Molecular Biology of the Gene Draft: Final Copy: Chapter 1

THE MOLECULAR BIOLOGY OF THE GENE

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Figure I-10- Schematic representation of how the sex chromosomes can determine sex. Here is shown a case in which males contain one x and one y chromosome, and females, two x chromosomes. This is the situation in both humans and Drosophila. In other species there is no y chromosome, and so diploid male cells contain one less chromosome than diploid female cells.

Figures I-11a and I-11b: The inheritance of a sex-linked gene in Drosophila. Genes located on sex chromosomes can express themselves differentially in male and female progeny. This is because, if there is one one x chromosome present, recessive genes present in this chromosome are always expressed. Here are shown two crosses, both involving a recessive gene (w, for white eye) located on the x chromosome. In (a) the male parent is a white-eyed (wY) fly, and the female, homozygous for red eye (WW). In (b) the male has red eyes (WY) and the female, white eyes (ww). The letter Y stands, here, not for an allele, but for the y chromosome, present in male Drosophila in place of a homologous x chromosome. There is no gene on the y chromosome corresponding to the (w) or (W) gene on the x chromosome.

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THE MENDELIAN VIEW OF THE WORLD

It is very easy to consider man unique among living organisms. He alone has developed complicated languages allowing meaningful and complex interplay of ideas and emotions. Great civilizations have developed and changed our world's environment in ways inconceivable for any other form of life. There has thus always been a tendency to think that something special differentiates man from everything else. This belief has found expression in man's religions, which try to find an origin for our existence and, in so doing, to provide workable rules for conducting our lives. It seemed natural to think that, just as every human life begins and ends at a fixed time, man had not always existed, but was created at a fixed moment, perhaps the same moment for man and for all other forms of life.

This belief was first seriously questioned just over 100 years ago when Darwin and Wallace proposed their Theories of Evolution, based upon selection of the most fit. They stated that the various forms of life are not constant, but are continually giving rise to slightly different animals and plants, some of which are adapted to survive and to multiply more effectively. At that time, they did not know the origin of this continuous variation, but they correctly realized that these new characteristics must persist in the progeny if they were to form the basis of Evolution.

At first, there was a great deal of furor against Darwin, most of it coming from people who did not like to believe that man and the rather obscene-looking apes could have a common ancestor, even if he occurred some 50-100 million years in the past. There was initial opposition also from many biologists who failed to find Darwin's evidence convincing. Among these was the famous Swiss-born

naturalist Louis Agassiz, then at Harvard, who spent many years writing against Darwin and Darwin's champion, T. H. Huxley, the most successful of the popularizers of Evolution. But by the end of the nineteenth century, the scientific argument was almost finished; both the current geographical distribution of plants and animals and their selective occurrence in the fossil records of the geological past were explicable only by postulating that continuously evolving groups of organisms had descended from a common ancestor. Today, the Theory of Evolution is an accepted fact for everyone but a fundamentalist minority whose objections are based not on reasoning but on doctrinaire adherence to religious principles.

An immediate consequence of the acceptance of Darwinian Theory is the realization that life first existed on our Earth some 1-2 billion years ago in a simple form, possibly resembling the bacteria, the simplest variety of life now existing. Of course, the very existence of such small bacteria tells us that the essence of the living state is found in very small organisms. Nonetheless, Evolutionary theory profoundly affects our thinking by suggesting that the basic principles of the living state are the same in all living forms.

The Cell Theory

The same conclusion is independently given by the second great principle of nineteenth century biology, the "cell theory". This theory, first put forward convincingly in 1839 by the German microscopists Schleiden and Schwann, proposes that all the larger plants and animals are constructed from small fundamental units called cells. All cells are surrounded by a membrane; and usually contain an inner body, the nucleus, which is also surrounded by a membrane, the nuclear membrane (Figure I-1 and I-2). Most important, cells arise only from other cells by the process of cell division. Most cells are capable of growing and of splitting

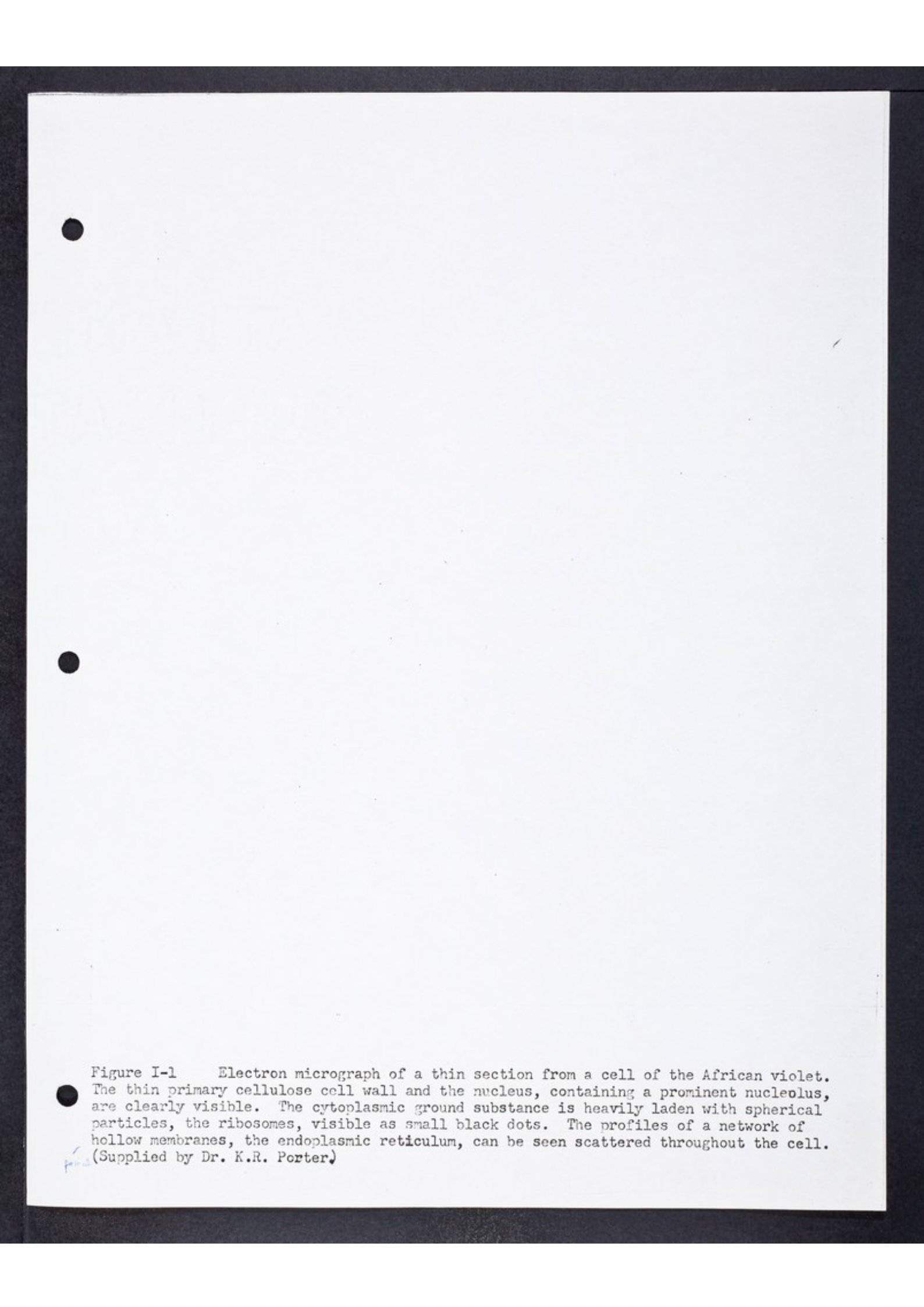


Figure I-1 Electron micrograph of a thin section from a cell of the African violet. The thin primary cellulose cell wall and the nucleus, containing a prominent nucleolus, are clearly visible. The cytoplasmic ground substance is heavily laden with spherical particles, the ribosomes, visible as small black dots. The profiles of a network of hollow membranes, the endoplasmic reticulum, can be seen scattered throughout the cell. (Supplied by Dr. K.R. Porter.)

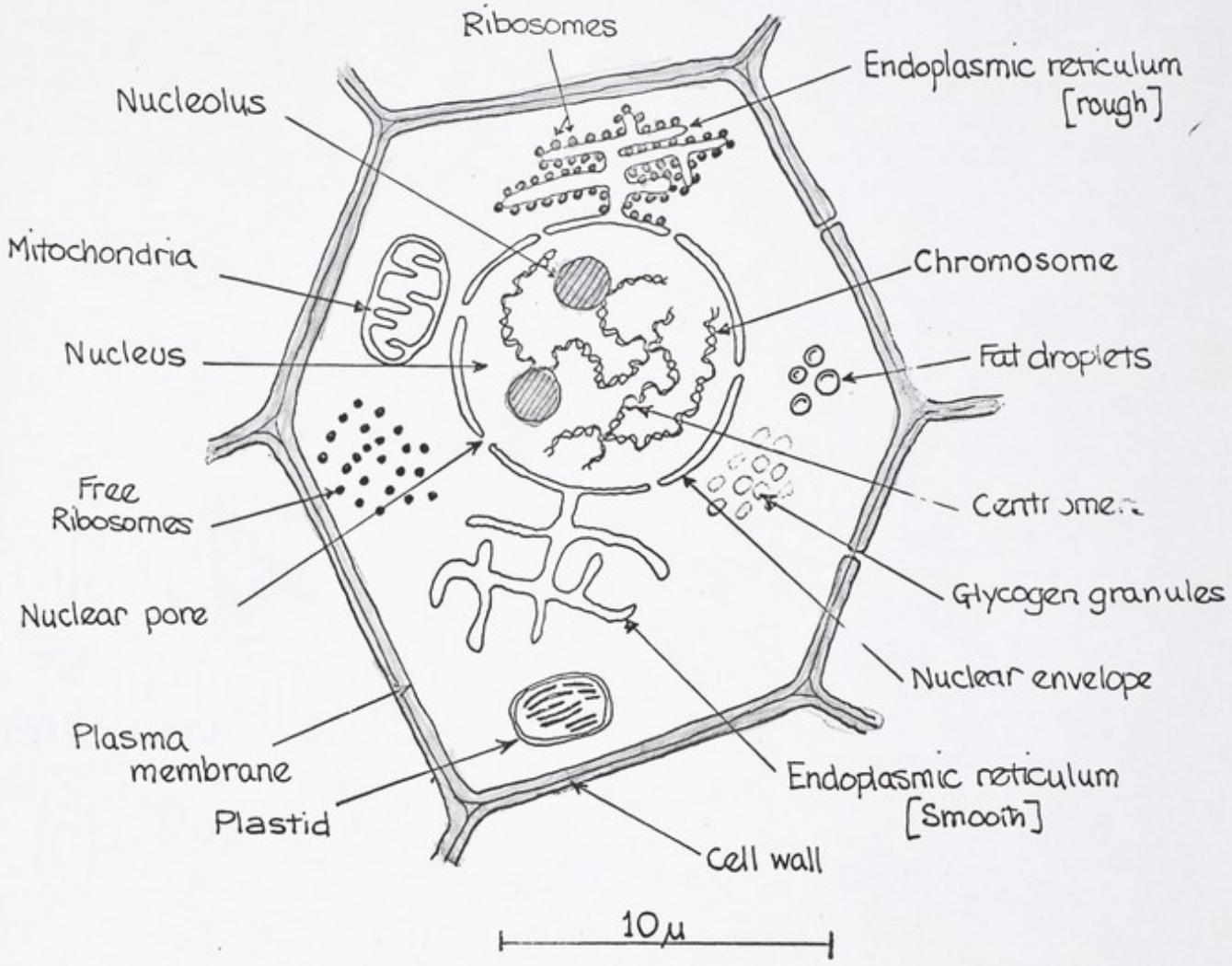


Figure 1 A Schematic view of the plant cell shown in Figure 2. The various components are not always drawn to scale. The plastid shown in the bottom of the cell will eventually transform into a chloroplast, the chlorophyll containing site of photosynthesis.

roughly equally to give two daughter cells. At the same time, the nucleus divides so that each daughter cell can receive a nucleus.

Mitosis maintains the parental chromosome number

Each nucleus encloses a fixed number of linear bodies, called chromosomes (Figure I- 3). Before cell division, each of the chromosomes divides to form two chromosomes identical to the parental body. This process, first accurately observed by Flemming in 1879, doubles the number of nuclear chromosomes. During nuclear division, one of each pair of daughter chromosomes moves into each daughter nucleus (Figure I- 4). As a result of these events (now termed "mitosis"), the chromosomal complement of daughter cells is usually identical to that of the parental cells.

During most of a cell's life its chromosomes exist as highly extended linear objects. Prior to cell division, however, they condense into much more compact bodies. The duplication of chromosomes chiefly occurs when they are in the extended state characteristic of interphase, (the various stages of cell division are defined in Figure I- 4). One part of the chromosome, however, always duplicates during the contracted metaphase state: this is the centromere, a body which controls the movement of the chromosome during cell divisions. The centromere always has a fixed location on a given chromosome. Its specific location, however, varies with the specific chromosome; in some it is near one end, and in others it occupies an intermediate region.

When a chromosome is completely duplicated except for the centromere, it is said to consist of two chromatids. A chromatid is transformed into a chromosome as soon as its centromere has divided, and is no longer shared with another chromatid. As soon as one centromere becomes two, the two daughter chromosomes begin to move apart from each other.

The regular lining up of chromosomes during the metaphase stage is accompanied

Figure I-3 The haploid complement of chromosomes from the leopard frog (rana pipens) magnified 2125 times. This photograph was taken with a light microscope by T.E. Powall of the Biological Laboritories, Harvard University. It shows the chromosomes when they have duplicated to form two chromatids held together by a single centromere.

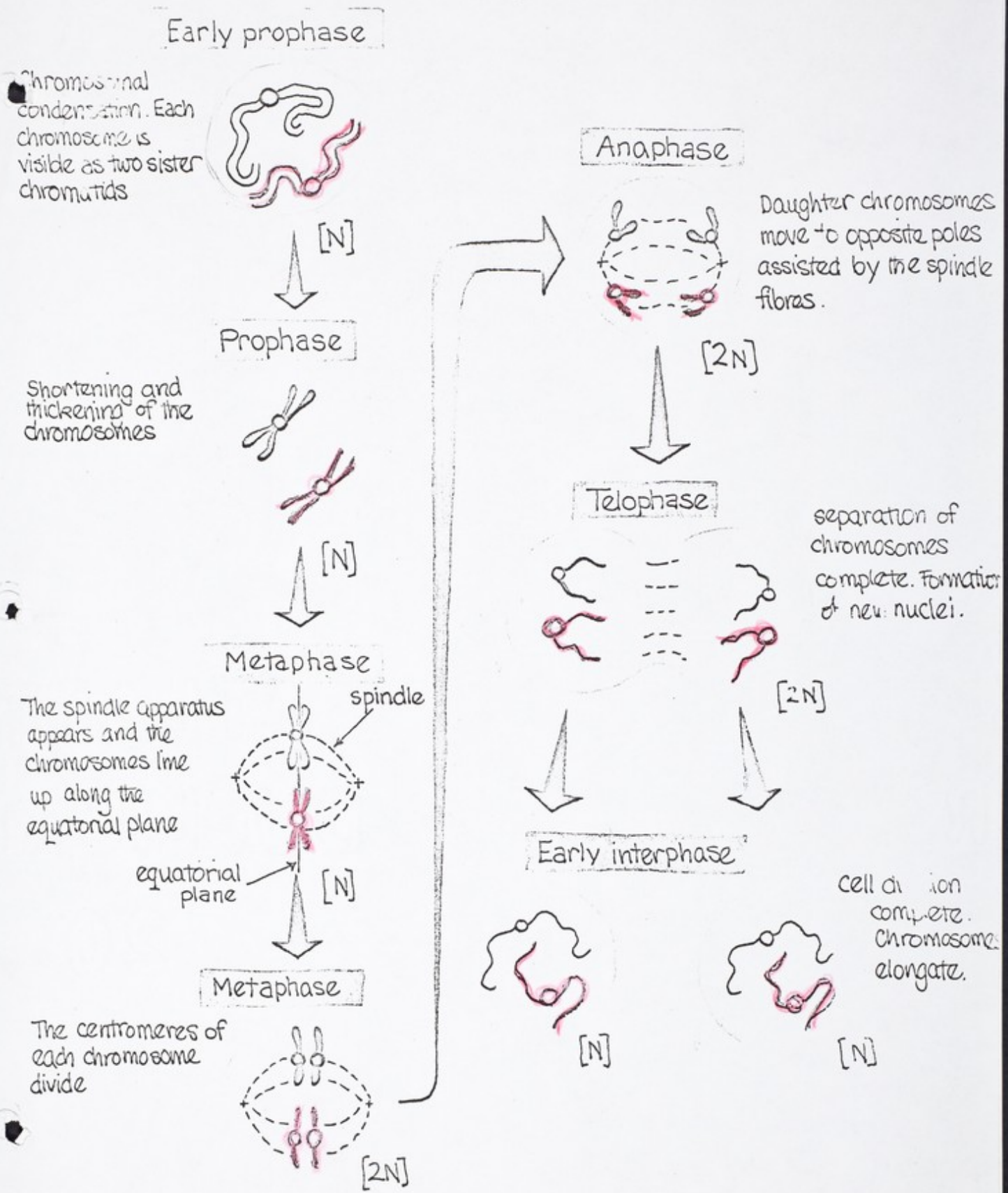


Figure I-4 Diagram of mitosis in a haploid cell containing 2 nonhomologous chromosomes.

by the appearance of the spindle. This is a cellular region, shaped like a spindle, through which the chromosomes of higher organisms move apart during the anaphase stage. Much of the spindle region is filled with long, thin protein molecules, which some people think are similar to the contractile proteins of muscles. If this resemblance is genuine, then perhaps the same mechanism which underlies the contraction of muscles also underlies the movement of chromosomes through the spindle.

There are also present in the nucleus of virtually every plant and animal cell objects called the nucleoli. There is at least one nucleolus per haploid set of chromosomes, and at least in some cells the nucleolus is connected to a specific chromosome. Until very recently the functional role of the nucleolus was completely obscure, though some cytologists (cytology is the study of cells) thought that they might be related to the formation of the spindle. Now, however, there are some strong hints that they are involved in the synthesis of ribosomes, small cellular particles upon which all proteins are synthesized.

Meiosis reduces the parental chromosome number

One important exception was found to the mitotic process. At the conclusion of the two cell divisions which form the sex cells, the sperm and the egg, (meiosis) the number of chromosomes is reduced to one-half of its previous number (Figure I-5). In higher plants and animals each specific type of chromosome is normally present in two copies: the homologous chromosomes (the diploid state). In sex cell formation the resulting sperm and egg each usually encloses only one of each type (the haploid state). Union of sperm and egg during fertilization results in a fertilized egg (zygote) containing one homologous chromosome from the male parent and the other from the female parent. Thus the normal diploid chromosome constitution is restored (FigureI-4).

While in higher plants and animals most cells are diploid, in lower plants and bacteria the haploid state is the most frequent condition, the diploid number existing only briefly following sex cell fusion. Usually almost immediately after fertilization

meiosis occurs to produce haploid cells. (Figure I-4).

The cell theory thus tells us that all cells come from pre-existing cells. All the cells in adult plants and animals are derived from the division and growth of a fertilized egg, itself formed by the union of two other cells, the sperm and the egg. All growing cells contain chromosomes, usually two of each type, and here again, new chromosomes always arise through division of previously existing bodies.

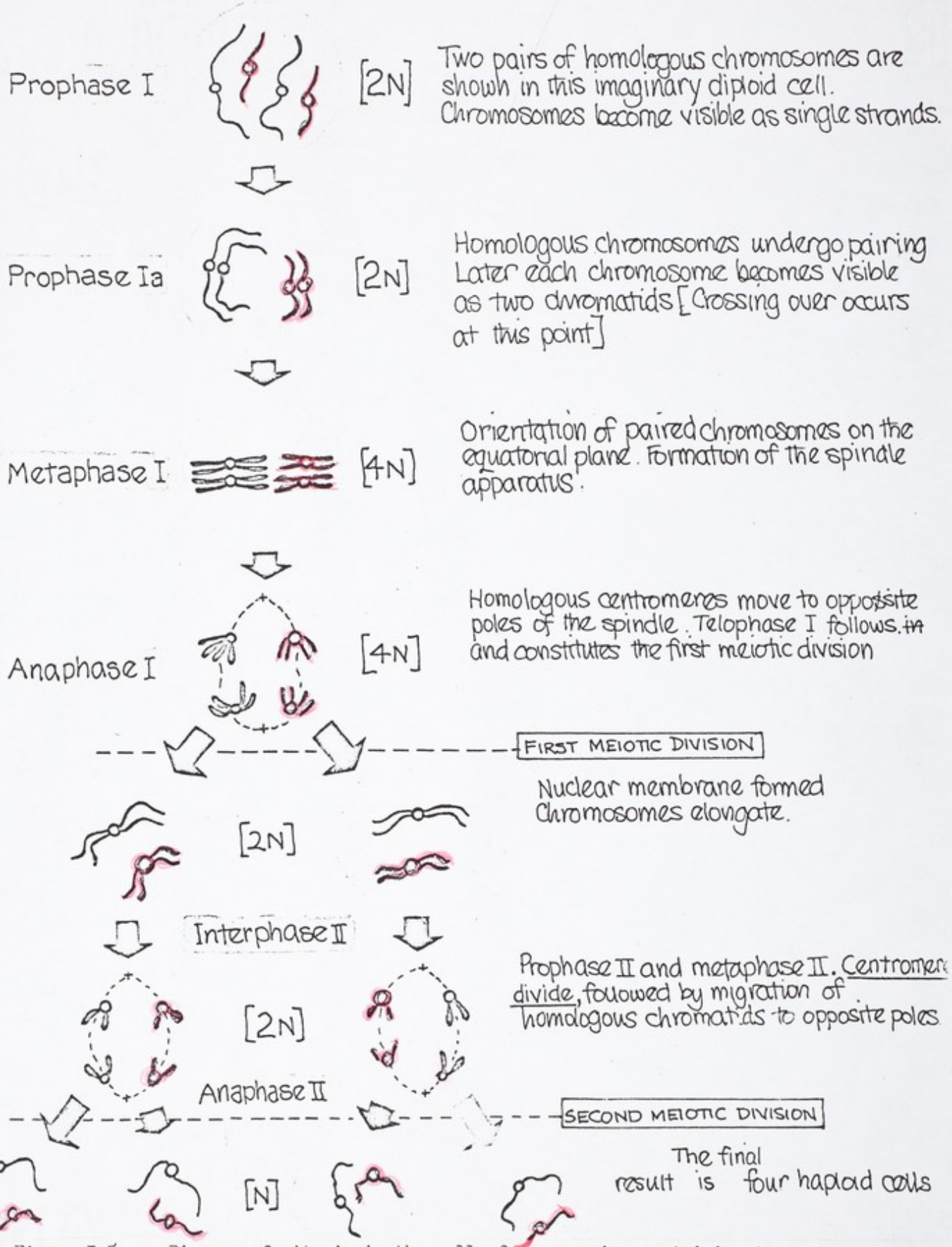


Figure I-5 Diagram of mitosis in the cell of an organism containing two pairs of homologous chromosomes.

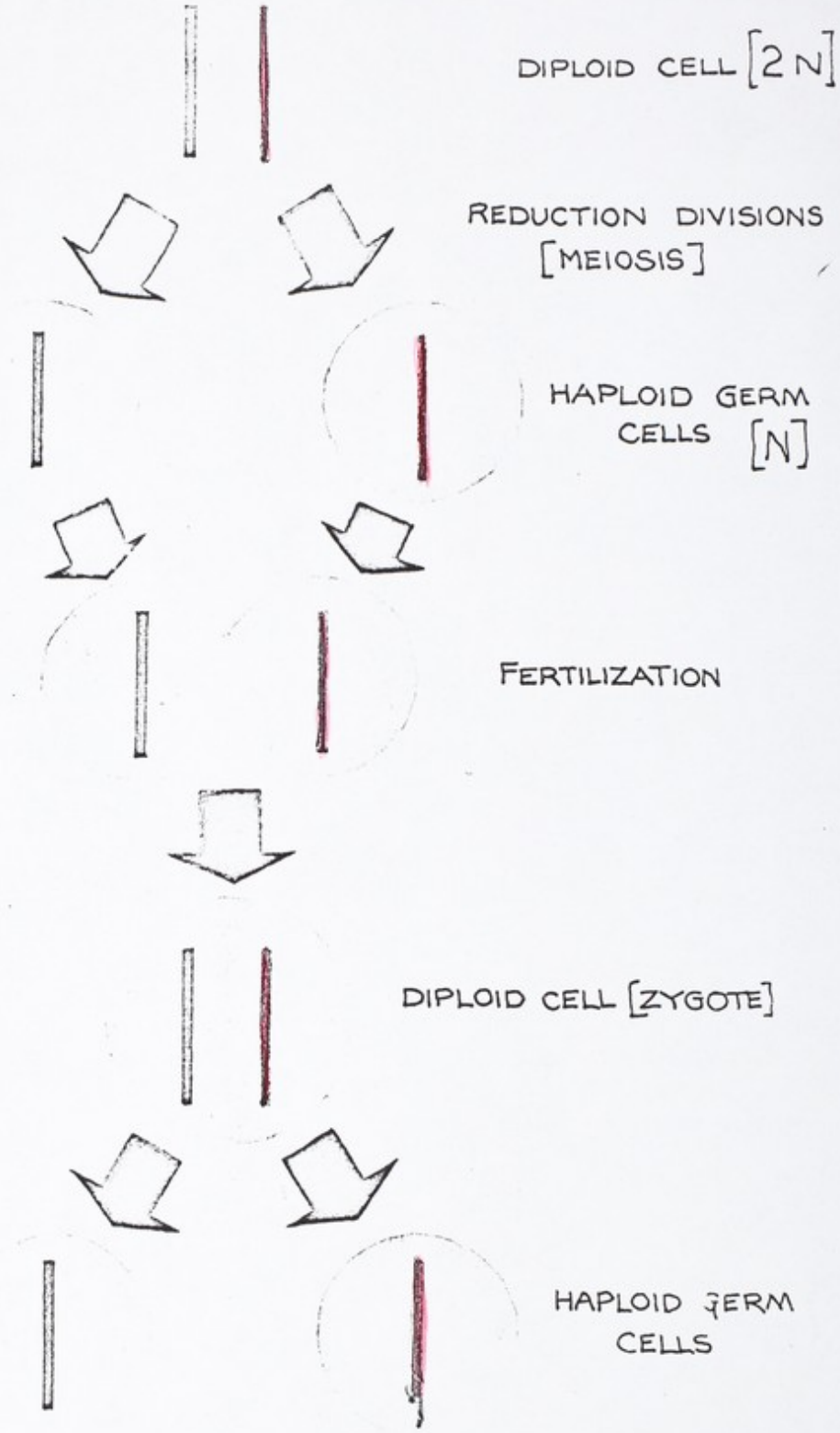


Figure I-6 Diagram of the alternation of haploid and diploid states which comprise the sexual cycle. The chromosome set derived from one parent is shown black, that from the other p..rt red.

The Cell Theory is universally applicable

Although the cell theory developed from observations about higher organisms, it holds with equal force for the more simple forms of life, such as protozoa and bacteria. Each bacterium or protozoan is a single cell, whose division usually produces a new cell identical to its parent, from which it soon separates. In the higher organisms, on the other hand, the daughter cells not only often remain together, but also often differentiate into radically different cell types, like nerve or muscle cells, while maintaining the chromosome complement of the zygote. Here, new organisms arise from the highly differentiated sperm and egg, whose union initiates a new cycle of division and differentiation.

Thus, although a complicated organism like man contains a very large number (up to 5×10^{12}) of cells, they all initially arise from a single cell. The fertilized egg contains all the information necessary for the growth and development of an adult plant or animal. Again the living state per se does not demand the complicated interactions which occur in complex organisms: but its essential properties can be found in single growing cells.

The Laws of Mendel

The most striking attribute of a living cell is its ability to transmit hereditary properties from one cell generation to another. The existence of heredity must have been noticed by early man as he witnessed the passing of characteristics, like eye or hair color, from parents to their offspring. Its physical basis, however, was not understood until the first years of the twentieth century, when in a remarkable period of creative activity the chromosomal theory of heredity was established.

Hereditary transmission through the sperm and egg became known by 1860, and already in 1868 Haeckel, noting that sperm consisted largely of nuclear material, postulated that the nucleus was responsible for heredity. Almost twenty years passed before the chromosomes were singled out as the active factors, because

the details of mitosis, meiosis and fertilization had to be worked out first.

Then it could be seen that, unlike other cell constituents, the chromosomes were equally divided between daughter cells. Moreover, the complicated chromosomal changes observed during meiosis which reduce the sperm and egg chromosome number to the haploid number became understandable if the chromosome number had to be kept constant. These facts, however, merely suggested that chromosomes carry heredity.

Proof came at the turn of the century with the discovery of the basic rules of heredity. These rules, named after their original discoverer Mendel, had in fact been first proposed in 1865, but the climate of scientific opinion had not then been ripe for their acceptance. They were completely ignored until 1900, despite some early efforts on Mendel's part to interest the prominent biologists of his time. Then de Vries, Correns and Tschermak, all working independently, realized the great importance of Mendel's forgotten work. All three were plant breeders doing experiments related to Mendel's, and each reached similar conclusions before they knew of Mendel's work.

Principle of independent segregation

Mendel's experiments traced the results of breeding experiments (genetic crosses) between strains of peas differing in well-defined characteristics, like seed shape (round or wrinkled), seed color (yellow or green), pod shape (inflated or wrinkled), and stem length (long or short). His concentration on well-defined differences was very important; previously many breeders had tried to follow the inheritance of more gross qualities, like body weight, and were unable to discover any simple rules about their transmission from parents to offspring. After ascertaining that each type of parental strain bred true (that is, produced progeny with particular qualities identical to those of the parents), Mendel made a number of crosses between parents (P) differing in

in single characteristics, (such as seed shape, or seed color). This yielded the striking conclusion that all the progeny (F_1 = first filial generation) had the appearance of one of the parents. For example, in a cross between peas having yellow seeds and peas having green seeds, all the progeny had yellow seeds. The trait which appears in the progeny is called dominant, while that which does not appear in F_1 is called recessive.

The meaning of these results became clear with the genetic crosses Mendel made between F_1 offspring. These crosses gave the most important result that the recessive trait reappeared in approximately 25% of the progeny, while the dominant trait appeared in 75% of them. For each of the seven traits he followed, the ratio in F_2 of dominant to recessive traits was always approximately 3:1. When these experiments were carried to a third (F_3) progeny generation, all the F_2 peas with recessive traits bred true (produced progeny with the recessive traits), while those with dominant traits fell into two groups: one-third bred true (produced only progeny with the dominant trait), while the remaining two-thirds again produced mixed progeny in a 3:1 ratio of dominant to recessive.

Mendel correctly interpreted his results as follows (Figure I-7). The various traits are controlled by pairs of factors (which we now call genes), one factor derived from the male parent, the other from the female. For example pure-breeding strains of round peas contain two genes for roundness (RR), while pure-breeding wrinkled strains have two genes for wrinkledness (rr). The round strain gametes each have one gene for roundness; the wrinkled strain gametes each have one gene for wrinkledness (r). In a cross between RR and rr, fertilization thus produces an F_1 plant with both genes, (Rr). The plant looks round because R is dominant over r. We refer to the appearance (physical structure) of an individual as its phenotype, and to its genetic composition as its genotype. Individuals with identical phenotypes may possess different genotypes; thus, to

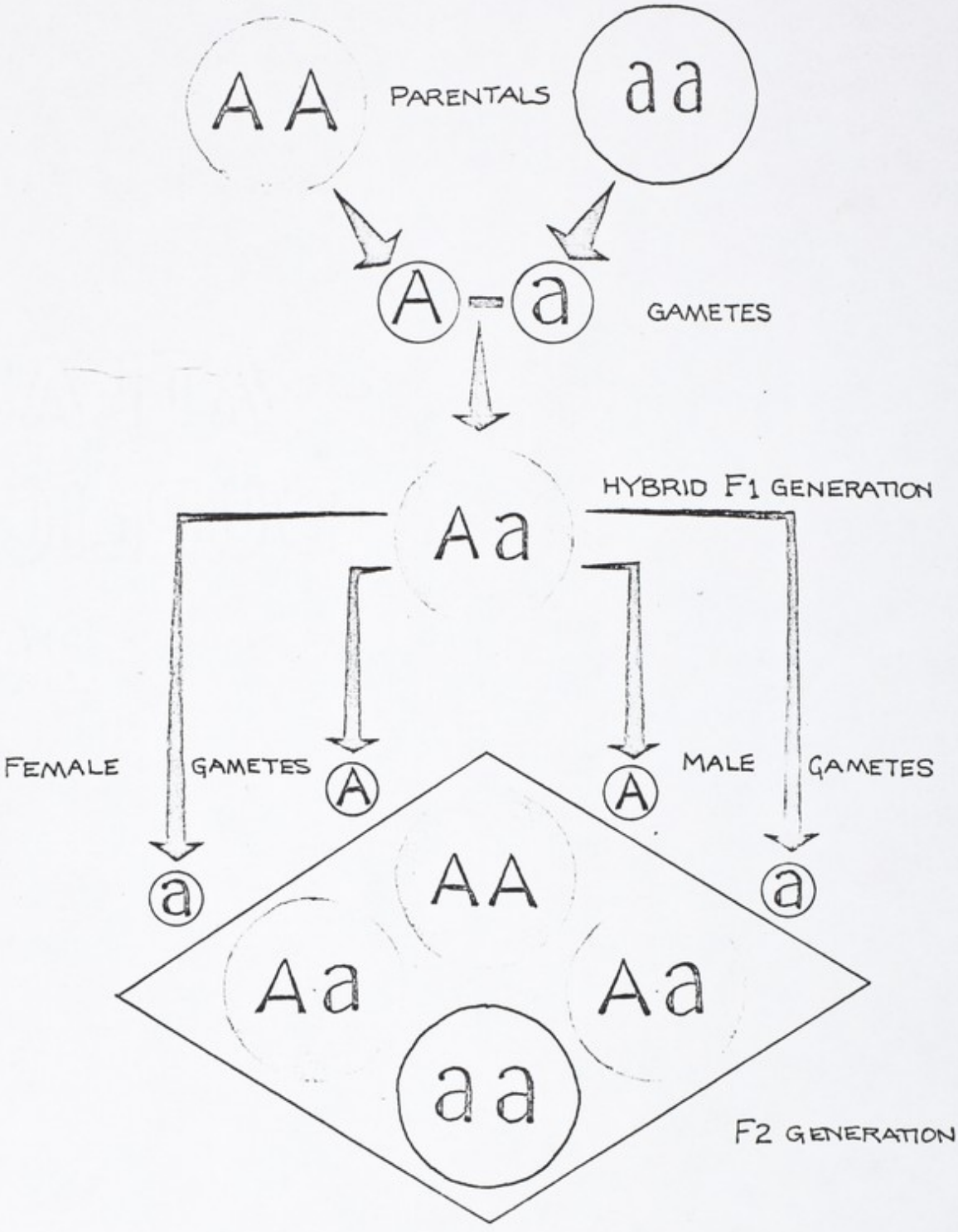


Figure I-6 Representation of how Mendel's First Law (Independent Segregation) explains the three-to-one ratio of dominant to recessive phenotypes among the F_2 progeny. (A)represents the dominant allele and (a) the recessive allele. The shaded circles represent the dominant phenotype, the unshaded circles the recessive phenotype.

determine the genotype of an organism it is frequently necessary to perform several generations of genetic crosses.

It is very important to notice that a given gamete contains only one of the two genes present in the organism it comes from (for example, either the R or the r, but never both), and that the two types of gamete are produced in equal numbers. Thus there is a 50:50 chance that a given gamete from an F_1 pea will contain a particular gene (R or r). This choice is purely random. We do not expect to find exact 3:1 ratios when we examine a limited number of F_2 progeny. Sometimes the ratio will be slightly higher, and other times slightly lower. But as we look at increasingly larger samples, we expect that the ratio of peas with the dominant trait to peas with the recessive trait will more and more closely approach the 3:1 ratio.

The reappearance of the recessive character in the F_2 generation indicates that recessive genes are neither modified nor lost in the hybrid (Rr) generation, but that the dominant and recessive genes are independently transmitted, and so able to segregate independently during the formation of sex cells. This principle of independent segregation is frequently referred to as Mendel's First Law.

Some genes are neither dominant nor recessive

In the crosses reported by Mendel, one of each gene pair was clearly dominant, and the other recessive. This behavior, however, is not universal. Sometimes the heterozygous phenotype is intermediate between the two homozygous phenotypes. For example, the cross between a pure breeding red snap dragon (Antirrhinon) and a pure breeding white variety gives F_1 progeny of the intermediate pink color. If these F_1 progeny are crossed among themselves, the resulting F_2 progeny contain red, pink, and white flowers in the proportion of 1:2:1 (Figure I-8). Thus it is possible here, to distinguish heterozygotes from homozygotes by their phenotype. We furthermore see that Mendel's laws do not depend for their

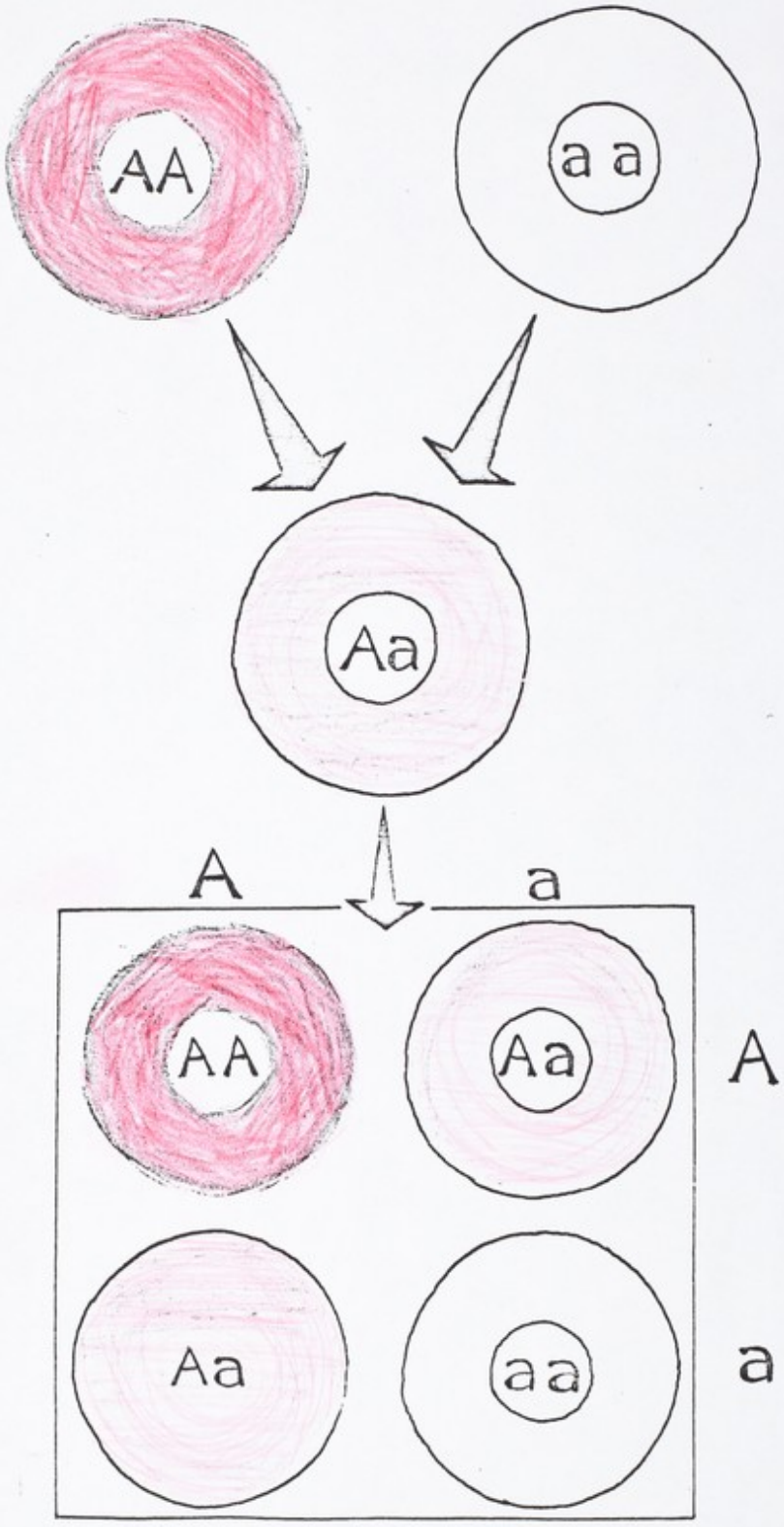


Figure ~~IV-3~~ I-8 The inheritance of flower color in the snap dragon. One parent is homozygous for red flowers (AA) and the other homozygous for white flowers (aa). No dominance is present, and the heterozygous flowers are pink. The 1:2:1 ratio of red:pink:white flowers is shown by appropriate coloring.

applicability on whether or not one allele of a gene pair is dominant over the other.

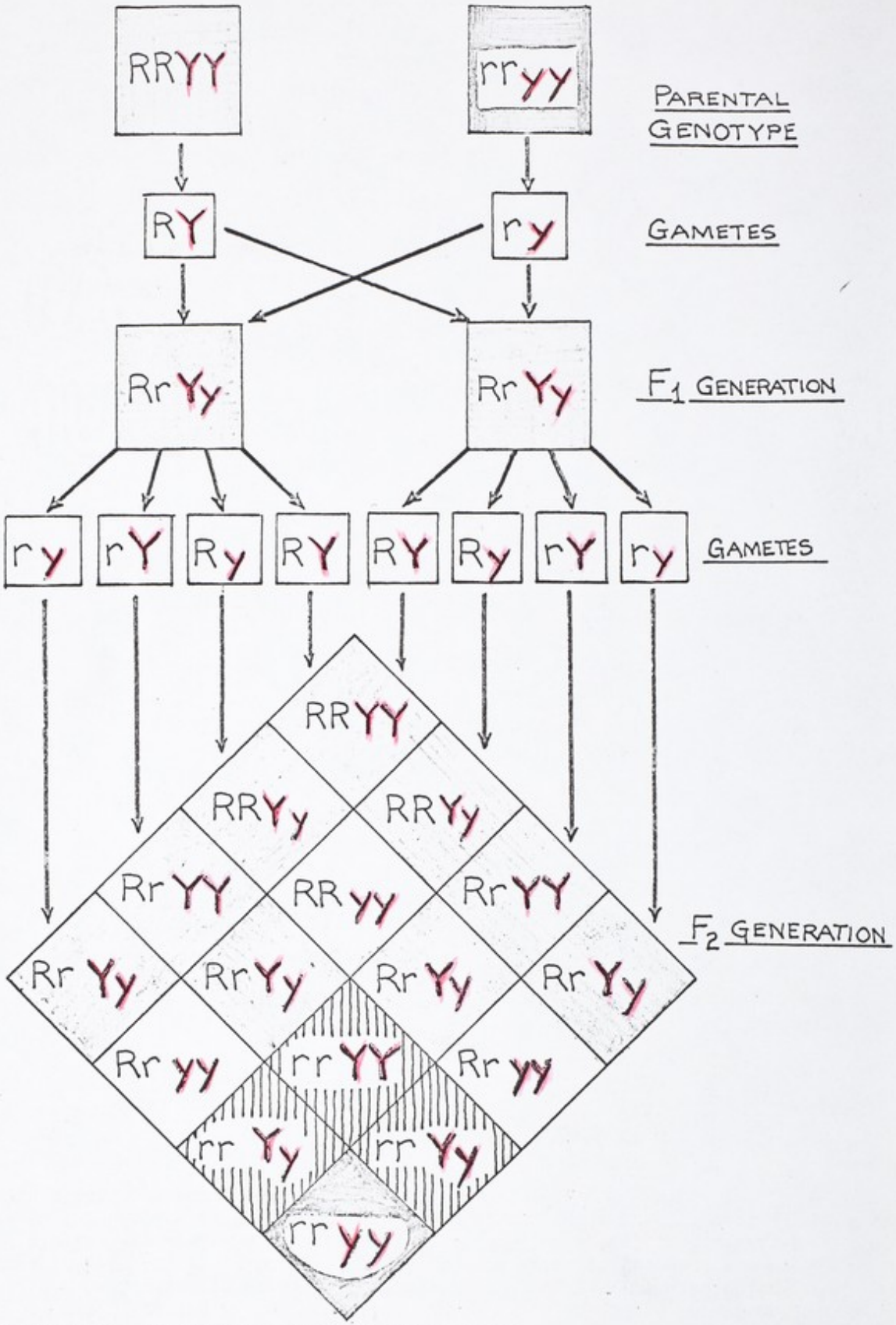
The principle of independent assortment

Mendel then extended his breeding experiments to peas differing by more than one character. As before, he started with two strains of peas, each of which bred pure when mated with itself. One of the strains had round yellow seeds, the other, wrinkled green seeds. Since round and yellow are dominant over wrinkled and green, all the F_1 generation produced round, yellow seeds. The F_1 generation was then crossed among itself to produce a number of F_2 progeny which were examined for seed appearance (phenotype). In addition to the two original phenotypes (round, yellow; wrinkled green), there had emerged two new types (recombinants): wrinkled yellow and round green.

Again Mendel found he could interpret the results by the postulate of genes, if he assumed that during sex cell formation, each gene pair is independently transmitted to the sex cell (gamete). This interpretation is shown in Figure I-9. Any one gamete contains only one type of inherited factor from each gene pair. Thus the gametes produced by an F_1 (RrYy) will have the composition RY, Ry, rY, or ry, but never Rr, Yy, YY, or RR. Furthermore, in this example, all the four possible gametes are produced with equal frequency. There is no tendency of the genes arising from one parent to stay together. As a result, the F_2 progeny phenotypes appear in the ratio of: 9 round yellow, 3 round green, 3 wrinkled yellow, and 1 wrinkled green. This phenomenon of independent assortment is frequently called Mendel's Second Law.

The chromosomal theory of heredity

A principal reason for the original failure to appreciate Mendel's discovery was the absence of firm facts about the behavior of chromosomes during meiosis and



I-9

Key to Phenotypes: grey squares = round yellow seeds
white squares = round green seeds
striped squares = wrinkled yellow seeds
black squares = wrinkled green seeds

mitosis. This knowledge was available, however, when Mendel's laws were reannounced in 1900, and seized upon in 1903 by the American, Sutton. In his classic paper, The Chromosomes in Heredity, he emphasized the importance of the fact that the diploid chromosome group consists of two morphologically similar sets, and that during meiosis every gamete receives only one chromosome of each homologous pair. He then used this fact to explain Mendel's results by the assumption that genes are parts of the chromosome. He postulated that the yellow seed and green seed genes are carried on a certain pair of chromosomes, and that the round seed and wrinkled seed genes are carried on a different pair. This hypothesis immediately explains the experimentally observed 9:3:3:1 segregation ratios (Figure I-). Though Sutton's paper did not prove the chromosomal theory of heredity, it was immensely important, for it brought together for the first time the independent discipline of genetics (the study of breeding experiments) and cytology (the study of cell structure).

Chromosomal determination of sex

There exists one very important exception to the rule that all chromosomes of diploid organisms are present in two copies. It was observed as early as 1890 that one chromosome -- then called an accessory chromosome and now the x chromosome -- does not always possess a morphologically identical mate. The biological significance of this observation was clarified by the American cytologist Wilson and his student Stevens in 1905. They showed that while the female contains a pair of x chromosomes, in the male the x chromosome is present only once. In addition, in some species, the male cells contain a unique chromosome, not found in females, called the y chromosome. They pointed out how this situation provides a simple method of sex determination: while every egg will contain one x chromosome, only half the sperms will carry one. Fertilization of an ovum by an x-bearing sperm leads to an xx zygote, which becomes a female, while ferti-

lization by a sperm cell lacking an x chromosome gives rise to male offspring (Figure I-1D). These observations provided the first clear linking of a definite chromosome to a hereditary property. In addition they elegantly explained how male and female zygotes are created in equal numbers.

The importance of the tiny Drosophila

Initially all breeding experiments used genetic differences already existing in nature. For example, Mendel used seeds obtained from seed dealers who must have obtained them from farmers. The existence of alternative forms of the same gene (alleles) raises the question of how they arose. One obvious hypothesis states that genes can change (mutate) to give rise to new genes (mutant genes). This hypothesis was first seriously tested, beginning in 1908, by the great American biologist Morgen, and his young collaborators, the geneticists Bridges, Muller, and Sturtevent. They worked with the tiny fly Drosophila. This fly, which normally lives on fruit, was found to be easily maintained under laboratory conditions, where a new generation can be produced every 14 days. Thus by using Drosophila instead of more slowly multiplying organisms like peas, it was possible to work at least 25 times faster, and also much more economically. The first mutant found was a male with white eyes instead of the normal red eyes. It spontaneously appeared in a culture bottle of red-eyed flies. Because virtually all Drosophila found in nature have red eyes, the gene leading to red eyes was referred to as the "wild type" gene, while the gene leading to white eyes was called a mutant gene (allele). (Figures I-11a + I-11b)

The white eye mutant gene was immediately used in breeding experiments, with the striking result that the behavior of the allele completely paralleled the distribution of an x chromosome (i.e. was sex linked). This immediately suggested that this gene might be located on the x chromosome, along with those genes controlling sex. This hypothesis was quickly confirmed by additional

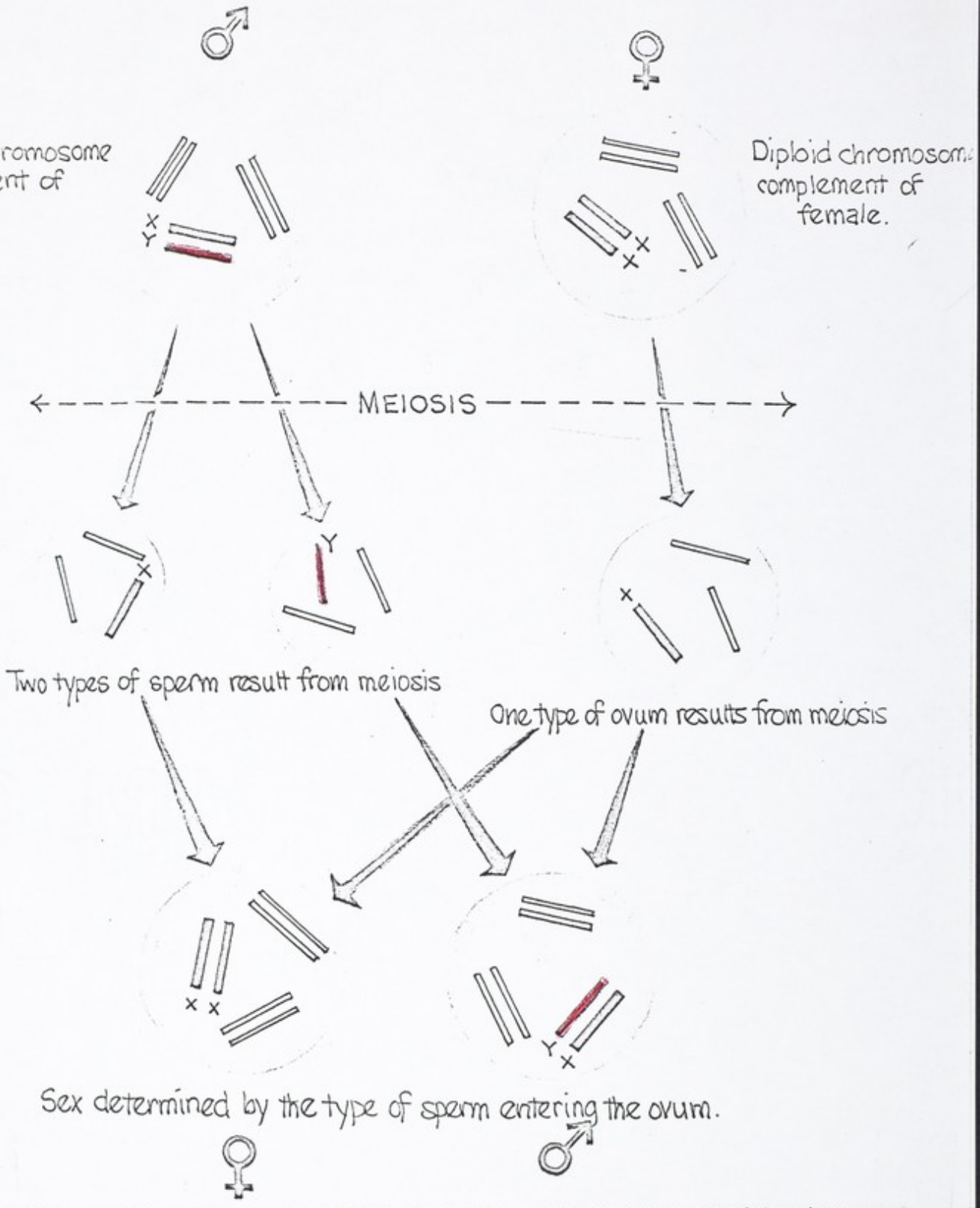


Figure I-10 Schematic representation of how the sex chromosomes can determine sex. Here is shown a case in which males contain one x and one y chromosome, and females, two x chromosomes. This is the situation in both humans and Drosophila. In other species there is no y chromosome, and so diploid male cells contain one less chromosome than diploid female cells.

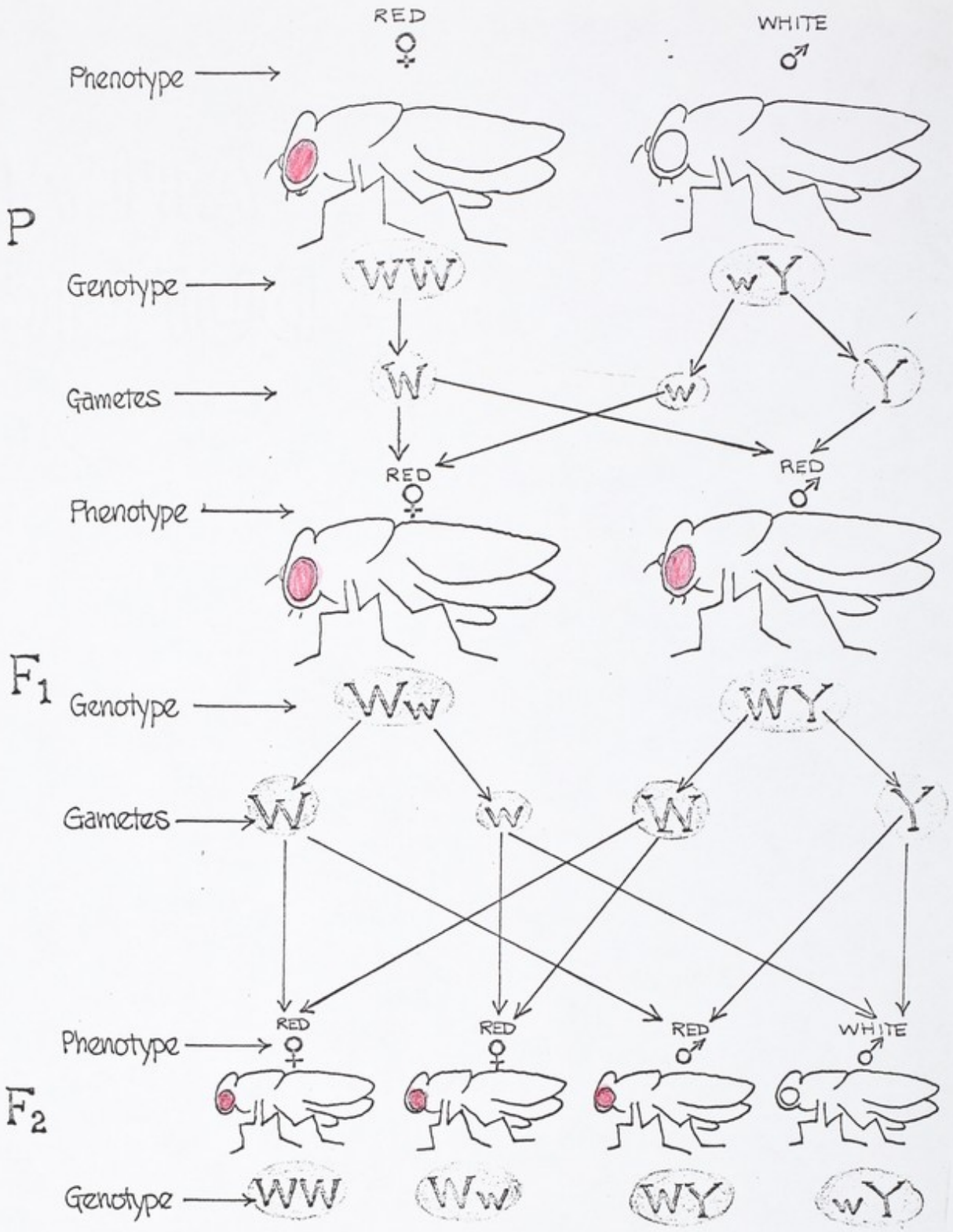
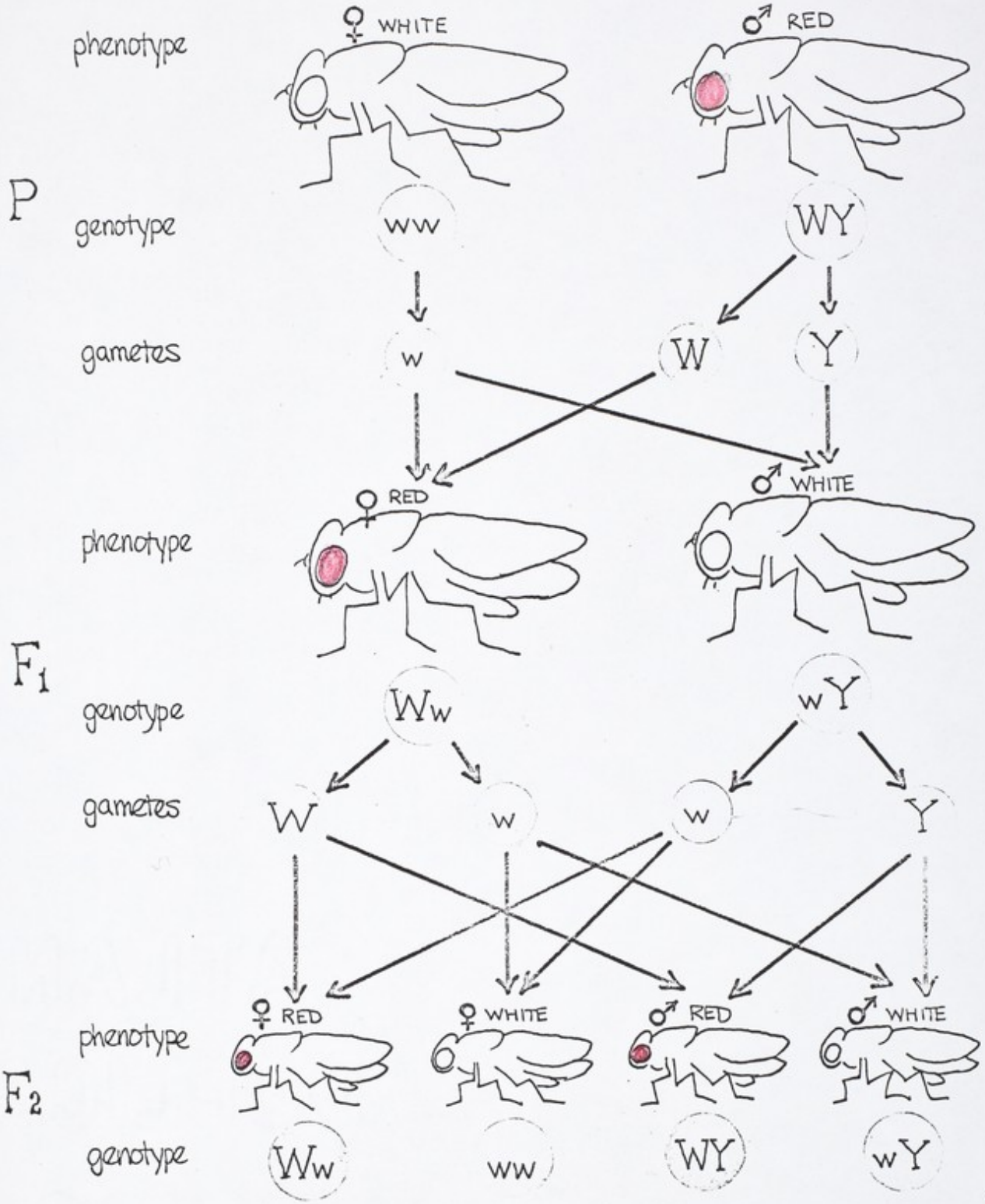


Figure I-11 The inheritance of a sex-linked gene in Drosophila. Genes located on sex chromosomes can express themselves differentially in male and female progeny. This is because, if there is one x chromosome present, recessive genes present in this chromosome are always expressed. Here are shown two crosses, both involving a recessive gene (w, for white eye) located on the x chromosome. In (a) the male parent is a white-eyed (wY) fly, and the female, homozygous for red eye (WW). In (b) the male has red eyes (WY) and the female white eyes (ww). The letter Y stands, here, not for an allele, but for the y chromosome, present in male Drosophila corresponding to the (w) or (W) gene on the x chromosome.


phenotype
♀ WHITE
♂ RED
P
genotype
ww
WY
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w
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Y
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Ww
wY
gametes
W
w
w
Y
♀ RED
♀ WHITE
♂ RED
♂ WHITE
phenotype
F2
genotype
Ww
ww
WY
wY

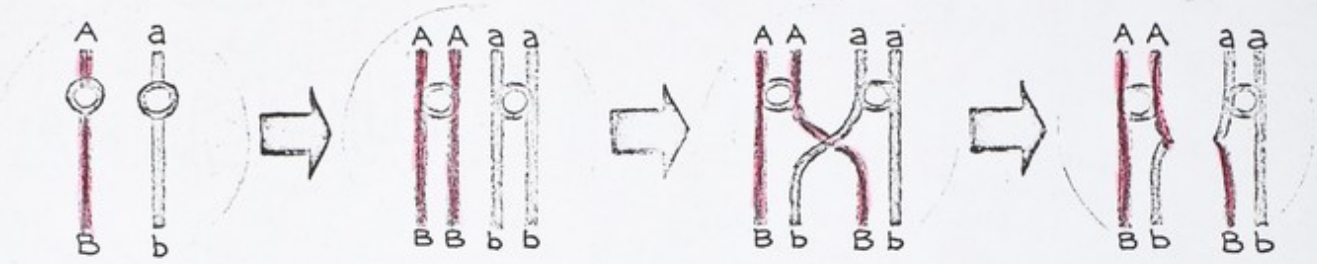
Figure I- [illegible]

genetic crosses using newly isolated mutant genes. Many of these additional mutant genes also were sex linked.

Gene linkage and crossing over

Mendel's principle of independent assortment is based on the fact that genes located on different chromosomes behave independently during meiosis. Often, however, two genes do not assort independently, because they are located on the same chromosome (linked genes). Numerous examples of non-random assortment were found as soon as a large number of mutant genes became available for breeding analysis. In every well studied case, the number of linked groups was identical with the haploid chromosome number. For example, there are four groups of linked genes in Drosophila, and four morphologically distinct chromosomes in a haploid cell.

Almost as soon as linkage was discovered, it was found often not to be complete. For example, one pair of genes assorted with each other 90% of the time, while another two assorted together 95% of the time. This meant that a mechanism existed for exchanging genes on homologous chromosomes. This mechanism is called crossing-over. Its cytological basis was first described by the Danish cytologist Janssens. At the start of meiosis the homologous chromosomes pair (synapse), with their long axes parallel. At this stage, each chromosome has duplicated to form two chromatids. Thus synapsis brings together four chromatids (a tetrad), which coil about each other. Janssens postulated that, possibly because of tension resulting from this coiling, two of the chromatids might sometimes break at a corresponding place on each. This could create four broken ends, which might rejoin crosswise, so that a section of each of the two chromatids is joined to a section of the other (Figure I-12). Thus recombinant chromatids might be produced that contain a segment derived from each of the original



Synapsis of homologous chromosomes

Duplication of chromosomes to form tetrads

Two chromatids bend across one another

The chromatids break at the point of contact and fuse with portions of the other chromatid.

Figure I-12 Janssens' theory of crossing over

homologous chromosomes.

Morgen and his students were quick to exploit the implication of Janssens' still unproven theory: that genes located close to each other on a chromosome would assort with each other much more regularly (close linkage) than genes located far apart on a chromosome. This immediately suggested a way to locate (map) the relative positions of genes on the various chromosomes (see Chapter IV for the details). By 1915 more than 85 mutant genes in Drosophila had been assigned locations, each a distinct spot on one of the four linkage groups or chromosomes (Table I-). The definitive volume which they then published, The Mechanism of Mendelian Heredity, showed the general validity of the chromosomal basis of heredity, a concept which ranks with the theories of evolution and the cell as one of the main achievements of the biologist's attempt to understand the nature of the living world.

Many genes control the red eye, (or any other complex character)

Mere inspection of the list of mutant genes (Table I-) reveals a very important fact: many different genes act to influence a single character. For example, 13 of the genes discovered by 1915 affect eye color. When a fly is homozygous for a mutant form of any of these genes, the eye color is not red, but a different color, distinct for the mutant gene, (e.g. carnation, vermillion). Thus there is not a one-to-one correspondence between genes and complex characters like eye color or wing shape. Instead, the development of each of these characters is controlled by a series of events, each of which is controlled by a gene. We might make a useful analogy with the functioning of a complex machine like the automobile: there are clearly a number of separate parts, like the motor, the brakes, the radiator, and the fuel tank, all of which are essential for its proper operation. While a fault in any one part may cause the car to stop

Table I-1. The eighty-five mutant genes reported in Drosophila melanogaster in 1915. The mutations fall into four linkage groups. Since four chromosomes were cytologically observed, this indicated that the genes are situated on the chromosomes. Notice that mutations in various genes can act to alter a single character, such as color, in different ways.

Group I

Name	Region affected
abnormal	abdomen
bar	eye
bifid	venation
bow	wing
cherry	eye color
chrome	body color
cleft	venation
club	wing
depressed	wing
dotted	thorax
eosin	eye color
facet	ommatidia
forked	spines
furrowed	eye
fused	venation
green	body color
jaunty	wing
lemon	body color
lethals,13	die
minature	wing
notch	venation
reduplicated	eye color
ruby	legs
rudimentary	wings
sable	body color
shifted	venation
short	wing
skee	wing
spoon	wing
spot	body color
tan	antenna
truncate	wing
vermilion	eye color
white	eye color
yellow	body color

Group IV

bent	wing
eyeless	eye

Group II

Name	Region affected
antlered	wing
apterous	wing
arc	wing
balloon	venation
black	body color
blistered	wing
comma	thorax mark
confluent	venation
cream II	eye color
curved	wing
dachs	legs
extra vein	venation
fringed	wing
jaunty	wing
limited	abdominal band
little crossover	II chromosome
morula	ommatidia
olive	body color
plexus	venation
purple	eye color
speck	thorax mark
strap	wing
streak	pattern
trefoil	pattern
truncate	wing
vestigial	wing

Group III

band	pattern
beaded	wing
cream III	eye color
deformed	eye
dwarf	size of body
ebony	body color
giant	size of body
kidney	eye
low crossingover	III chromosome
maroon	eye color
peach	eye color
pink	eye color
rough	eye
safranin	eye color
sepia	eye color
sooty	body color
spineless	spines
spread	wing
trident	pattern
truncate intensf.	wing
whitehead	pattern
white ocelli	simple eye

functioning properly, there is clearly no reason to believe that the presence of that component alone is sufficient for proper functioning.

The origin of genetic variability through mutations

It now became possible to understand the hereditary variation which is found throughout the biological world, and which forms the basis of the Theory of Evolution. Genes are normally copied exactly during chromosome duplication. Rarely, however, changes (mutations) occur in genes to give rise to altered forms. This process is necessarily very rare; otherwise many genes would be changed during every cell cycle, and offspring would not ordinarily resemble their parents. There is instead, a strong advantage in there being a small but finite mutation rate; it provides the constant source of new variability necessary to allow plants and animals to adapt to a constantly changing physical and biological environment.

Surprisingly, however, the results of the Mendelian geneticists were not avidly seized upon by the classical biologists, then the authorities on the evolutionary relations between the various forms of life. Doubts were raised about whether genetic changes of the type studied by Morgen and his students were sufficient to permit the evolution of radically new structures, like wings or eyes. Instead, they believed that there must also exist more powerful "macro-mutations", and it was these that allowed great evolutionary advances.

Gradually, however, these doubts vanished, largely due to the efforts of the mathematically more powerful geneticists, the American Sewell Wright, and the Englishmen Fisher and Haldane. They showed how, considering the very long life of the earth, the relatively low mutation rates formed for Drosophila's genes, together with only mild selective advantages, would be sufficient to allow the gradual accumulation of new favorable attributes. By the 1930's,

biologists themselves began to reevaluate their knowledge on the origin of species, and to understand the work of the mathematical geneticists. Among these new Darwinians were the biologist Julian Huxley, (a grandson of Darwin's original publicist T. H. Huxley,), the Russian-born American geneticist Dobzhansky, the American paleontologist (student of fossils) Simpson, and the German-born American ornithologist Mayr. In the 1940's all four wrote major works, showing, each from his special viewpoint, how Mendelianism and Darwinism were indeed compatible.

Early speculations about what genes are and how they act

Virtually immediately after the rediscovery of Mendel's laws, geneticists began to speculate both about the chemical structure of the gene and how it acts. No real progress could be made, however, since the chemical identity of the genetic material remained unknown. Even the realization that both nucleic acids and proteins are present in chromosomes did not really help, since the structure of neither was at all understood. The most fruitful speculations focused attention on the fact that genes must be, in some sense, self duplicating: their structure must be exactly copied every time one chromosome becomes two. The possibility was considered that the gene and the primary gene product (perhaps a protein) were one and the same; if this were true (which it is not), then one primary replication process would be sufficient for both the gene and its product. Ideas of this sort immediately raised the profound chemical question of how a complicated molecule could be precisely copied to yield exact replicas.

Some physicists also became intrigued with the gene, and when quantum mechanics burst on the world in the late 1920's, the possibility arose that perhaps to understand the gene it would be necessary to master the subtleties of the most advanced theoretical physics. Such thoughts, however, never really

took root, since it was obvious that even the best of physicists or theoretical chemists could not worry about a substance whose structure still awaited elucidation. There was only one fact which they might ponder: Muller's 1927 discovery that x-rays induce mutations could be used to estimate the size of the gene. But even here so many special assumptions had to be made that virtually no one, not even the estimators themselves, took the estimates very seriously.

Preliminary attempts to find a gene-protein relation

The most fruitful endeavors to find a relationship between genes and proteins examined the ways in which gene changes affect which proteins are present in the cell. At first this was very difficult, since no one really knew anything about the proteins which were present in structures like the eye or the wing. It was thus clear that only very few of the many genes then described would be suitable for an understanding of the gene-protein relation. One of the first useful examples came from a study of a hereditary disease affecting amino acid metabolism. There occur in humans spontaneous mutations affecting the ability to metabolize the amino acid phenylalanine. When individuals homozygous for the mutant trait feed on food containing phenylalanine, their inability to break it down results in a toxic level of phenylalanine. The existence of such diseases, an example of the so-called "Inborn Errors of Metabolism", suggested as early as 1909 to the English physician Garrod that the wild type gene is responsible for the presence of a particular enzyme, and that in a homozygous mutant, the enzyme is congenitally absent.

This general hypothesis of a gene-enzyme relationship was extended in the 1930's by work on flower pigments and the pigments of insect eyes. In both cases evidence was obtained that a particular gene affected a particular step in the formation of the pigment. However, the absence of fundamental knowledge about the structures of the relevant proteins did not permit deeper examination of the gene-protein

relationship, and no assurance could be given either that most genes control the synthesis of proteins (by then it was suspected that all enzymes were proteins), or that all proteins are under gene control.

It thus became obvious to the Mendelian geneticists as early as 1935 that future experiments of the sort successful in elucidating the basic features of Mendelian genetics were unlikely to yield productive evidence about how genes act. Instead it would be necessary to find biological objects more suitable for chemical analysis. They were aware that the contemporary state of chemistry was completely inadequate for a fundamental chemical attack of even the most suitable biological systems. Very fortunately, however, the limitations in chemistry did not deter them from learning how to do genetic experiments with chemically simple molds, bacteria, and viruses. As we shall see, the necessary chemical facts became available almost as soon as the geneticists were ready to use them.

Summary

The study of living organisms at the biological level has led to three great generalizations: (1) Darwin's and Wallace's Theory of Evolution by Natural Selection, which tells us that today's complex plants and animals are derived by a continuous evolutionary progression from the first primitive organisms, (2) the Cell Theory, the realization that all organisms are built up of cells, (3) the Chromosomal Theory of Heredity, the understanding that the function of chromosomes is the control of heredity.

All cells contain chromosomes, which are normally duplicated prior to a cell division process (mitosis) which produces two daughter cells, each with a chromosomal complement identical to that of the parental cell. In haploid cells there is usually just one copy of each type of chromosome, while in diploid cells there are usually two copies (pairs of homologous chromosomes). A diploid cell arises by fusion of a male and a female haploid cell (fertilization), while haploid cells are formed from a diploid cell by a distinctive form of cell division (meiosis) which reduces

the chromosome number to one-half of its previous number.

Chromosomes control heredity because they are the cellular locations of genes. The existence of genes was first discovered by Mendel in 1865, but it wasn't until the first half of the twentieth century that their importance was realized. Each gene can exist in a variety of different forms called alleles. Mendel proposed that a gene for each hereditary trait is given by each parent to each of its offspring. The physical basis for this behavior is in the distribution of homologous chromosomes during meiosis: one (randomly chosen) of each pair of homolggous chromosomes is distributed to each haploid cell. When two genes are on the same chromosome they tend to be inherited together (linked genes). Genes affecting different characters sometimes are inherited independently of each other: this is becaase they are located on different chromosomes. Linkage is, in any case, seldom complete, because during meiosis homolggous chromosomes attach to each other, and often break at homologous and rejoin crosswise. (crossing-over). This attaches genes initially found on a paternally-derived chromosome to gene groups originating from the maternal parent.

Different alleles of the same gene arise by inheritable changes (mutations) in the gene itself. Normally genes are very stable and are exactly copied during chromosome duplication; mutation normally occurs very rarely, and usually has harmful consequences. It does, however, play a positive role, since the accumulation of the rare favorable mutations provides the basis for the genetic variability which the Theory of Evolution presupposes.

For many years the structure of the genes and the chemical way in which they control cellular characteristics were completely mysterious. As soon as large numbers of spontaneous mutations had been apprehended, it became obvious that a one gene - one character relationship does not exist, but that all complex characters are under the control of many genes. The most sensible idea, postulated clearly by Garrod as early as 1909, was that genes affect the synthesis of enzymes.

However, in general, the tools of the Mendelian geneticists, organisms like the corn plant, the mouse, and even the fruit fly, Drosophila, were not suitable for chemical investigations of gene-protein relations. For this type of analysis, work with much simpler microorganisms became indispensable.