CHAPTER VIII

CROSSING OVER AND CHROMOSOMES

THERE are several occasions in the maturation period of the germ-cells when it would seem that there might be an opportunity for an interchange between like chromosomes. Such an occasion might be found at the time when the thin threads twist around each other; or it might be found after fusion of the threads, or possibly after a general breaking up of the chromosomes and reunion of the pieces. Unfortunately the cytological evidence does not furnish explicit information as to the stage at which interchange takes place.

It has also been suggested that crossing over may take place at a still earlier stage in the germ-tract, *i.e.*, long before the time of maturation, even in the early embryo. Fortunately, it has been possible to obtain critical genetic evidence showing approximately the time when crossing over takes place. This evidence was obtained by Plough in his work on the influence of temperature on crossing over in *Drosophila melanogaster.*

The way in which Plough's experiment was carried out was as follows: Females homozygous for three mutant factors in the second chromosome, *viz.*, black, purple, curved, were mated to wild-type flies. Some of these females were kept in an incubator, some in an ice-box, and some were kept at room temperature; under one or the other of these conditions they laid their eggs which hatched and produced larvæ and pupæ and flies. The daughters were then mated to black, purple, curved males, and remained under the same temperature conditions until their offspring hatched. It was found that there was more crossing over in the offspring of the pairs kept at a high and at a low temperature than in those kept at room

temperature. Later the crossing over values for intermediate points was also obtained, and from these data the curve shown in Fig. 56 was made.

At a low temperature (about 10° C.) crossing over is increased as compared with a somewhat higher temperature (18–27° C.). Room temperature (22° C.) lies in that part of the curve where there is the least amount of crossing over. The amount then rises suddenly until about 29° and remains high till 31° C. is reached. The apparent fall after this temperature, as shown by the curve, may not be significant. The flies fail to lay eggs or may die at about this point.

In the foregoing experiment the eggs, larvæ and adult flies had been kept continuously at the same temperatures. If, however, the heterozygous virgin females reared at high temperature are back-crossed to the triple recessive males, and kept afterwards at normal temperature (22° C.) it is found that only the first ten-day output of such females shows the high crossing over values. The value drops during the following ten days. If a correction is made for a change in the crossing-over value due to age—since age, as Bridges has shown, causes a lowering in the value—still the effect of the early period is found to have begun to disappear after ten days, and soon completely disappears.

In still another way the influence of temperature may be shown. Heterozygous females that had lived at normal temperatures are mated to triple recessive males, and then exposed for the first seven days to 31.5° C. At first the normal crossover values are found, as seen on comparing Fig. 40 with Fig. 39 which is the control. The latter drops slightly from the second to the eleventh day. About the eighth day the heat effects begin to show (Fig. 40), and there is a sudden and considerable rise in the curve, that lasts for ten days, when it drops back to normal, corresponding with the removal of the flies from the high to normal temperature, *i.e.*, after the seven-day exposure.

From data of this kind it is possible to locate the stage in the development of the egg when the heat is affecting it. If, for instance, we know how long after subjecting a

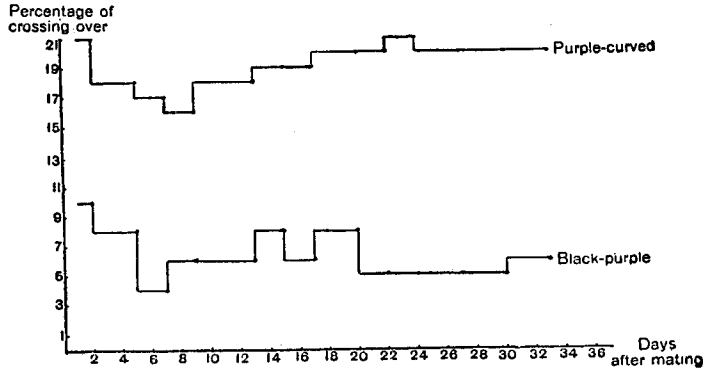

FIG. 39.—Curve showing the influence of temperature on crossing over; control. (After Plough.)

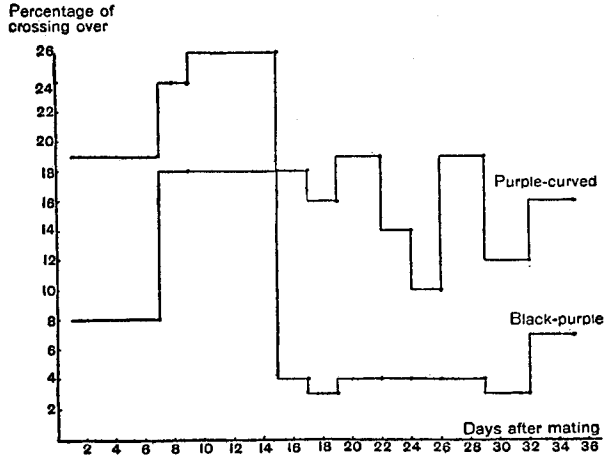

FIG. 40.—Curve showing the influence of temperature on crossing over. (After Plough.)

female to a high temperature, the effect of heat on crossing over begins to be observed in her offspring, and also how many eggs are laid by the female before this influence is manifest, we can tell approximately in what stages heat

affects crossing over. Furthermore, if we keep eggs, larvæ and pupæ in a high temperature, and then find out how many eggs have been affected by the high temperature, we can find out to what stage the eggs must have developed in order that crossing over may be influenced. Plough has made this calculation, and finds that only the eggs that have reached the stage where conjugation of the chromosomes takes place are affected—all the earlier stages are not influenced. It follows that the initial effect appears at about the time of conjugation of the chromosomes, but whether the crossing over occurs at this critical stage or some effect only is then produced that later affects the crossing over is not specifically shown. Nevertheless, I am inclined to think it more probable that the crossing over is actually changed at the time the heat acts (rather than afterwards), because in general most reactions of living things to environmental influence take place immediately rather than after a long interval. However this may be, the fact of prime importance in this work is that earlier than the period of conjugation of the chromosomes crossing over does not take place.

Expressed in numbers of eggs, the results show that in a just-hatched virgin female there are from 125 to 175 eggs that will be laid before the effects of heat are shown. In the females that have just hatched about 150 eggs are present that have passed beyond the conjugation period. This number (150) agrees with the estimated number of eggs (125–175) first laid that are not affected, and establishes the conclusion that after conjugation of the chromosomes crossing over cannot be influenced any more than it could before that period. The results clearly establish, then, that crossing over cannot be affected earlier than the conjugation, but can be affected at the time when the conjugation is known to occur.

As already pointed out, the chromosomes become drawn out into long threads at the synaptic period, and in many animals and plants these threads have been shown

to place themselves at this time in pairs. The double threads shorten later to take on the form of the ordinary chromosome. How the earlier, long thin thread (leptotene thread) is changed into a thick thread when the chromosomes condense is not known. According to several accounts the thread coils spirally within the wall of the "chromosome," at first in a loose coil, then in a tightly twisted coil. This idea of a coiled thread, or core, in a condensed chromosome is one that fits in very well with the idea that the thin thread represents the essential element in the chromosome that retains its original continuity even when the chromosome is condensed into a short rod or even into a ball. Unfortunately the evidence in favor of this view is by no means well established.

At the time when the threads conjugate, the evidence in several forms, such as *Batracoceps, Tomopteris,* etc., shows that when the conjugating pairs are U-shaped, the union begins apparently at both ends of the U at the same time. When the chromosomes are rod-shaped (in the last telophase) the evidence fails to show whether the union begins at both ends simultaneously or at one end only.

As the union between the threads progresses the parts not yet united can often be seen to be twisted about each other. They not only overlap, but they seem to be wrapped around each other.

Whether the threads are split lengthwise before their union can not be stated for all cases. It is certain that no splitting has been seen in several animals, but in one case (*Ascaris*) the threads have been found to be split lengthwise before they conjugate.

For a short time following the union of the threads they come in close contact with each other, and give the impression of having fused into a single thread. Usually before the nuclear wall breaks down to release the thick threads, a split can be seen again extending throughout the length of the threads. Not infrequently another longitudinal split appears in each half resulting from the

first split, so that four parallel strands appear. It is customary to call the split, that is supposed to correspond to the line of union of the maternal and paternal chromosome, the primary split or reductional split, and the split that corresponds to the longitudinal division within the

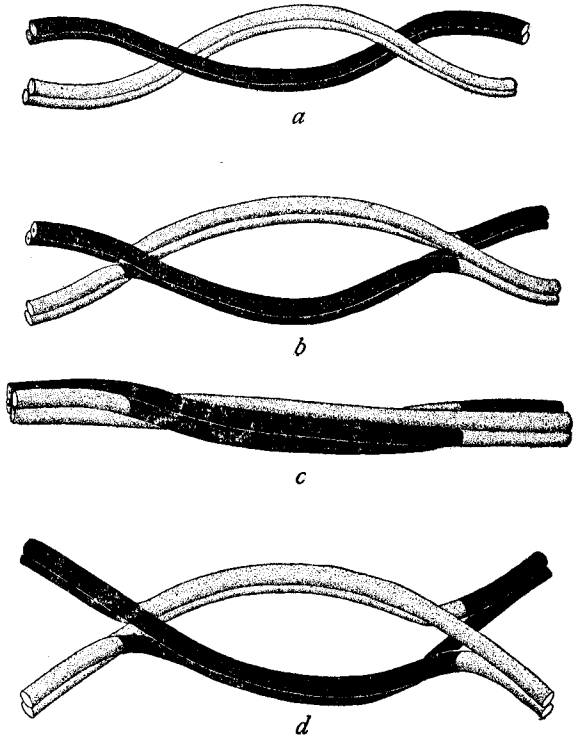

FIG. 41.—Diagram showing crossing over of two chromosomes at the four-strand stage *a*, *b*, and the subsequent opening out of the tetrad, *d*.

maternal or the paternal chromosomes, the secondary or equational split. Only in very special cases is it possible to be able to say which is the primary and which is the secondary split. In fact, whenever crossing over takes place in the four-strand stage this distinction fails to have much meaning.

There are certain questions connected with crossing over that are illustrated by the following models (Figs. 41, 42, 43). In these models of tetrads the dotted rod, split lengthwise, stands for a maternal chromosome, and each of its halves may be called a strand. The split in the rod is the secondary (or equational) split. The black rod, also split lengthwise, stands for the paternal chromosome.

In Fig. 41, *a*, the two split rods are represented as twisted about each other. If the two inner strands break and the cords interchange at the levels, where they first come into contact with each other (Fig. 41, *b*), and then

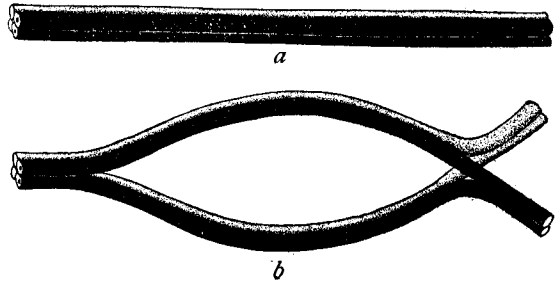

FIG. 42.—Scheme showing the opening out of the strands of the tetrad, *a*, in two planes; *b*, according to Robertson and Wenrich.

later the four strands come to lie side by side, *i.e.*, "fuse," the result will be that shown in Fig. 41, *c*. Two of the strands represent crossovers in the sense that an interchange has taken place between a maternal and a paternal strand; and if at the first spermatocyte division, when the threads begin to pull apart, the maternal separate from the paternal threads, two threads may be seen actually crossing each other (Fig. 41, *d*). They are here the two non-crossover strands, but if the two strands thrown to the left had been thrown to the right the two crossover strands would cross over. The scheme is essentially the same as the chiasma of Janssens, but the strands that cross may or may not (as here) represent the crossover strands.

The next two figures (Fig. 42, *a, b*) show how Robertson and Wenrich interpret the crossed threads, that they have observed in the spermatogenesis of some of the grasshoppers. The four strands are represented as conjugating side by side in Fig. 42, *a*. When the strands begin to open out preparatory to the first spermatocyte division the two maternal separate from the paternal at the ends of the tetrad, while in the middle of the tetrad the opening up involves the separation of a maternal and a paternal strand from a maternal and a paternal. In other words, the tetrad opens up in two planes at right angles to each other. This scheme also gives an appa-

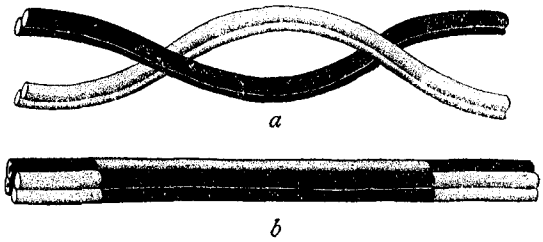

FIG. 43.—Scheme showing crossing over involving both strands of each chromosome.

rent crossing of the strands at the level where the opening out in one plane passes over into the opening out in the other plane, but there has been no real crossing over of the strands in the sense of interchange between them. Theoretically this explanation is sound, and moreover seems to be supported by observations in cases where the maternal and the paternal strands can be identified. The results undoubtedly show that the occurrence of crossed threads in cases where the split occurs in two planes does not necessarily imply that crossing over has taken place; but, on the other hand, as has been shown (in Fig. 41) a similar figure may also necessarily result after crossing over of the threads. In a word, the crossed-strand stage is not *ipso facto* evidence that it must have come about according to Robertson's

scheme. It should also be observed that the scheme rests on the assumption that no twisting has preceded the stage of the crossed threads, or, if such has taken place it has no relation to the resulting chiasma. Yet crossing of the threads is an observed fact.

A third scheme (Fig. 43, *a*, *b*) makes both maternal strands interchange with both the paternal ones. This scheme has at least one formal advantage over the other two in that it represents the four strands, after crossing over, as in position to lie side by side in the tetrad, so that the two longitudinal splits that reappear later lie in the same plane throughout their length. This seems more in accord with many of the observations that are recorded. If, during the following stages, the tetrads open out by the separation of the maternal from the paternal strands the crossed threads that result correspond to those in the first scheme (Fig. 41). At present it is not possible to decide between these different modes of representing crossing over. They may all occur. Their discussion shows little more than certain possibilities involved in the situation.

Details of Spermatogenesis

Some of the stages in the spermatogenesis of a grasshopper, *Phrynotettix,* as described by Wenrich, are shown in the following figures. The material furnishes certain details concerning the "resting stages" of the nuclei preceding synapsis more completely than any other, and it serves also to illustrate clearly the relationship of the chromosomes to the vesicles into which they pass (or which they form) during the resting stages. The figures also show how the threads emerge from the vesicles in which they appear to have been contained during the resting stages, and how the opening out of the tetrads in two planes gives the appearance of chiasma according to Wenrich.

During the time when the germ-cells are increasing in number by division there is a resting stage after each

division during which the chromosomes expand into a sort of vesicle, as seen by comparing Figs. 44, *a* and 44, *b*. An optical cross section of the stages shown in the last figure is represented in Fig. 44, *c*. An older stage is seen in Fig. 44, *d*. The stage of greatest diffusion of the chromatin material within its vesicle is seen in this figure, where the outlines of each vesicle are still visible. As the nucleus gets ready for another divi-

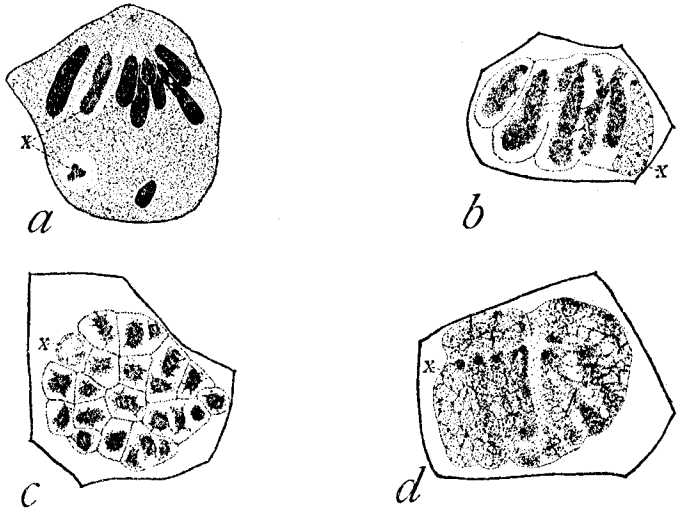

FIG. 44.—Spermatagonial cells in the last phase of division, *a*, and the following resting stages, *b*, *d*. (After Wenrich.)

sion the vesicles become more distinct (Fig. 45, *a*, *b*), and soon a coiled thread can be seen to be present in each vesicle (Fig. 45, *c*). As the thread thickens (Fig. 45, *d*), a longitudinal split appears in it, which indicates the plane of division of each chromosome at the next division.

At the last spermatogonial division, the chromosomes of the two daughter nuclei form vesicles, as they have done in earlier divisions (Fig. 46, *a* and *b*). But changes begin to take place that carry the chromosomes through a very different series of stages from those seen in preparations

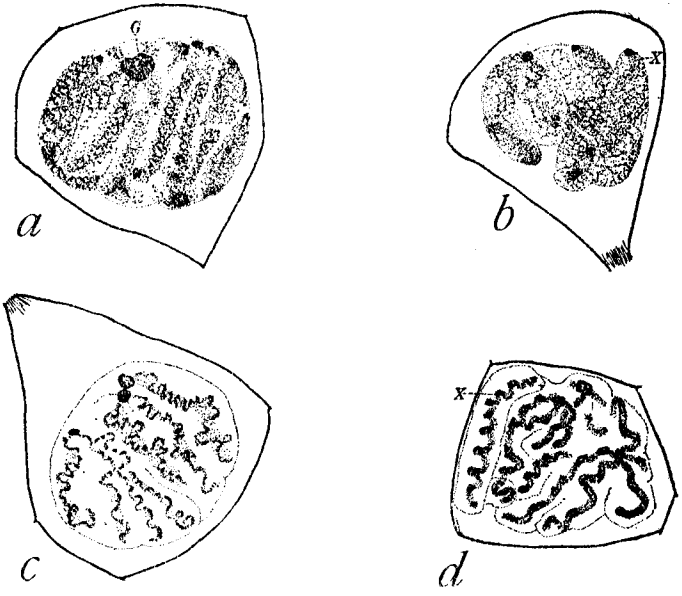

FIG. 45.—Cells emerging from the resting stages preparatory for the next spermatagonia division. (After Wenrich.)

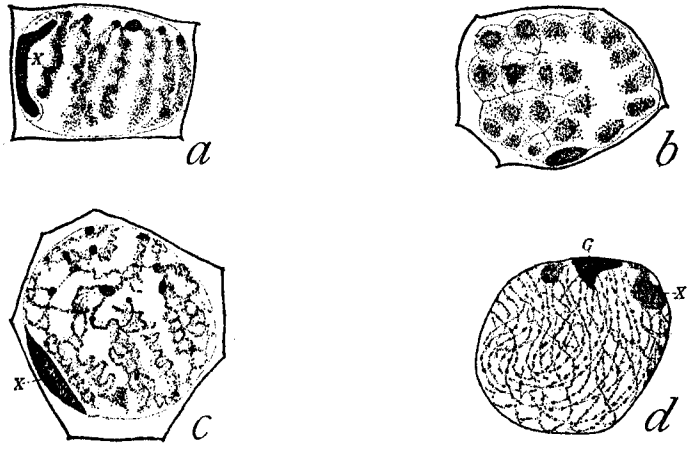

FIG. 46.—Cells emerging from their last spermatagonial division, *a*. *b*; passing into the synapsis stage, *c*, *d*; (After Wenrich.)

for the ordinary spermatogonial (or somatic) cell-divisions. Each chromosome vesicle begins to show a coiled thread (Fig. 46, *c*). Each thread next becomes longer and longer (Fig. 46, *d*) until the whole nucleus is filled with them. One or both ends can often be seen at the "distal pole" of the cell, where deep-staining nucleoli are present. The cells are now in the so-called thin thread, or leptotene stages.

The threads next come together in pairs beginning at the distal end of the chromosomes (the zygotene stage,

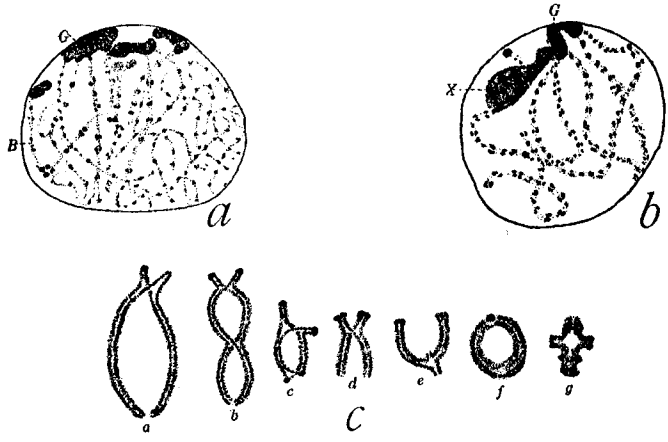

FIG. 47.—Formation of a thick thread after synapsis, *a*, *b*; and the following condensation of a tetrad, *c*. (After Wenrich.)

Fig. 47, *a*). When the fusion is complete and all the threads are double (Fig. 47, *b*), the stage is called the thick thread or pachytene stage. There are half as many threads now present as at the beginning. A longitudinal split is present in the chromosome throughout these stages along the line of fusion of the two thin threads. Wenrich identifies the split as the "primary split."

Another longitudinal split at right angles to the other one soon appears (Fig. 47, *c*), thus forming tetrads, each composed of four chromosomes. The tetrads next shorten,

opening out in various ways to produce figures like those shown in Fig. 47, *c*.

The sex-chromosome (*X*) that has no mate in the *Phrynotettix* male, and hence has not conjugated, has only one longitudinal split (a dyad). The cell, the primary spermatocyte, with its nucleus next divides. Eleven autosomes go to each pole, and the sex-chromosome failing to divide at this time goes to one daughter cell only. The secondary spermatocytes are produced—half with 12, half with 11 double chromosomes. A short resting stage follows —the chromosomes again becoming diffuse, *i.e.*, forming vesicles. They soon reappear and a second division takes place, producing the spermatids—the daughter cells of the secondary spermatocytes. Half of these have 12, half 11 chromosomes—the *X*-chromosome having divided at the second division.

Wenrich found it possible to identify certain of the chromosomes and was thus enabled to follow a few of them through several successive stages. Eight consecutive stages in the history of chromosome "*B*" of *Phrynotettix* are shown in Fig. 48. Indications of the primary split are present in *a, b, c,* the secondary split appears first in *d*. The evolution of the thread continues as the tetrad becomes placed in the spindle in such a way that the first separation of the chromosomes takes place along the secondary split, *i.e.*, the first division is equational. Wenrich found in several other individuals of this species that this same chromosome pair "*B*" consist of unequal members as shown in Figures 48, 2 *a–h* and 3 *a–d*. In 48, 2 *c* a distinct crossing of the threads is present. The shape of the contracted chromosome (*f g h*) and its position on the spindle show that one of the longer, and one of the shorter strands passes to one pole, and similarly a longer and shorter to the other pole. The division here is in the plane of the secondary split, *i.e.*, equational. The inequality in length of the conjugating pair makes this conclusion certain in this case.

In the second division of this chromosome the longer thread separates from the shorter one—the second is therefore reductional. It is evident, especially from this

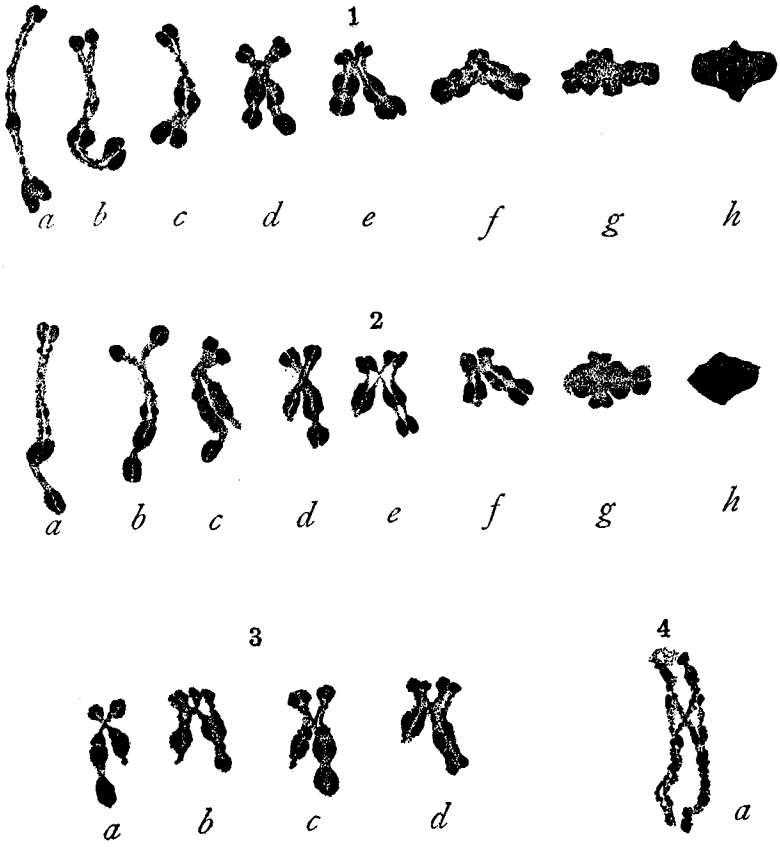

FIG. 48.—A pair of chromosomes "*B*" in conjugation, 1; the same pair in conjugation in another individual in which one chromosome is shorter than the other, 2; same in a third individual, 3; later stage showing chiasma of threads, 4. (After Wenrich.)

last example, that the crossing of the threads is not an indication that the division of the chromosome is necessarily different from what it is when there is no such crossing. What is more important is that the crossed

threads furnish no proof that an interchange must have taken place earlier, but neither does it furnish any evidence that interchange had not taken place. For example, the most obvious interpretation of Fig. 48, 2 *d* is that the upper end of the tetrad has separated in the plane of the secondary split (in anticipation, as it were, of the separation about to take place in this plane), and has separated in the lower part of the same tetrad in the plane of the primary split. This interpretation does not involve any real crossing over in the sense that the two crossed threads had previously broken and interchanged, as Janssens'

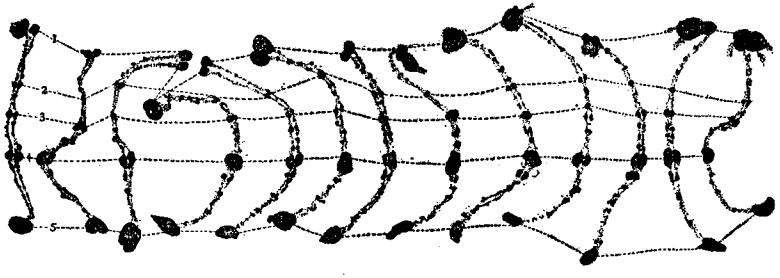

FIG. 49.—The same chromosome pair in conjugation from thirteen different cells. (After Wenrich.)

chiasmatype assumes on the ground that the two granules (threads) in contact at the upper end of the tetrad must be related to each other in the same way as are those further back in the tetrad.

This last assumption is the foundation of Janssens' view, but has no longer sufficient evidence to support it, even though none opposes it. Nevertheless, it should be clearly understood that evidence such as this, derived from Wenrich's results, can not possibly be held to show that an earlier interchange or crossing over has not occurred. If it had, such a figure as this (*c*) would, as explained above, be a consequence to be expected.

The constancy of the beading of the chromosomes in each individual is most remarkable. Its significance for

the linear order of the material of the chromosomes cannot be overestimated. As a further example Wenrich gives identical stages of the same chromosomes (Fig. 49), each of the figures is from a different individual. The identity in size and in location of the principal beads in the series is obvious.

Robertson has also brought forward a case of an unequal pair of chromosomes and interpreted the facts as opposed to the crossing-over hypothesis. He found two cases in a grasshopper of the genus *Tettigidea* in which there was a very unequal pair of chromosomes. The shorter piece conjugated consistently with only one part of the longer chromosome, as shown in the next figure

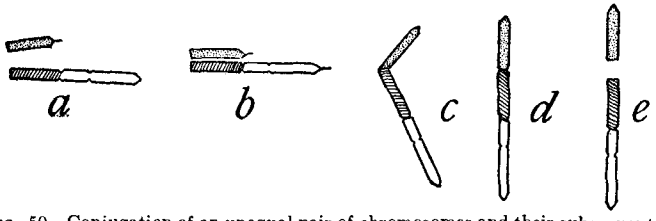

FIG. 50—Conjugation of an unequal pair of chromosomes and their subsequent separation. (After Robertson.)

(Fig. 50, *a, b*). At the first maturation division the two chromosomes separated, as shown in (*c, d, e*). It would be difficult to find a more excellent illustration of the persistence of the individuality of the chromosomes after conjugation, and the case falls equally in line with the view that conjugation takes place only between those parts of the chromosome that are alike, *i.e.*, composed of the same series of genes. How, then, could this case, so admirably suited to support the chromosome theory be turned against the chiasma theory? Only, I think, through a misconception of the essence of the theory. Robertson says: "In both types of unequal tetrads we have very strong evidence that the homologous chromosomes, on entering the side-to-side pairing process of synapsis, remain as distinct individuals, retain their identity throughout the period, and come

out of it with at least the same size they had on entering it. Each pairing chromosome maintains its distinct individuality during this period. This is opposed to the idea of Janssens ('09) and Morgan ('11), as expressed in the theory of chiasmatype. In their theory they assume that homologous chromosomes in parasynapsis twist about each other and fuse. On splitting, a plane passes down the fused body, regardless of the previous spiral fusion plane, resulting in two daughter chromosomes which may not be identical with the two chromosomes which entered the process. Each new one may contain parts of both original chromosomes. If such had been the case, the separation or formation of a short and a long chromosome out of the first chromosome with such regularity of size, etc., as we have shown, could not have occurred." On the contrary, even if crossing over had occurred within the region where the short and the long pieces came together, the separation would be expected still to be exactly that described by Robertson; for the genetic evidence points very clearly to the conclusion that the interchange involves exactly equal and opposite parts. There is no reason to suppose that regions outside the conjugating region would be affected; on the contrary, all the genetic evidence would lead us to expect no such effects.

Summary of Evidence

If we have found Janssens' evidence inadequate as a demonstration of crossing over, what other evidence is there in the history of the chromosome to which an appeal can be made? First, there is the undisputed fact that at the time when the chromosomes come together they spin out into long, thin threads which, as they meet, lie over and under each other, so that the line of fusion is in a spiral plan. Later, when the fusion is complete, it is no longer possible to follow the plane of union, but unless the chromosomes slip around each other after crossing

over—for which there is no evidence—one member of the pair must lie on one side of its mate in one region, and on the other side in other regions. Second, when the thick thread splits anew just before condensing into the tetrad it is so difficult to follow the course of the split in all cases that it cannot be affirmed that it always lies in one plane throughout the length of the chromosome, but if such should turn out to be the case, as so often figured, it would appear to mean that the crossing over had taken place and been obliterated by the time the condensation began. Third, evidence such as that described by Wenrich—of which sort there are other cases but none quite so clear—indicates that the chromosomes are enclosed in vesicles until they begin to spin out each into a long thread. Interchange of the sort called for by the genetic evidence could scarcely take place until the walls of the sacs had disappeared. The thin thread stage that follows would seem best to fulfill the conditions called for by the genetic evidence. The moment the primary split appears after the two threads have fused there would seem to be precluded any further chance for crossing over, as the genetic evidence suggests. This analysis leads, then, to the thin-thread stage as the most favorable stage for the requirments of the genetic evidence.

It is well known that most of our information about the maturation stages is derived from the male, because of the greater ease of obtaining the critical stages, and in preparing material. We are handicapped in discussing crossing over to a large extent by the fact that we must appeal largely to the evidence of spermatogenesis. In *Drosophila* at least there is no crossing over in the male. On the other hand, Nabours has recently found evidence in one of the grasshoppers that crossing over occurs both in the male and female. In this case evidence from the male would be more to the point. Whether genetic crossing over occurs in the male of *Batracoseps* and *Tomopteris*, we do not know.

In the female of some insects, amphibians, selachians and annelids, the thin-thread stages in the form of U-shaped loops have been described—stages that are so much like those of the male that the argument for one would seem to extend to the other. But again this proves too much, and we have yet to learn what cytological differences exist in cases where crossing over occurs in one sex and not in the other. On the whole, then, while the genetic evidence is favorable in all essentials to the theory of interchange between homologous chromosomes, it must be confessed that the cytological evidence is so far behind the genetic evidence that it is not yet possible to make a direct appeal to the specific mechanism of crossing over on the basis of our cytological knowledge of the maturation stages. The idea that the chromosomes disappear as such and go into some sort of suspension during the resting stage is an old idea. O. Hertwig thought that the chromosomes did actually "dissolve" at this time and "recrystallize" at each division stage. Goldschmidt elaborated a view of crossing over that rests on the assumption that the homologous genes are set free in the resting nucleus and may become interchanged during reconstruction. Aside from certain inherent contradictions in Goldschmidt's scheme (the most obvious ones have been pointed out by Sturtevant and by Bridges), it stands in contradiction to the one most certain fact that we know about crossing over, *viz.*, that not single genes but whole blocks of genes are involved—in fact, the most common sort of interchange involves the two entire pieces of each chromosome.

The general idea that the genes become dissociated during the resting phases is disproven by the way in which they come together. The genetic evidence from *Drosophila* shows that when crossing over occurs, let us say at the middle of the chromosome, all of the genes of each half of each pair hold together—and exchange as large pieces. Now if the genes are dissolved at each rest-

ing stage, there can be given no explanation as to why homologous genes should not recombine in all possible combinations with other genes. But this is exactly what does not happen. If it be supposed that the chromosomes dissolve only partly into chains of genes, it is still not obvious why the chains of one chromosome should be identical with those of the other (its homologue) as they must be to recombine properly; for, in neighboring nuclei other chains are forming—as the crossing-over results indicate—involving breaking at all possible levels.

Bateson and Punnett have proposed a theory of crossing over that is called reduplication. It is fundamentally different from the one here adopted. Although I think this theory outlawed by the evidence that Plough has obtained, and made impossible by certain other considerations that will be given later, the theory is so interesting that it may be briefly stated. Bateson suggests that at some time early in the embryo segregation may take place involving heterozygous pairs of factors. In the actual case presented only two such pairs are involved. As a "symbolic presentation" of the situation Bateson gives the diagram drawn in Fig. 51.

Although the dichotomous method of separation is utilized in the second line of figures to show reduction of the two pairs at once, such figures could obviously bear no relation to the ordinary process of cell division—nor do they, I understand, pretend to be. After separation (segregation) the cells that get *AB* and *ab* are represented as dividing faster than the cells *Ab* and *Ba,* hence there will be more of them in proportion as the two rates of division differ.

Bateson's view is open to the following criticisms:

1. The evidence from *Drosophila,* where many linkage ratios are known, gives no support to the view that these ratios fall into relatively few dichotomous schemes, such as Bateson's hypothesis calls for. Other forms also fail to fit such a view. On the contrary, the ratios fall into no

such groups as those given by Bateson. Even were it possible to suppose that in each case a different reduplication occurred (*i.e.*, a different number of generations was passed through), still, as said above, it is not obvious that the linkage series stands in any such numerical (*i.e.*, dichotomous) relation as the view demands.

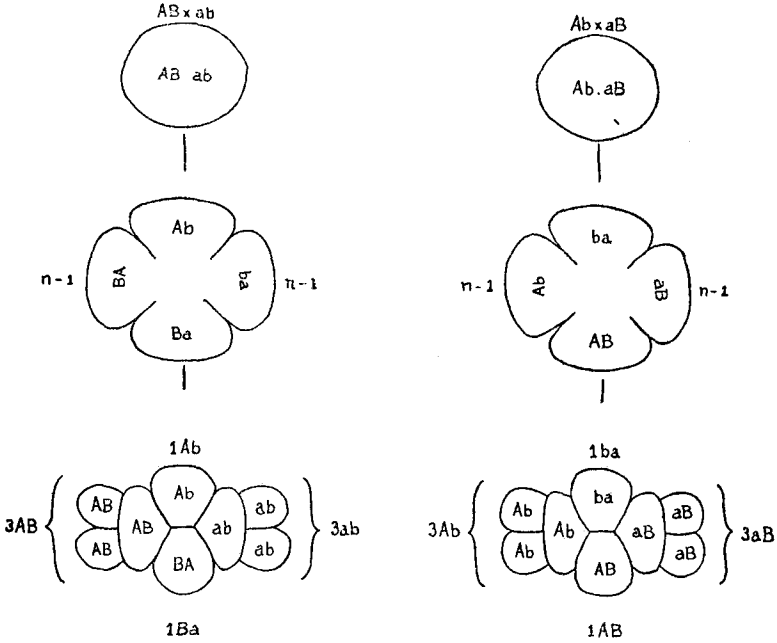

FIG. 51.—Two schemes illustrating the idea of reduplication by Bateson and Punnett; the three figures to the left illustrating "coupling," and the three to the right "repulsion."

2. If reduplication occurred at an early stage in the germ tract, we should expect to find in any organ of limited size, as a stamen, that there would be a likelihood that it would contain for the most part a particular kind of cell. Altenburg tested out this view with pollen of the primrose and found no evidence in favor of a limited distribution—on the contrary, he found that all the linkage combinations

were present in each stamen in the expected proportions. These and other difficulties make it improbable that linkage can be the result of this kind of reduplication.

Bateson and Punnett formulated their hypothesis at first for only two pairs of linked factors. When it was shown that three pairs of factors could show linkage, Bateson and Punnett assumed that all three pairs of factors might segregate at the same time (or in three successive divisions), the observed ratios being due, as before, to unequal division rates later. Trow has suggested that in such cases the segregation and reduplication for the third pair of factors might not occur until that for the first two pairs was completed. This view seemed to meet certain inadequacies of the former hypothesis, but meets with certain difficulties on its own account. One of the most obvious of these objections is, as Sturtevant has pointed out, that the number of cell divisions, necessary to produce some of the higher ratios that are known, would produce a mass of cells thousands of times larger than the animal itself.