GENETIC MAPS OF THE AUTOSOMES IN DROSOPHILA PSEUDOOBSCURA

C. C. TAN

Wm. G. Kerckhoff Laboratories of the Biological Sciences, California Institute of Technology, Pasadena, California

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INTRODUCTION

DROSOPHILA PSEUDOOBSCURA is of considerable interest as an object for the study of a number of genetic problems, especially those connected with the hybrid sterility and the methods of race and species formation. As a basis for these studies it is necessary to work out the genetic and the cytological maps of the chromosomes of this species. The present paper deals with the genetic maps of its autosomes.

MATERIALS AND METHODS

Since the two races of *Drosophila pseudoobscura* are different in gene arrangement—they differ in an inversion in each limb of the X chromosome and an inversion in the second chromosome (TAN 1935a,b, KOLLER 1936)—and since strains of the same race from different geographic origins may also differ in gene arrangement, it becomes imperative for the purpose of this work to use always a definite standard strain. Race A strain from Georgetown, Texas, was chosen.

All mutant stocks of race A *D. pseudoobscura* kept in this laboratory were used. The author is indebted to Miss BEERS of the University of Southern California for the use of some mutants she discovered. Some mutant stocks were sent here from the Genetics laboratory of Edinburgh University, Scotland, through the kindness of Drs. KOLLER and CREW, to whom the author wishes to express his appreciation. In the course of the investigation a few new mutants were discovered by the author and some of them proved valuable because of their favorable locations. Prof. STURTEVANT kindly let me use some of the new mutants he had found.

For linkage experiments all cultures were raised in an incubator at $25 \pm .5°C$.

DESCRIPTION AND LOCALIZATION OF MUTANT GENES

Since the present paper is confined to the autosomes of *D. pseudoobscura*, only the autosomal mutants will be briefly described below. In each chromosome mutants will be described more or less in the order of their discovery.

The second chromosome

Bare (Ba). Bare was discovered by STURTEVANT (1934) in race A. It is characteized by the shortening of all the macrochaetae. The character can be easily distinguished from the wild type, and the viability and productivity of the heterozygous flies are excellent. Homozygous Bare flies are occasionally viable.

Smoky (Sm). Smoky, another dominant gene in this chromosome, was found by Miss BEERS (1934) in race B. It has been successfully transferred to race A. The mutant gene causes the thickening and branching of wing veins, especially the second longitudinal vein. The minimum expression is

TABLE 1

Two point linkage experiments.

LOCI	TYPE OF CROSS	NON-CROSSOVERS		CROSSOVERS		TOTAL
		Chromosome II				
bi Ba	*bi*×*Ba*	135	145	21	7	308
		Chromosome III				
Sc cv	*Sc*×*cv*	181	175	141	158	655
		Chromosome IV				
in j	*in j*×+	199	177	59	26	461
in j	+×*in j*	354	341	54	73	822
in Cy	*in*×*Cy*	128	104	117	119	468
j tg^3	+×*j* tg^3	149	157	103	112	521
j Cy	*Cy*×*j*		886		235	1121
tg^3 *Cy*	tg^3 *Cy*×+	261	288	30	54	633

the formation of a small delta-like structure at the distal end of the second longitudinal vein. It is invariably lethal when homozygous. Smoky also has the effect of roughening the surface of the eyes. By crossing to Bare and backcrossing the F_1 Bare Smoky females to wild type males, it was found that the crossing over frequency between the two exceeds 40 per cent.

Glass (gl). The recessive mutant glass was discovered and described by CREW and LAMY (1935a). The eye is reduced in size and surrounded by a smooth colorless rim. The pigment in the central space appears to be greatly reduced in amount and leaves only a pinkish or reddish hue. The character can be easily recognized with the naked eye. The viability and fertility are about as good as in wild type. In a cross between *Sm Ba/gl* ♀ ×*gl* ♂, the result (table 2) shows crossing over values of 42.0 per cent between Bare and Smoky, 18.9 per cent between Bare and glass, and 46.3 per cent between Smoky and glass. Since the smallest number of flies were represented by Bare-glass and Smoky classes, which must be double cross-

overs, the sequence of the three genes in the chromosome must be Smoky-Bare-glass, Bare being nearer to glass than to Smoky.

Bithorax (bi). The gene bithorax was first reported by CREW and LAMY (1934b) as an autosomal recessive mutant, causing an enlargement of the balancers. In extreme cases, the balancers may take the form of wing-like organs. The mutant flies are relatively weak and their viability is low. Sometimes the character may overlap normal.

DONALD (1936) described the mutant in detail and reported that the locus of bithorax is 25 units from glass. My data from the cross *bi gl/Sm Ba* ♀ to *bi gl* ♂ (table 3) show that bithorax is 28.9 units from glass. Between

TABLE 2

Three point linkage experiments.

LOCI	TYPE OF CROSS	NON-CROSSOVERS		SINGLE CROSSOVERS				DOUBLE C.O.		TOTAL
				REGION 1		REGION 2		1, 2		
				Chromosome II						
Sm p ps	*Sm p ps*×+	134	59	50	95	81	45	39	39	542
Sm Ba gl	*gl*×*Sm Ba*	90	164	137	52	36	27	20	20	546
bi p Ba	*bi p*×*Ba*	226	206	20	9	7	5	4	4	481
bi p ps	*bi p ps*×+	223	138	10	9	165	89	7	4	645
bi gl ps	+×*bi gl ps*	119	21	44	21	24	10	6	5	250
				Chromosome III						
or Bl Sc	*Bl Sc*×*or*	160	175	19	13	34	39	1	1	442
or ab pr	*or ab pr*×+	251	146	9	29	182	94	7	18	736
or Sc pr	*or Sc pr*×+	74	45	17	19	23	17	5	6	206
				Chromosome IV						
in j tg^3	*in j tg*^{3}×+	172	90	21	59	70	104	13	5	534
tg^3 *Cy Ro*	*Ro*×*tg*^{3} *Cy*	157	160	19	11	5	2	0	0	354

glass and bithorax lies the gene Bare, which gives 21.2 per cent of recombinations with glass on one side and 9.7 per cent with bithorax on the other (table 4).

Pink (p). The gene for pink eye color was found in a cross of the La Grande-2 strain to orange Scute purple by STURTEVANT in 1934. It is recessive and located near Bare. Pink itself is variable and sometimes overlaps wild type. However, in combination with orange, a third chromosome gene, it can be easily classified. As a matter of fact, orange alone gives a bright red eye color (like vermilion), while in combination with pink it gives a true orange color.

According to STURTEVANT, pink is located close to Bare and the two gave only a few per cent of crossing over. An experiment involving the cross of *bi p/Ba* ♀ with *bi p* ♂ gave 7.7 percent of crossing over between

bithorax and pink and 4.2 per cent between pink and Bare (table 2). The sum of the two, 11.9, is somewhat higher than the value, 9.7 (table 4), that was obtained directly between bithorax and Bare. Despite the unexpectedly high number of observed double crossovers, it is justifiable to put the gene pink between bithorax and Bare.

Pauciseta[1] (*ps*[1]) *and its allele pauciseta*[2] (*ps*[2]). The gene pauciseta[1] (*ps*[1]) was discovered by Miss GROSCURTH in the Chehalis-4 strain (race A), several generations after this strain had been derived from a single female caught in nature. The mutant is characterized by the absence of some bristles, especially the anterior dorso-centrals and the scutellars. In some cases, however, all bristles may be present, but one or both anterior dorso-centrals become somewhat more slender than normal. Unless the bristles are carefully examined, pauciseta[1] can be easily overlooked.

Pauciseta[2] was found by the author as a single female in progeny of a cross between *ct y m sn v se/s*[2] ♀ with *ct y m sn v se* ♂ in March 1936. The fly had some bristles missing on both the thorax and the scutellum. It was mated with wild type males and produced exclusively wild type flies in F_1, which in F_2 produced 365 wild type and 122 pauciseta[2]. When pauciseta[2] males were mated to bithorax glass pauciseta[1] females, the progeny showed the pauciseta character, indicating that the new pauciseta is an allele of the old one. Since the two are indistinguishable, the new pauciseta may be designated as *ps*[2].

To localize the gene pauciseta in the second chromosome, two sets of experiments were carried out. They were *bi p ps/+* ♀ by *bi ps ps* ♂ and *bi gl ps/+* ♀ by *bi gl ps* ♂. The result as summarized in table 2 enables us to localize the gene pauciseta on the right end of the second chromosome.

The third chromosome

Orange (*or*). Orange eye color was found by LANCEFIELD and later described by CREW and LAMY (1934a). It is recessive and cannot be distinguished from the sex-linked vermilion eye color. The mutant can be easily classified even in combination with most other eye colors. The viability and productivity of the mutant fly are as good as in normal.

Purple (*pr*). The gene purple, another recessive eye color mutant, was found by CREW and LAMY (1932). It is a translucent color ranging from yellowish brown to chestnut. In males the testicular sheath appears colorless. When it is combined with orange, the eye becomes grayish white in color.

Scute (*Sc*). Scute, a dominant gene causing the absence of most bristles on the thorax and the head, was also reported by CREW and LAMY (1934a). The homozygous forms of Scute can be distinguished from the heterozygotes by the rough eyes and the absence of some microchaetae. Accord-

TABLE 3

Four point linkage experiments.

LOCI	TYPE OF CROSS	NON-CROSSOVERS		SINGLE C.O.						DOUBLE C.O.						TRIPLE C.O.		TOTAL
				REG 1		REG 2		REG 3		I, 2		I, 3		2, 3		I, 2, 3		
										Chromosome II								
Sm bi Ba gl	*bi gl*×*Sm Ba*	186	210	199	100	17	20	78	45	28	13	60	28	1	7	7	1	1000
										Chromosome III								
or ab Ja pr	*or ab pr*×*Ja*	286	146	14	43	44	24	108	45	4	8	4	4	6	2	1	2	741
or Ja Sc pr	*or Sc pr*×*Ja*	205	152	36	37	15	10	42	54	1	1	5	5	4	3	0	5	575
or Ja pr cv	*or pr cv*×*Ja*	273	202	39	40	85	98	59	68	12	7	17	4	7	8	2	1	922

ing to CREW and LAMY (1934a) Scute is located half way between orange and purple, each of which gives about 25 per cent of crossing over with Scute. Due to its favorable location and its clear cut expression, Scute has proved to be an extremely valuable mutant in this group.

Crossveinless (cv). The mutant crossveinless was reported by CREW and LAMY (1934b) as an autosomal recessive, which causes the absence of the posterior crossvein. The anterior crossvein may either be absent or incomplete. The mutant is easily distinguishable, if the wing is not damaged. According to DONALD (1936) crossveinless is located 17 to 26 units from

TABLE 4

Total data for each pair of loci.

CHROMOSOME II			CHROMOSOME III			CHROMOSOME IV		
LOCI	% OF CROSSING OVER	TOTAL FLIES EXAMINED	LOCI	% OF CROSSING OVER	TOTAL FLIES EXAMINED	LOCI	% OF CROSSING OVER	TOTAL FLIES EXAMINED
Sm bi	43.6	1000	*or Bl*	7.7	442	*in j*	17.1	1817
Sm p	41.2	542	*or ab*	9.7	1477	*in tg*	47.6	534
Sm Ba	22.8	1546	*or Ja*	15.8	2235	*in Cy*	50.1	468
Sm gl	46.5	1546	*or Sc*	21.8	1223	*j tg*	38.6	1055
Sm ps	50.0	542	*or pr*	36.7	3180	*j Cy*	41.9	1121
bi p	5.9	1126	*or cv*	42.5	922	*tg Cy*	11.5	987
bi Ba	9.7	1789	*Bl Sc*	17.0	442	*tg Ro*	10.5	354
bi gl	29.2	1250	*ab Ja*	12.3	741	*Cy Ro*	2.0	354
bi ps	41.6	895	*ab pr*	36.7	1477			
p Ba	4.2	481	*Ja Sc*	6.8	575			
p ps	39.6	1187	*Ja pr*	23.6	1497			
Ba gl	21.2	1546	*Ja cv*	38.0	922			
gl ps	18.0	250	*Sc pr*	21.6	781			
			Sc cv	45.6	655			
			pr cv	18.0	922			

purple. This is in close agreement with my data, which give 18.0 per cent of crossing over between purple and crossveinless (table 4).

Jagged (Ja). Jagged wing is a dominant mutation found by the author. Usually, only the inner margin of the wing is notched. In extreme cases, the whole wing may become strap-like and bear notches on all margins. Three such flies, two females and one male, appeared in a cross between *bi gl/Sm Ba* ♀ and *bi gl* ♂ in December 1935. A single Smoky Bare Jagged female was successfully crossed to three wild type males, producing 52 Jagged and 63 non-Jagged flies, equally distributed among both sexes. This shows that Jagged is an autosomal dominant mutation.

The test for the viability of the flies homozygous for Jagged wing indicates that Jagged is lethal when homozygous.

The locus for Jagged has been determined, by the combined results of

several different type of four-point crossing over experiments (table 3), to lie 6.8 units to the left of the gene Scute (table 4).

Abrupt (*ab*). Abrupt longitudinal vein is a recessive mutant found by the author. It produces a shortening of the fourth longitudinal vein. In extreme cases, the vein may abruptly stop just below the posterior crossvein. The mutant cannot be distinguished from either short, a sex-linked gene causing the shortening of the third and fourth longitudinal veins, or short-4 which was described by CREW and LAMY (1935a) to produce the shortening of the fourth and fifth longitudinal veins. In order to avoid confusion, separate names are here proposed to designate these mutants. Short (*s*) remains to designate the sex-linked one. The fourth chromosome one, originally known as short-4 (s_4), is renamed incomplete (*in*). The name abrupt (*ab*) applies to the one in the third chromosome, which is now under discussion.

The origin of abrupt can be traced back to the orange purple stock. In three cultures of *na/or Sc pr* ♀ by *or pr* ♂, there appeared several males whose wings were indented at their inner margin. The gene concerned was found to be allelomorphic to the beaded originally described by LANCEFIELD (1922). It is known as bd^2. From a cross between *or* bd^2 and *or* sibs, three *or pr* females appeared to have their fourth longitudinal vein shortened. By mating them to wild type sibs, a good many orange abrupt purple flies of both sexes were obtained. Since all abrupt flies had orange purple eye color, the mutant must have originated in the *or pr* stock. When *or ab pr* flies were mated to incomplete (*in*), all F_1 individuals were wild type, indicating that abrupt and incomplete are not allelomorphs. An *or ab pr* stock was soon established. At the beginning some abrupt flies appeared to overlap normal. But after several generations of selection and inbreeding, the character became more pronounced and at the same time bred true.

The locus for the gene abrupt has been found to lie between orange and Jagged. As shown in table 4, it is closer to the former (9.7 units) than to the latter (12.3 units).

Blade (*Bl*). Blade wing is a dominant discovered by STURTEVANT (unpublished). The wing assumes a blade-like shape. It is easily classifiable and has normal viability and productivity. STURTEVANT crossed *or/Bl Sc* ♀ to *or* ♀ and found Blade to give 7.7 per cent of crossing over with orange and 17.0 per cent with Scute. With his permission the result of this cross is also included in table 2.

The fourth chromosome

Curly (*Cy*). Curly, a dominant gene, was discovered by Miss BEERS (1934) in race B. The wing may be curled either upward or downward.

Flies homozygous for Curly are always inviable. In heterozygous condition, the mutant fly is fully viable and fertile. Curly can be easily distinguished from normal. The gene has been successfully transferred from race B to race A.

Jaunty (*j*). The gene jaunty was reported by CREW and LAMY (1935a). It is recessive, and the mutant flies show a slight upturning of the tip of the wing. Occasionally, it overlaps normal.

Incomplete (*in*). The gene incomplete, another fourth chromosome recessive found by CREW and LAMY (1935a), was originally described by them as short-4. According to them incomplete and jaunty gave about 10 per cent of crossing over. Recently, DONALD (1936) reported that the two gave 15 to 16 per cent of recombinations. My data as shown in table 4, giving 17.1 per cent of crossing over, is in close agreement with the result of DONALD.

Multiple alleles of tangled (*tg*). The first mutant of tangled, tangled[1] (*tg*[1]), was discovered by CREW and LAMY (1934b), who described it as fused. Later (1935a) they changed the name to tangled. It is recessive; in the mutant flies the second and third longitudinal veins come together at their distal ends, often with extra crossveins. The same also occurs with the fourth and fifth longitudinal veins. In extreme cases, the wing may be tilted up.

A single tangled[2] male, similar to tangled[1], except for having few extra veins, was found in Bare orange Curly stock. It was mated to wild type females, and in F_2 several *Ba or Cy* tg^2 and *or* tg^2 flies were obtained.

A single tangled[3] male fly found in the Curly stock had only one extra crossvein connecting the distal ends of the second and the third longitudinal veins. It was mated to wild type females, giving in F_1 all wild type flies and in F_2 8 tg^3 and 435 wild type ones. The great excess of wild type flies over the mutant type may be accounted for by presence of some modifier in the wild type parent, for after several generations of selection, tg^3 started to breed true.

A fourth allele of tangled, tg^4, was obtained by Dr. STURTEVANT in the echinus stock. The mutant flies resemble very much either tg^1 or tg^2. When they were crossed to each of the other three tangled alleles, all offspring were tangled.

Among the four alleles of tangled, tg^3 was most favorable for experimental purposes, because tg^3, having only one clear extra crossvein instead of the many characteristic of tg^1, tg^2, or tg^4, can be distinguished in combination with other fourth chromosome wing mutants. Moreover, tangled[3] wings, unlike other alleles of the same locus, are never tilted at the tip. Hence, for genetic analysis of the locus, tg^3 flies have been almost exclusively used.

According to DONALD (1936) tangled and jaunty gave 44 to 49 per cent of crossing over and the sequence of the genes was assumed to be incomplete-jaunty-tangled. My data, given in table 4, are in agreement with DONALD's in regard to the sequence of genes but show less crossing over between *in* and *j*. This difference is probably due to the fact that the tangled used by DONALD was tg^1, which, being more extreme, may obscure the classification of flies in the combination of tangled and jaunty.

Rough (*Ro*). Rough is a dominant eye mutation found by Miss GROSCURTH in race A. It causes irregular arrangement of eye facets. Not infrequently the mutation overlaps normal. A single experiment involving the cross of tg^3 *Cy/Ro* ♀ by tg^3 ♂ gave 9.5 per cent of crossing over between tg^3 and *Cy* and 2.0 per cent between *Cy* and *Ro*. The absence of *Ro Cy* and tg^3 flies indicates that *Ro* is located to the right of *Cy*.

CONSTRUCTION OF THE GENETIC MAPS

According to MORGAN, STURTEVANT and BRIDGES (1925) four autosomal groups of genes were reported by LANCEFIELD with 4, 6, 1 and 2 recognized loci respectively. These, together with the sex-linked group, made the total number of linkage groups 5, which is equal to the haploid chromosome number of the species. In spite of the facts that the autosomal linkage groups reported by LANCEFIELD were imperfect, and that most of his mutants are now lost, some of them have been found to be the same as some of the ones described above. For this reason, the numbering of the second and the third linkage groups still follow the system of LANCEFIELD. The numbering of the fourth linkage group is justified on the basis of the cytological evidence which shows that the representative gene of this group is located in the rod-shaped chromosome and not in the chromosome now designated as the fifth, in which no gene is represented here.

A summary of linkage data is presented in tables 1–3. The first column at the left shows not only the loci concerned but also their sequence. Under the column headed "type of cross," types of two parents involved in the cross are shown, the one on the left being female and the one on the right male. In the third and the following columns classes are entered under the headings indicating the type of crossing over they represent. In every case the class which includes the individuals bearing the wild type allele of the most left-hand locus concerned is placed first, and is followed by the contrary class. The results are, of course, obtained from the backcrosses of the F_1 hybrids to multiple recessives.

Table 4 shows the total data for each pair of loci in the second, third and four chromosomes respectively. The resulting maps are shown in figure 1.

It is, of course, realized that these maps are only preliminary ones, and

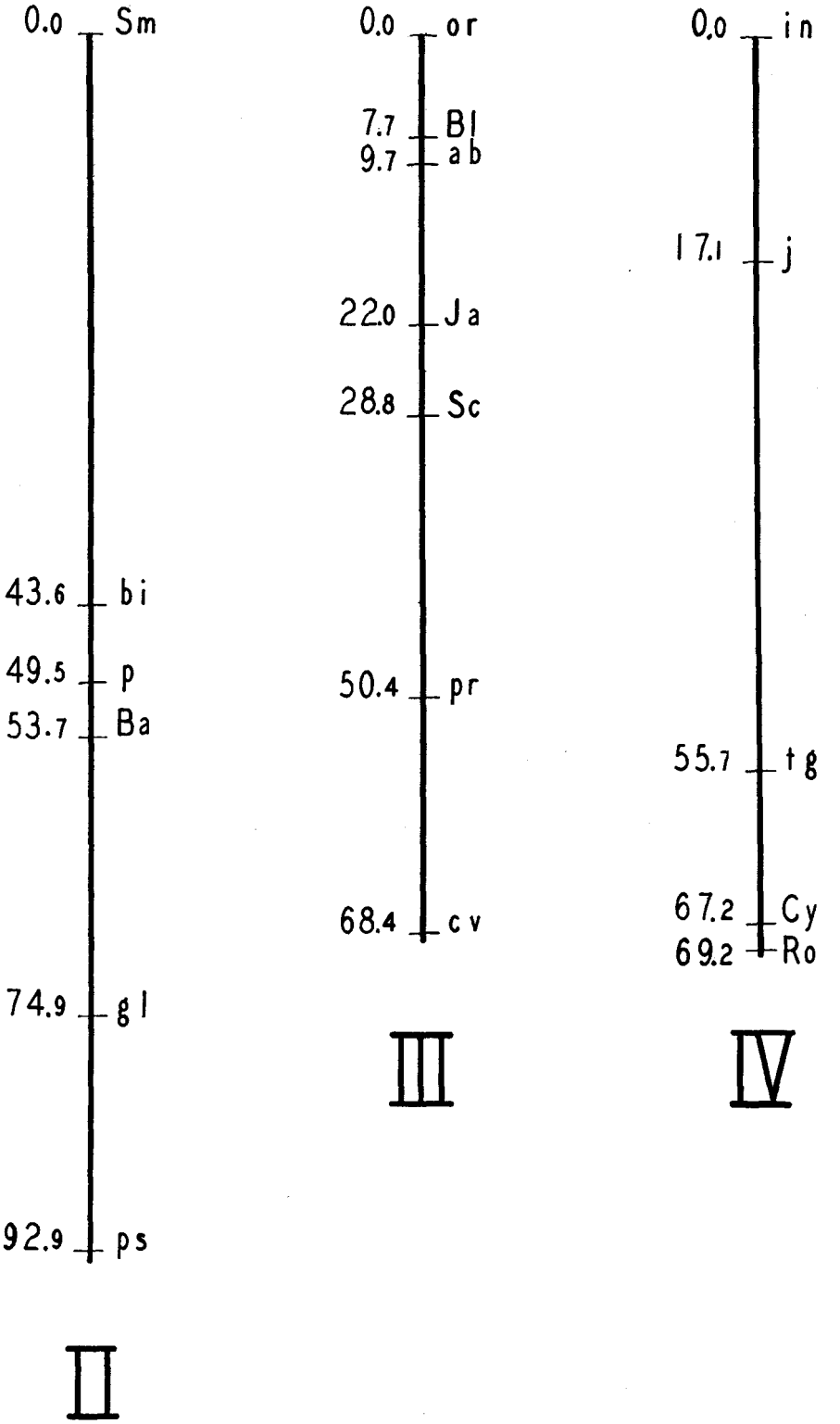

FIGURE 1.—Genetic maps of the three autosomes of *Drosophila pseudoobscura*. II—the second chromosome, III—the third chromosome, and IV—the fourth chromosome.

that both the total map-lengths and the distance between the genes may prove to be larger than here indicated. The data presented in table 4 are in general self consistent. Several apparent discrepancies may, however, be noticed; for instance, the value shown between Smoky and bithorax is slightly higher than between Smoky and pink or Smoky and Bare, although the maps show the reverse relation. Similar discrepancies are observed in the values between crossveinless and Scute and between crossveinless and Jagged. These discrepancies are probably due to differential viability of the classes concerned in different experiments.

In determining the map distances, the genes lying nearest to the spindle fibre attachment in each chromosome are taken as the zero points or the left ends. The evidence leading to the determination of the locations of the genes in relation to the spindle fibres will be published elsewhere in connection with the cytological maps. Suffice it to mention here that the loci known to lie nearest to the spindle fibre attachments are Smoky (*Sm*) in the second, orange (*or*) in the third, and incomplete (*in*) in the fourth chromosome. Hence, these loci are taken as zero points.

Recently, CREW and LAMY (1935a) attempted to homologize the second chromosome of *D. pseudoobscura* with the left arm of the V-shaped third chromosome of *D. melanogaster*, and the fourth chromosome of *pseudoobscura* to the right arm of the V-shaped second chromosome of *melanogaster*. In view of the facts that relatively few genes are now known in *D. pseudoobscura* and that the two species cannot be crossed to effect a direct test, it seems to the author that attempts to establish the homology of genetic maps of the two species are rather hazardous. Indeed, DOBZHANSKY and TAN (1936) show that even the two closely related species, *D. pseudoobscura* and *D. miranda*, have no single chromosome in common. *D. pseudoobscura* and *D. melanogaster*, being much less closely related, have the gene arrangement in their chromosomes altered beyond recognition.

The author wishes to express his gratitude to Professors TH. DOBZHANSKY and A. H. STURTEVANT for encouragement, advice and use of their stocks.

SUMMARY

1. The mutants belonging to the three autosomal linkage groups of *Drosophila pseudoobscura* are briefly described.

2. The genetic maps of the three autosomes of the species are constructed as shown in figure 1.

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