Research Notes

Alikanian, J. The maternal effect. While studying mutations in the left end of the sex chromosome of Drosophila melanogaster, I obtained in 1940 a rather interesting lethal mutation - l_{76} (for details of my method of obtaining lethals see Zoological Journal N). Males carrying the lethal mutation never emerged if there was no deletion in their genotype. Females homozygous for this lethal were fully viable, fertile, and for obtaining offspring were further mated with lethal males, always carrying a deletion. In the progeny there always appeared both wild-type females (carrying the deletion) and yellow females (without the deletion) homozygous for l_{76}. A thorough genetical analysis of the lethal was carried out by me, but unfortunately all the experimental records were lost during the war. It is of peculiar interest to note one phenomenon - the existance of tumors in larvae. The time of action of l_{76} in males was rather late and was manifested in adult larvae up to pupae formation. All such larvae possessed tumors in the form of free-lying formations in the body cavity. Their shape and dimensions varied. The color varied from brown to black. The larvae without the deletion are yellow. There is another interesting feature. The larvae of males carrying the tumors mentioned above appeared only in the progeny of females homozygous for l_{76} and not carrying the deletion 215. No larvae with tumors were found in the offspring of females carrying the lethal and the deletion 215. This is an interesting example of the maternal effect. It is necessary to point out that these tumors appeared in larvae in the process of their growth, so that larvae that were at first normal began after a few hours to manifest tumors.

Barigozzi, C. and Trapani, E. (unpublished data). Wing bristles in wild samples of melanogaster from Luino, Civate, and Gaiano were counted in order to determine the cell size. The three samples are significantly different. Through reciprocal crosses a maternal effect is demonstrated. Cell size in males is smaller than in females, but the entity of the difference varies in the three strains. It seems to prove an influence of the Y chromosome.

Bird, M. J. X chromosome of Drosophila subobscura. 0 m (miniature). 14.5 pm (plum). 32.5 ct (cut). 68.5 bg (bulge). 86 sn (singed). 91 cp (copper). 128 c(X) (crossing-over suppressor). 136.8 bnz (bronze). 138 lethal I(1)40j, lost. 143 v (vermilion). 143 cv (crossveinless). 147 dw (dried wing). 155 lethal I(1)41 K, lost. 169 N (Notch). 183 lz (lozenge). 191.6 w (white eye). 198.2 y (yellow). 198.6 sc (scute). 198.7 scl (short costal). 200.2 wi (withered wing). 220? bb (bobbed), lost.

The order of v and cv is as given; the distance is about 0.14 unit. The order of sc and scl is uncertain. lz and w are male sterile, as visual stimuli are needed for mating in this species. Hence lz cannot be combined with N, and the distances between cv and w are doubtful, as is that between cp and cv, which give 39.7% crossing over. The distances given are in general longer than the recombination values, an allowance being made for multiple crossing over. The homology of m, ct, v, N, lz, w, y, sc, and bb with the corresponding melanogaster genes is highly probable. Some other genes have been lost after partial location. In two six-point counts in which m and pm segregated with ct sn cp v sc, 3 flies in 2839 showed five-fold crossing over.

Bird, M. J. (2) A gene affecting the male genitalia in Drosophila subobscura.

A wide range of structural defects occurs in the male genitalia of cv^2, but not of cv^1, which produces indistinguishable effects on venation. The normal organs differ from those of D. melanogaster mainly in that the testes are slightly elongated ellipsoids of a bright orange color. The most abnormal form has two spherical testes, but no ducts, sperm pump, or external genitalia; the least abnormal form has one colorless degenerate vas efferens still attached to a testis. The commonest abnormal types are: (a) Right and left sides separate, each consisting of a vas efferens joined proximally to a testis and distally to a paragonium; no vas deferens. (b) One testis detached and spherical, the other joined at one point to two vasa efferentia each ending in a paragonium, no vas deferens. In both types the system floats loose in the body cavity; and there may be a small reduced sperm pump. Detached testes are colored, and in the few cases where a vas efferens was not attached to a testis it was still colored. The percentage of abnormals in cultures from brother-sister matings ranged from 40.7% of 221 flies to 8.4% of 155 flies. In five generations of inbreeding it has not fallen systematically, so there has been no selection of modifiers.

Bonnier, G., Rasmuson, B., and Rasmuson, Marianne The Bar facet number.

The mutant Eb, described by Bonnier, Nordenskiöld and Bagman (Hereditas 29: 113-133, 1943; earlier symbol in DIS-16: i; in Bridges and Brehme, The mutants of Drosophila melanogaster, denoted by I) occurred in a homozygous B female with attached-X's. After detachment linkage tests showed Eb to lie very close to the right of B. Salivaries showed that the Eb chromosome has lost one of the two B segments. The phenotypic effects of Eb were found to be: Eb/+ gives round eyes, whereas Eb/B and B Eb/+ give eyes of the homozygous B type. Before making facet-counts the strains were made homozygous and isogenous by marking the X with a number of known recessive mutants and by repeated brother-sister matings. It was then found (cf. Hereditas 29, 1943) that the original Eb/B females had an average of 84.27 facets per eye, and the B Eb/+ an average of 40.57. In the new strains of Eb/B, which were received after crossing over in the B Eb/+, the average facet number was 89.97. The chief interest lies in the fact that, though the original and the recovered B/Eb strains have facet numbers of the same orders of magnitude, yet the difference is statistically significant.

This problem has been tested further. Again a very great number of generations (not counted, but probably more than 100) of strong inbreeding preceded the experiment. From the above-mentioned strain of B Eb/+, with an average of 40.57 facets per eye, which now acted as the original one, three new strains were secured through crossing over at 3 different instances (strains nr 1, 2, 3). From strains 1 and 2, strains of B Eb/+ were again secured by crossing over at different instances (strains 1C, 1E, 1F, and 1H from strain 1, and strains 2T, 2U, 2V, and 2Y from strain 2). From all these strains counts were made of the facets of the right eyes of females, bred simultaneously in the same incubator. The results were as follows

Type	Strain	No. of the eyes counted	Average No. of facets
B/Eb	1	446	69.32
"	2	411	66.65
"	3	74	98.87
B Eb/+	Original	92	39.67
"	1C	372	42.99
"	1E	387	39.61
"	1F	19	54.55
"	1H	313	40.47
"	2T	411	42.16
"	2U	378	46.43
"	2V	118	56.24
"	2Y	178	40.18

To this it must be added that in one single culture bottle of the original strain B Eb/+ a decisively different number of facets was found - viz., 30.70 (40 eyes). This bottle is not included in the above record. Disregarding this bottle (it will be discussed, and a more detailed description given, in a later publication), it is found that the difference between the old and the new counts in the "original" strain of B Eb/+ - viz., 40.57 - 39.67 - is statistically insignificant. On the other hand, differences that are statistically very significant are found between the different B Eb/+ strains, and it can be shown that they are not wholly due to environmental causes.

Gersh, Eileen Sutton Chemically induced phenocopies in D. melanogaster. Flies of Oregon-R stock were raised on food containing (1) potassium arsenite (about 0.03% by weight), (2) boric acid (about 0.05% by weight). Eggs or larvae were transferred to the special media within 48 hours of laying. Small numbers of adult flies were obtained, which showed the following effects: (1) Effects of $KAsO_2$. (a) Dull and somewhat wavy wing surface in 16 out of 29 flies (ca. 55%). (b) Grade I melanism: very dark trident, accessory longitudinal stripe on each side of trident, and pigment at posterior end of scutellum in 16 out of 29 flies (ca. 55%). (cf. mutant ptg.) (c) Grade II melanism: very dark trident and accessory lateral stripe, but no scutellar pigment in 9 out of 29 flies (ca. 31%). (d) 5 out of 29 flies (ca. 14%) completely normal. (2) Effects of H_3BO_3. (a) Melanotic tumors in an undetermined proportion of larvae. (cf. mutant l(1)7.) (b) Reduced eyes of various grades up to completely eyeless in 6 out of 21 flies (ca. 29%). (cf. mutant ey). (c) Abnormal aristae in the 4 flies with greatest reduction of eyes (ca. 19%). (cf. mutant ss^a.) (d) 15 out of 21 flies normal. At higher doses fewer flies were obtained, but a larger proportion of abnormalities. Allowing for differences in the dosages used, it may be considered that these results provide substantial confirmation of those published by Rapoport (Bull. Biol. et Med. Exp. de l'URSS. 7: 415, 1939), at least with regard to the effects of arsenic and boron compounds.

Gloor, H. Phenocopies of tetraptera and bithorax mutants. By treating Drosophila eggs at an early embryonic stage (within 2-4 hours after oviposition) with chemical agents, mainly ether, it is possible to obtain in a considerable percentage (up to 40%) adult specimens with the metathorax modified in a characteristic way. The sclerites, legs, and halteres resemble in a variable degree the corresponding organs of the mesothorax.

Thus it is evident that those modifications arising from ether treatment may be considered as phenocopies of the mutant characters tetraptera, bithorax, etc. In very extreme cases a complete and in most details normal-looking second mesothorax instead of a metathorax is formed. In our laboratory H. Niggli obtained a similar effect by treating eggs with a phenol solution.

Hinton, Taylor Salivary chromosomes of irradiated larvae. Eggs and young larvae of Drosophila melanogaster were treated with X-rays. Just prior to pupation half of the larvae were taken from the culture bottle and smear preparations made of the salivary glands. The other larvae were allowed to pupate and hatch. Of the 70 that hatched, 7 showed malformations in the development of the eyes, legs, or wings. Examination of the salivary-gland chromosomes of the first group of 100 larvae failed to reveal any chromosomal disturbances, even though a search was made for small aberrations.

Hinton, Taylor (2) Further study on position effect in In(2LR)40d. A more detailed analysis than was first made of the effect of temperature upon the phenotypic expression of this inversion shows no significant effect of temperatures ranging from 15° to 26° C. However, the severity of the effect on the offspring is increased with the aging of the parents. The effect is also increased in individuals heterozygous for clot and the inversion, but decreased in individuals heterozygous for In(2LR)40d and the Cy inversion. The effect of age and other inversions is being tested further.

Lieb, Margaret, Valencia, J., and Muller, H. J. New translocations between the X and 4th chromosomes. Using Dubinin and Sidoroff's technique of crossing irradiated males to females containing cubitus eyeless and looking for cubitus among the offspring, we have during the past year obtained stocks of fifteen translocations between the X and 4th chromosomes. Approximately 9000 F_1 females were examined, derived from fathers given 3500 to 5000 r of X-rays. In five of the X4 translocations the male is both viable and fertile, and in four of them the break lies in the euchromatic region between the loci of yellow and forked, and has been determined cytologically. The breeding work was done by Lieb and the salivary examinations by Valencia. Before the salivary examinations were made, the positions of the breaks had first been determined approximately by crossing the males to females having attached X's containing various marker genes. The positions of the breaks in the salivary chromosomes of the four stocks are as follows: 4C3, 9A1, 11A7, 11B16. In addition to these four, a salivary examination has been made by Valencia of the X4 translocation which Sidky found while at Edinburgh, and which is commonly designated as "Sidky a." This break is approximately at 13B8-9. In view of the loss of almost all of the X4 translocations designated by the symbol A, possession of the above translocations fills, we believe, a present need. In addition to these translocations numerous translocations were found between the second or third chromosomes and the fourth, but no attempt was made to locate the position of the breaks in these cases. They have been sent to Dr. Griffith, of the University of Missouri, and we understand that he will subject some of them to salivary examination.

Milani, R. Induction of mutations with Na_3PO_4. In 1941 I tried to induce mutations in D. melanogaster by treating with sodium phosphate. Measuring the frequency of lethals on the X chromosome by the ClB method, I obtained; (a) Normal eggs in 11% solution during 7 hours 30 minutes: 4 lethals over 1047 chr. (0.38% ± 0.19).

(b) Dechorionated eggs in 11% solution during 5 hours: 2 lethals over 477 chr. (1.76± 1.22). (c) Feeding (10gr. in 480 of food): 13 lethals on 477 chr. (d) Control (in Ringer): 3 lethals on 545 chr. (0.55 ± 0.32). In (c) each of 3 males gave more than 1 mutation (2-2-3). Although the number of chromosomes tested is small, the results reached in the feeding experiences are significant, I think.

Moree, Ray. Probable X detachment and phenotypic elimination. Several + w^e females were discovered in stock y ♀ x w^e ♂ (y + ♀ x + w^e ♂). New w^e females replaced and eventually eliminated y males; several y females were recultured without w^e females to prevent loss of original stock. Since productivity of w^e females is about twice that of y females, w^e females probably result from detachment of X chromosomes. Cytological examination to be made as soon as microscope is available.

Morgan, L. V. Loci of sv^n and ey^2. It is known that the loci of sv and ey lie outside the range of Df4(M)4 in the fourth chromosome. The region that includes the loci has been more narrowly defined by use of the 4-2L fragment of T(2;4)b. The break in chromosome 4 is proximal to 102D1 in the distal range of 102C (Lewis, U). Flies homozygous for normal second chromosomes and carrying the 4-2L fragment and one mutant fourth chromosome were recognized as hyperploids by somatic characteristics. They showed exaggeration of the mutant, sv^n or ey^2, indicating that the loci of both of these mutants are distal to nearly all of region 102C.

Muller, H. J. Two mutants of mosaic expression not caused by gross rearrangement of heterochromatin. It was reported by the author in 1935 (Proc. 15th Internat. Physicl. Cong.: 587-589) that the curious mosaically expressed mutations in D. melanogaster, of X-ray origin, which he had earlier found (Am. Nat. 64: 245-246, 1930) to be associated with structural changes, and accordingly called "eversporting displacements," always involve a placing of the affected genes near to a "chromocentral" (now usually called "heterochromatic") region--a finding later concurred in by Schultz (Proc. Nat. Acad. Sci., 22: 27-33, 1936). Demerec on the other hand had still earlier (Proc. 4th Internat. Cong. Plant Sci. 1: 943-946, 1926) reported several cases of mosaically expressed mutations of spontaneous origin in D. virilis, which were not associated with gross structural changes and which gave evidence of being of a different nature, dependent on an unstable condition of the affected genes themselves. We have now found two cases in D. melanogaster, one spontaneous and one induced by X-rays, in which there is an appearance of somatic mosaicism much like that in the rearrangements involving heterochromatin, but in which there is no evidence of a structural change of microscopic dimensions having occurred, and certainly no gross change involving heterochromatin. As both stocks appear to be stable, there is no evidence of germinal mutability. It is conceivable that one or both cases are similar to Demerec's miniature gamma in virilis, for which there is good evidence that there is high gene mutability but that this is confined to somatic tissue; but it is also conceivable that the mosaicism is purely a gene manifestation in our cases (as in some mammalian spotting), or involves some semipermanent change like that in the eversporting displacements, if not some new phenomenon.

One of these cases (that of spontaneous origin) involves a mutation of a locus that had presumably been normal to an allele designated as mw, and called "mottler of the white series" (or "mottler" for short). This is a recessive sex-linked gene with locus near (within a few units of) vermilion,

but not allelic to it. In an otherwise normal genotype this mutant gene produces no visible effect, as it is--like the various mutants called "cream" a, b, etc., discovered by Bridges--a specific modifier of apricot and of other alleles of intermediate expression belonging to the white-locus series. In connection with these, it makes the color much lighter, especially in the female. Unlike what is found with Bridges' specific modifiers, however, the color is made patchy, and greatly resembles that of many of the lighter mottled alleles of white that are caused by the juxtaposition of the white locus to heterochromatin. And as in those cases, there is a considerable variation between individuals, as well as between the two eyes of the same fly, though also a positive correlation between the latter. Classifiability always seems possible, however.

The other mutant, X-ray induced, involves the mutation of what was originally the normal allele of forked bristles to an allele called f^m ("forked-mosaic"). Again the flies vary considerably from one another, many seeming like ordinary forked but a high percentage showing coherent patches of the body, often of considerable size, in which the bristles are quite normal; and occasionally practically the whole fly seems normal. Stocks phenotypically like this were obtained in abundance in Belgovsky and Muller's work (Rec. Gen. Soc. Amer. 6: 139-140, 1937) in which a heterochromatic inversion of Bar (B^{MI}) was irradiated, but here the heterochromatin had presumably been involved (as in the yellow effects of scute 8 studied by Nuzhdin). In our present case, on the other hand, the crossing-over tests made so far indicate f^m to be in the usual location for f^+, and to have approximately normal crossover frequencies with other loci. Moreover, salivary examinations by J. Valencia have given evidence of not even a minute (microscopically visible) rearrangement.

Both cases are being followed further. Among other things, it is desired to determine what effect, if any, is produced in them by the presence of an extra Y.

Postscript (added October 7, 1946): Since the above was written, it has been found that the stock of forked-mosaic, although possessing a certain germinal stability in appearing not to mutate frequently all the way to normal, does within certain limits, to be more accurately determined later, have some tendency to inheritable variation in the germ cells. That is, it responds to selection, and does so in a way not readily to be explained by the segregation of modifying genes or of a Y chromosome. This would seem to place it in the general category of unstable genes such as those reported by Demerec referred to above, and would make it the first case of the kind in L. melanogaster.

Novitski, E. Triploidy in D. pseudoobscura. To detect triploidy in D. pseudoobscura, ♀♀ of the constitution or Bl Sc pr/Inv (Cuernevaca) were mated to or ab pr ♂♂ and the F_1 non-or pr ♀ were examined for evidence of Bl and Sc (presumed genotype or ab pr/or Bl Sc pr/Inv (Cuernevaca)). Out of 13,068 non-or pr ♀ , one showed slight Bl and Sc characters and in addition had the coarse bristles and large wing cell size characteristic of triploids. When backcrossed to an or ab pr ♂, she produced two or ab pr ♀ , one or Bl Sc pr ♀ (2N), one + ♀ (2N), and one + ♀ (3N). The constitutions of the F_1 indicate that three of the autosomes marked were present in the parental ♀ . The F_1 triploid x or ab pr ♂ yielded one or ab pr ♀ (2N), one or ab pr ♂(2N), and three + ♀ (all 2N). An oogonial metaphase of one of the latter showed that she was XXY.

The very low fertility of the 3N ♀ and the failure to perpetuate the triploid condition is undoubtedly caused by the larger number of chromosomes (five in pseudoobscura as compared with four in melanogaster), which leads to a

larger percentage of unbalanced zygotes. No intersexes, supermales, or superfemales were obtained; it seems likely that the inviability of the 2X3A, 3X2A, and 1X3A zygotes is much greater in pseudoobscura, where the X chromosome contains roughly twice as much euchromatic material as in melanogaster.

Philip, U. A. Cytology of males in cultures with recessive crossover suppressor.

Thirty-one males from 5 mothers homozygous for the sex-linked crossover suppressor c(X^2) (see note by H. Spurway) were examined. The results were:

Mating	Patroclinous Males		Regular Males				
	XO	XY	XO	XY	YYY	XYYY	XYYYY
XX or XXY ♀ x XYY ♂	1	0	0	3	5	0	0
XXY or XYYY ♀ x XY ♂	3	5	4	7	1	1	1

The male with 4 Y's had also 3 dot chromosomes, and no external genitalia. In addition, five other males hemizygous for c(X) were examined. Three were XY, one XYY, and one XYYY. XXY females form eggs containing all 3 or none of the sex chromosomes, producing XO exceptional males and XXYY exceptional females. XXYY females produce XYY eggs, giving rise to XYYY sons when mated to XY males. Meiosis in c(X) males is not much modified. The chromosomes lag at anaphase. But no unexpected chromosome numbers were found in cells undergoing second meiotic division. Pairing at metaphase I is complete. In males with more than one Y chromosome, including that with 4 Y's, all the sex chromosomes form a multivalent, the Y's associating with the X near the centromere. The association between X and Y is obviously not due to chiasma formation. In crosses between XXY females and XYY males using other stocks no XYYY males were found among large numbers. No male in the progeny of c(X) c(X) females was found to be trisomic for one of the long autosomes, though such males had been obtained before.

Poulson, D. F. and Boell, E. J. Cholinesterase in embryonic tissues of Drosophila.

Experiments carried out by means of the Cartesian diver technique provide information on the development of cholinesterase activity in normal and deficient zygotes. ChE activity is negligible in normal and Notch-deficient zygotes 10 hours of age or younger (at 23° C.) and in unfertilized eggs. ChE activity increases between 10 and 14 hours and rises rapidly until time of hatching of normal larvae. At 24 hours the ChE activity in normal embryos averages 12.8 m.μ.l. CO_2/embryo/hour; in N^8 embryos (male) it averages 34.0 m.μ.l. CO_2/embryo/hour. The activity of N^8 embryos is 2.7 times that of normal, which is very close to the ratio of volumes of nervous systems at that time. N^8 embryos have a higher ChE activity from the time that this is first measurable, between 10 and 14 hours. Activity rises considerably after 24 hours in N^8 embryos and reaches a maximum of 56.0 m.μ.l. CO_2/embryo/hour at 40 hours, after which it declines until the death of the embryo. Central nervous systems were isolated from the rest of the tissues in normal embryos and larvae at 18.5 and 24 hours, and ChE activities of nervous system and remnant were determined separately. ChE activity in the nervous system is roughly 40 times that of the rest of the tissues. The method is sufficiently sensitive so that it is possible to carry out determinations on single isolated nervous systems as well as on single embryos. The results will be described in full elsewhere.

Rendel, J. M. and Suley, A. C. E. Eye buds from larvae of a cardinal subobscura stock injected into larvae of wild-type melanogaster and subobscura stocks, gave implants indistinguishable from the host-type eye. Injected into v melanogaster hosts, the implants were cd. As cardinal is sex-linked, it is probably homologous with vermilion. The name has therefore been changed to vermilion. Preliminary experiments suggest that poppy and scarlet are autonomous in + melanogaster and are therefore not homologous with cn. But these results require confirmation.

Spurway, H. An extreme example of delay in gene action in D. subobscura. Females homozygous for the autosomal recessive grandchildless (gs) themselves appear normal but produce cultures in which all the flies are sterile, irrespective of the male to which they are mated. gs/gs males are normal and produce normal offspring (unless mated to a gs/gs female). The abnormality is transmitted as an autosomal recessive through both males and females. The locus is in the 3rd linkage group with hoary, rough and net. Almost all the sons of a gs/gs female have minute spherical testes like those found in miranda x pseudoobscura hybrids. No cell divisions can be found (Philip). About 3% of these sons have one or two normal testes. These are normally fertile. No intermediates have been found. In the daughters of gs/gs females the ovaries remain in the condition they normally show in young pupae (Fahmy). No fertile or probably fertile females have been found. One of our lines containing the condition, now at the F_{13}, has become balanced. It is interesting to compare both the anatomical effects and the mode of inheritance of this gene with those postulated by Dobzhansky to explain sterility in his pseudoobscura hybrids.

Spurway, H. (2) A sex-linked recessive crossover suppressor in D. subobscura. Females homozygous for a recessive gene c(X) 6 units from cv and v produce 18% of exceptional offspring. These females may themselves be matroclinous and therefore XXY, or they may have been obtained in the F_3 from the cross c(X) XY♂ x +/+ XX♀, and therefore XX. In such females crossing over in the X and the three autosomes in which it has been examined is reduced to about 3% its normal value. The salivary-gland chromosomes have not been examined, but crossing over in c(X)/+ females is normal. There is little heterogeneity among the progeny of different c(X)/c(X) flies, and male and female exceptions occur in equal numbers. Philip gives other data confirming that it is a recessive disturbing disjunction and that its effect is confined to females. The analogy with Gowen's gene c(3)G in melanogaster is obvious.

Vogt, Marguerite. Endocrine factors in Drosophila. Studies on the function of the ring gland led to the distinction of several factors produced by the gland; i.e., a factor producing metamorphosis and probably secreted by the "large" ring-gland cells, a gonadotropic factor, produced by the "small" cells (= corpus allatum) and possibly also having an inhibitory effect on metamorphosis, and a factor inhibiting the coloring process of the puparium and delaying the time of puparium formation, secreted by the "fuchsinophilic" ganglion cells (= copora cardiaca). Further, a hormone produced by the ripening ovary was found; it inhibits the secretion of the corpus allatum and accelerates the metamorphosis of imaginal discs.

Vogt, Marguerite (2) Studies on the state of determination of the larval imaginal discs in Drosophila. Reduplications of legs, antennae, palpi, ocelli and supernumerary facets were obtained by dividing the imaginal discs up to the first half of the last instar. These results, together with the investigation of the "temperature-effective"

period of the mutants aristopedia (ss^a), proboscipodia (pb), and of the gene combination sc, ec, ct; Lfd^r-Ldlers, which leads to duplications of antennae and palpi, lead to the conclusion that the imaginal discs of Drosophila are not finally determined by the last instar. The regulating power, however, as well as the capacity for changes in differentiation, does not extend to the imaginal disc as a whole but is restricted to the "organ-fields" within the imaginal disc.

Technical Notes

Hughes, Roscoe D. Notes on mites infesting Drosophila cultures. Some observations on a species of mite commonly infesting Drosophila cultures are recorded here as being of possible interest to Drosophila workers. The stocks upon which these observations were made were found in a Drosophila culture obtained from one of the biological supply houses. Dr. H. H. J.Nesbitt has tentatively identified these specimens as belonging to the genus Anoetus (Histiostoma), family Anoetidae. The species which has been studied is similar to the species gracilipes (Banks). It is also very probable that the species in question is the same as that named Histiostoma genetica by Stolpe. (Anat. Rec. (Supplement) 72: 133-134). The mites were reared satisfactorily on any of the Drosophila culture media in general use. The standard cornmeal-molasses-agar medium was used predominantly in these studies. The adult male mite may be readily distinguished from the female by the greater thickness of the posterior pair of legs of the former, and the relative uselessness of these legs in crawling over the surface of the medium. This pair of legs is dragged behind in an extended position. The thinner, more fragile posterior pair of legs of the female is used in crawling movements, although to a lesser extent than her first three pairs of legs. Also the adult male is distinguished from the adult female by his smaller size, although this is a variable character. Under favorable conditions—i.e., with freshly made food and a saturated atmosphere--the life cycle of this species may be completed within five days. The normal life cycle, however, appears to be about six days under average conditions. Parthenogenesis definitely occurs in this species. The offspring of unfertilized females (obtained from isolated eggs) are all males. Thus a single female-producing egg, inadvertently transferred from a contaminated Drosophila stock, may be the starting point of a new colony. The nonmigrant form rarely leaves the culture medium except under very crowded conditions. On exposure to normal laboratory atmospheric conditions, the nonmigratory form becomes desiccated in a short time. For the above reasons it is doubtful that this form is an important source of contamination. From the limited observations available at this time, it appears that the longevity of the nonmigrant form is approximately fourteen days or less. The migrant is the real source of contamination in Drosophila cultures. It appears that certain six-legged larvae, or possibly first-stage nymphs, metamorphose into small pink forms, which are especially well adapted for distributing the species. These migrant forms are much more active than nonmigrants, and, when they first make their appearance in a culture, tend to crawl away from the culture medium. They resist desiccation considerably longer than non-migrants, but thrive best under moist conditions. A favorite collecting place for migrants is on the under side of culture-bottle caps.∧ The migrants have survived in vials of water for over two months at room temperature. It is probable that they may survive for even longer periods under favorable conditions. Eventually the migrant forms metamorphose into either male or female nonmigrant forms. Migrants do not occur in large numbers when culture

∧The migrants (hypopial stage) possess a sucking disc on the ventral surface, which is used in attaching to Drosophila. They possess no mouth parts.

conditions are favorable. Under crowded conditions, especially in old stock bottles, they may occur in enormous numbers.

Moree, Ray. A long-boiling culture medium. In order to overcome difficulties of sterilization and storage we have developed the following culture medium. Water 1000 cc., plus agar-agar 20 cc. (9 grams); molasses 150 cc.; mix cornmeal 100 cc., brewer's yeast 30 cc., sodium propionate 10 cc., then slowly add water 140 cc. Agar-agar is dissolved by heat, molasses added next, and cornmeal-yeast-propionate mixture added last. Boil gently, with intermittent stirring, for about one hour; pour into bottles, plug, and steam-bath for one hour. Agar-agar is powdered type, and all measurements of solids are "loose measurements" (i.e., not settled or tamped). One hour of boiling sterilizes satisfactorily and usually gives right consistency to medium; consistency may have to be adjusted by more or less boiling. Some ingredients have given much trouble with bacteria, and sometimes mold—hence the long boiling period. Steam bath (in galvanized can) has proved better than dry heat of baking oven for sterilizing. If cornmeal settles, medium should be shaken up before it solidifies. Cultures are easy to make and suffer a minimum of contamination by microorganisms; when moistened occasionally with distilled water, they have been kept unrefrigerated for one month andlonger without spoiling.

Muller, H.J. Stock list: changes in manner of classification. Instead of numbering our stocks consecutively from beginning to end as in the past, we have divided them up into a number of groups each designated by a small letter and have numbered them within these groups after arranging them pretty much on the usual alphabetical basis within the groups (but see modification noted in second paragraph). This arrangement has several advantages: first, the stocks are more easily found after one becomes acquainted with what groups the letters stand for; second, when stocks are added to or subtracted from the original list there is not nearly so much disturbance of the numbering. Moreover, the addition of the letter, instead of making the designation of the stock a longer one, actually makes it on the average a little shorter, in the case of a long stock list, owing to the fact that there are more than ten letters in the alphabet.

We have also inserted a new column between the number of the stock and the designation of its genotype, entitled "Signal." This we use in two cases. First, in case a stock is a complicated one, which would not readily be found by looking for it in its regular place in the alphabetical scheme, as happens particularly when the first gene in the formula is not the one which we think of as most characteristic of the stock and for the purpose of keeping which the stock is maintained. In that case the signal is so chosen as either to represent or suggest the characteristic gene or genes, or else consists of a made-up word or phrase such as "plex" or "rucuca" or "Cy Fl only," chosen so as to suggest, in abbreviated form, the stock's composition or purpose. The second use of the signals is encountered in cases where, although the stock is not a complex one, it is being kept for the sake of some gene or genetic combination with which the formula does not begin and which, therefore, would not be easily found in the stock list when the latter is represented in the alphabetical order. Thus, for example, in our stock list there are a number of combinations containing the heterochromatic inversion associated with the bar mutation called B^{M1}. Instead of scattering these around through the stock list, we have gathered them all together and given all of them the signal "B^{M1}" so that they can be more easily found, and we have arranged them alphabetically within this signal group. Now in cases where stocks are given a signal, the alphabetic

arrangement of these stocks in the list as a whole follows the spelling of the signal rather than of the genetic formula itself, inasmuch as it is the signal which a person would first look for in trying to find the stock. We find that this arrangement is much more convenient than the old one, although we have not yet used it as extensively as it probably should be used in our own list.

Finally, in making up our present stock list, we have not adhered as rigorously as before to the classification of stocks into pure first, Y, second, third, and fourth chromosomes, then multiples of each type, etc. Although still using this classification in a general way, we have, for example, classed with the second-chromosome stocks those which involve some gene or even translocation connected with some other chromosome, in cases where the stock was being kept primarily for its second-chromosome content; as when, for example, the inversion-translocation combination termed "Indp" or "IndpT23" was being used as a balancer, even though there is a deletion-insertion translocation present here in which a piece has been taken from the second chromosome and inserted into the third. This sort of liberalization of the classification, although it might make the stock list less easy to use for a robot, makes it much easier for an ordinary human being, or even geneticist, to find things that he is looking for.

Muller, H. J. Nomenclature changes adopted in stock list. A good nomenclature should, of course, always have the utmost brevity consistent with ready understandability and clarity. This is particularly important in genetic formulae when they become complicated. With this in mind, we have adopted a number of minor improvements in nomenclature in our present stock list. Some of these will be apparent and understandable, without comment here, when the stock list is read, but others call for some comment. In designating an inversion or translocation we no longer enclose the letters which name that rearrangement in parentheses following the symbol In or T, respectively, but show them immediately after those symbols without a space between as well as without parentheses. In the case of a translocation, the chromosome numbers, shown by X, Y or Arabic numerals, and without spacing between them and between the T and the numbers, are used, and are followed by the further specification of the translocation, also without spacing or parentheses. In the case of an inversion, the number of the chromosome in which the inversion occurs is not given either in parentheses or otherwise when it is obvious from the formula in what chromosome the inversion is--as, for example, when it follows or precedes the symbol of a known gene in that chromosome. Now in the case of an inversion like the inversion associated with scute 8, in which a given gene that is symbolized is regularly associated with the given inversion, we do not, as some authors have in the past, first, give the symbol of the gene and then write In and then in parentheses the symbol of the gene again, used this time to designate the inversion with which it is associated. Instead of this, we merely show the symbol of the gene or genetic combination, such as sc^8, and then follow this by a comma, which is in turn followed by the symbol In. Here, the comma is used to signify that the inversion symbolized after the comma is regularly connected with the gene symbolized before the comma; thus there is no need to write the symbol for the gene a second time. The same system is used in the case of a translocation when that translocation is regularly connected with a given gene, as in the case of that connected with scute 2, which would be symbolized in full as follows: sc^2, TX2. When, however, it may be presumed to be well known to the reader that a given inversion or translocation is regularly connected with a given gene which is symbolized, we have often omitted the sign for inversion or translocation, as the case may be, in the interests of brevity and readability.

Thus, for example, we have seldom inserted the symbols for inversion connected with the symbol for the gene Curly, taking it for granted that those inversions are present. When, however, Curly is present only with the inversion of the left arm of the second chromosome, then we have specifically designated this inversion, using the symbol InCyL; and the presence of this symbol in a formula without the corresponding one for the inversion in the right arm would let the reader know that this inversion alone was present in the second chromosome along with the gene for Curly in the given case. We have further departed from formalism in the case of combinations resulting from crossing over between two inversions—for example, inversions associated with some scute effect—by starting the formula for the chromosome (or the part in question) with the symbol of the gene ordinarily associated with the inversion whose left end is present and ending the formula for the chromosome(or part) with the designation of the gene ordinarily associated with the inversion whose right end is present, even though that gene itself may not be present, as in the combination sc^{S1} sc^{4}. Here, it will be realized by those who understand the structure of scute inversions, the right end of the $scute^{4}$ inversion is present but not the gene $scute^{4}$ itself, even though that appears to be represented in the formula. This will not, however, cause confusion on the part of those who understand the matter, and few but those will need to know the meaning of the symbol.

As before, we use a period to designate attachment between parts which in the normal individual are not attached, when it seems helpful to designate such attachment at all. Of course, in the showing of ordinary mutual translocations the attachments are not represented. In cases where there are two points of attachment between a given part that is represented and another part, which is represented by a preceding symbol or by the symbol of genes contained in it, we use as before a colon instead of a period. In addition to using a comma in the way described in the preceding paragraph, we use it also in the following connection. When there are genes or genetic arrangements to be shown in regions of the chromatin which ordinarily occupy two different chromosomes, such as the second and third, but which in the present instance are somehow connected with one another, as by a translocation, we show the contents of the chromatin derived from one of the original chromosomes first, using the usual symbols for the contained genes, inversions, etc., and then insert a comma, after which we put in the symbols for the genes in that portion of chromatin derived originally from the other chromosome. Suppose, for example, we had a translocation between the second and third chromosomes, and one of these chromosomes contained the genes of the so-called "apl" combination, while the other contained the genes of the so-called "rucuca" combination. We would then show first the symbol T23 and the designation of the translocation, all without spaces, then a space, then the symbols of the genes of the "apl" combination, then a comma, and then, without a space after the comma, the symbols of the genes of the "rucuca" combination. This matter is gone into detail here because in the given translocation there would really be a rearrangement of genes and if one wished to show them literally arranged in the form in which they existed in the given chromatin, one would have to rearrange the genes entirely and thus make the formula very different from the ordinary one and correspondingly hard for the reader to follow. Moreover, since the new arrangement would often be unknown, it might not be possible to show it properly. The present symbolism avoids this difficulty. We do, however, show the genes in their proper arrangement in cases of inversions where that arrangement is known. A further item of our symbolism, which should perhaps be recalled here also, is that we use an equals mark (=) after a period to designate that there is another chromosome containing genes just like those already shown and that this chromosome is attached to the other one, as in the case of attached X's, which form the only class of cases for which this symbolism is so far used. We should also mention

that, in cases where there is no knowledge to the contrary and the symbol "In" following a comma is placed after a given gene symbol, it is implied that there is reason to believe that the character effect associated with the gene symbol has been produced by a position effect of the inversion noted. When the small letter "h" directly follows, without spacing, the symbol "In" it is further indicated that, while one of the breaks of the given inversion lies close to the gene designated by the symbol, the other break lies in the heterochromatic region. So, for example, the full symbol for the genotype of mottled 4 would be w^{m4}, Inh.

The present nomenclature of alleles, in which the date of discovery is used in the superscript, is in general an unsatisfactory one because it is far too long. We stated, in a note sent in to the DIS in which the nomenclature of alleles was originally taken up, that such a symbolism would require an abbreviation of the abbreviation; but through some error this remark of ours was not included, and therefore we restate it here. Nevertheless, we have still followed this usage in some cases, although we intend to shorten our symbols of such alleles eventually.

In designating either inversions or translocations or other rearrangements, the ideal scheme would, of course, be, except in cases of very well-known ones that can be shown by a simple symbol and will then be widely understood, to give the locus of at least the more important break in the chromosome, either by the symbol of a known gene very close to the break or by the designation of the position of the break in the salivary chromosome as mapped by Bridges. This is particularly desirable when for any reason the translocations or inversions are being used primarily because they have the break in the specific position which it occupies. We have a number of these translocations, involving chromosomes X and 4, as shown in another article, and in these cases the position of the break in the X is known and the translocation has been designated according to this position. The scheme then is for the symbol to begin "TX" and then to state in parentheses the approximate point at which the break in the X occurs as shown in Bridges' map, and after the parenthesis to designate the other chromosome involved, in this case 4. Here, unlike the cases previously mentioned, a parenthesis is desirable so that the letters and numbers within will not be confused with those outside the parenthesis and will be understood to refer to the salivary map position. We have thus designated not only our own recently obtained translocations between X and 4 in this way, but also those from other sources which we have in our laboratory and whose positions of breakage in the X happen to be known, and we have arranged them in our stock list in order of the position of the break in the X, beginning at the left end.

Wallace, Bruce. A new proposal for the control of mites. Prompted by the success of the U. S. Army in its use of benzyl benzoate as a chigger preventive, we have made preliminary tests of the effects of this substance on "white" mites and D. pseudoobscura. Benzyl benzoate is an oily substance (boiling point, 323° C.) which is soluble in alcohol and insoluble in water. Water suspension can be made by adding water to alcohol solutions or, less successfully, by the use of soap. Mites placed on paper toweling treated with 2%-4% benzoate-alcohol solution from which the alcohol has evaporated die in three to five minutes. Adult D. pseudoobscura walk and rest on such toweling in culture bottles with impunity. Toweling treated with 8% solution, however, seems to be injurious to the flies. Developing larvae crawl onto treated paper to pupate and emerge as imagoes without harm. One cc. of 5% benzoate-alcohol solution was poured over and worked into the food of a culture bottle. A small peice of food

containing developing larvae was placed in this treated culture. Larvae made their way into the untouched portions in the bottom of the culture and later emerged to pupate. Population-cage experiments, which are invariably ruined by mite infections (see Wright and Dobzhansky, Genetics 31: 127, 1946), apparently can be saved by the use of benzyl benzoate. Two methods have been tried to date. In the first, the infected cage was generously sprayed (Holmspray atomizer, 98 cents) inside and out with a 2% alcoholic solution. Cups were wiped and food surfaces were sprayed with the same solution. Adults were drowned by the spraying, but larvae within the food survived and have now emerged from pupae as flies. The second method consists of merely transferring cups from the infected cage to a new one after careful wiping and spraying of the food surface. No mites have reoccurred in cages treated by these methods. Cages are now sprayed periodically in an attempt to prevent mite infections. All vulnerable exterior cracks, crevices, corks, plugs, and such are sprayed lightly every two weeks. We hesitate to recommend this treatment without reservation because of the small amount of experimentation we have done with it. However, our preliminary results seem to justify our recommendation that benzyl benzoate be used experimentally on other types of mites and other species of Drosophila. It was through the kind suggestion of Dr. H. T. Spieth of Columbia University that our attention was called to the use of benzyl benzoate. Dr. Charles Michener of the American Museum of Natural History supplied information about the concentrations most likely to be effective.