for 24 hours at 100 rpm at  $28^{\circ}$ C, tissue-like cell aggregates were reconstituted in the center of the micro-beakers. Cell aggregates obtained from eye-antennal disc cells of the Oregon-R strain had an average diameter of 0.6 mm, whereas eye-antennal disc cells from the Bar strain formed smaller and looser aggregates of an average diameter of 0.4 mm. In the staining preparations of the aggregates ommatidium-forming cells formed some cell clusters, which were separated and sorted out from those of antenna-forming cells.

Wing disc cells from the Oregon-R strain formed after 24 hours of rotation aggregates of 0.5 mm in diameter which showed a characteristic structure different from that of eye-antennal disc cells. The differences in histogenesis and organogenesis of different imaginal discs are now further studied.

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- 3. Kuroda, Y., 1968, Proc. XII Internatl. Congr. Genet., Vol. II: 100.

Mohamed, Aly H. and Patricia A. Kemner. University of Missouri-Kansas City, Kansas City, Missouri. Cytogenetic effects of hydrogen fluoride on D. melanogaster.

Wild type (Oregon-R) D. melanogaster males were subjected to hydrogen fluoride gas and crossed to untreated Oregon-R virgin females and to tester virgin females of the genotype Pm dp b/Cy sp2; Sb/D CxF. Salivary gland chromosomes smears of the  $\rm F_1$  larvae from the first cross indicated the presence

of chromosomal aberrations such as duplications, deficiencies and inversions. The second cross has been used to determine the induction of subvital, semi-lethal and lethal mutations on the second chromosome.

Abrahamson, S., W. C. Kiriazis and E. M. Sabol. Dept. of Zoology, University of Wisconsin, Madison, Wisconsin. A Storage Effect of Ethyl Methane Sulfonate (EMS) on the Induction of Translocations in Drosophila Sperm.

Experiments were performed to determine the effect of EMS, a monofunctional alkylating chemical, on the induction of translocations, including mosaic translocations, in fresh mature sperm and stored mature sperm.

Oregon-R males aged for 5-6 days were fed a  $0.0125~\mathrm{M}$  solution of EMS for

24 hours (following procedures of Lewis and Bacher). These males were mated to bw/bw;e/e  $\varsigma\varsigma$  for 24 hours and then discarded. The females were allowed to lay eggs for 2 more days, and then transferred to non-yeasted media. After 10 more days the females were transferred back to yeasted media and allowed to lay for 3 days. Progeny obtained from the first three days of egg laying were products of fresh mature sperm; progeny recovered from the last brood were products of stored mature sperm. The  $F_1$  males containing the treated chromosomes (wild-type males) were mated singly to bw/bw;e/e  $\varsigma\varsigma$ . The  $F_2$  cultures were screened for translocations. Six to ten wild-type males from each normal  $F_2$  culture, those not exhibiting translocations, were further tested for mosaic translocations by single pair matings with bw/bw;e/e  $\varsigma\varsigma$ .

One Y-2 translocation was recovered from 271 fertile cultures from fresh mature sperm. Although only 20 vials were fertile in the stored mature sperm test, three 2-3 translocations were recovered, giving a translocation frequency of 0.15. This is a remarkable increase over the translocation frequency of 0.004 obtained in the fresh mature sperm test.

No mosaic translocations were recovered after testing 6-10 males from each of 110 F2 normal cultures from fresh sperm nor from the 17 aged sperm cultures. However, two other kinds of mosaics were observed in the stored sperm test. Among the progeny of the 6 males from F2 normal culture #7 two of these vials showed that bw/bw of &00, bw/bw;e/e of &00, and +/+ &00 all

were present in equal ratios. The number of +/+ of was in both cases about half the number of other classes and only one or two e/e of equal were recovered. Abnormal ratios have been observed through three more generations. The second mosaic was a lobed eye phenotype associated with all classes in one vial from another test group. Further tests will be performed to determine if a chromosome aberration is responsible for these two cases.

Although only a few cultures were tested from the stored mature sperm, it is clear that there is a storage effect of EMS on translocations in mature sperm. It is interesting to note that the mosaics recovered were also found in the stored sperm sample. Work supported by NSF grant (G-19394) and the Wisconsin Alumni Research Foundation.

Development of a 2-2 egg or 0 egg is possible only in case of fertilization by complementary male gamete. Males bearing isochromosomes 2 produce 50 per cent of aneuploid gametes 2-2 and 0, which enables to use these males to trace

2-2 and 0 gametes in XXY females. As the remaining half of the male gametes bears only one isochromosome 2, all the euploid eggs will be dominantly lethal. In the first experiment 456 females  $In(1)d1-49+B^{M1}$ , so  $v \ B^{M1}/sc^7 \ v \ f/Y^{BS}$  were crossed to C(2L)RM,b;C(2R)RM,cn males. The progeny (comprised of 11 individuals) included the following phenotypes:  $2^{op}$  sc;b cn,  $1\circ v$  sc,  $1\circ b$  cn,  $3\sigma \sigma$  sc  $v \ B^{M1}$ ,  $2\sigma \sigma$  sc  $v \ f$ ,  $2\sigma \sigma$  sc  $v \ f$ . In the second experiment 384 females  $In(1)d1-49+B^{M1}$ , sc  $v \ B^{M1}/sc^7 v \ f/Y^{BS}$ ; SM1,Cy were crossed to similar males. The progeny (comprised of 231 individuals) is shown in the table:

	Reg.females		Reg.males		Exc.females	Exc.males
	XX	XXY	XY	XYY		24
Matroclinic 2-2	75		76	-	2	26
(Cy) Patroclinic 2-2 (b cn)	1	15	-	19	16	1

The obtained preliminary data show that heterozygous inversion SM1,Cy in XXY females increases the frequency of the 2 chromosomes non-disjunction by more than an order of magnitude. Further, it may be suggested that in the process of aneuploid 2 formation in  $XXY^{BS}$ ; SM1,Cy females a considerable role is played non-homologous pairing of X-2 and Y-2.

Wakahama, Ken-Ichi<sup>1</sup>, Osamu Kitagawa<sup>2</sup>, and Costas D. Kastritsis<sup>3</sup>. 1. Department of Biology, Shimane University, Matsue, Japan; 2. Department of Genetics, Tokyo Metropolitan University, Tokyo, Japan; 3. Department of Anatomy, Southwestern Medical School, Dallas, Texas. Chromosomal variation and sexual isolation in the Drosophila nasuta complex.

The Drosophila nasuta complex has a wide distribution in the tropical and subtropical regions, and consists of at least 12 species.

Two groups (the Okinawa-Formsa group and the Hawaii-Samoa group) recognizable by some major morphological characters, were utilized for a study on chromosomal variation and reproductive isolation.

For the study of sexual isolation between groups, the Multiple Choice Method  $\,$ 

was applied. From the 16 cases studied it became apparent that the Oriental group is completely sexually isolated from the South Pacific group showing an isolation index of 1.00 or nearly 1.00.

In addition this, sexual isolation was studied by calculating the percentage emergence of flies from the eggs oviposited by each cross-mating. In this experiment, complete sexual isolation was also seen between the Hawaiian and the Okinawan strains, showing the percentage of 0.06.

The species belonging to the nasuta complex exhibit a very long arm in the salivary gland chromosomes as well as three medium arms and a dot. The salivary gland chromosome complement of the Okinawan strain, however, did not show a dot chromosome in contrast to all other strains