CHAPTER 11

The Experimental Polyploids

11.1: 1937 — Beginning of a New Era in Polyploidy

Colchicine replaced practically all the techniques used to double the number of chromosomes in plants. The procedure was new and could easily be fitted to many different kinds of plants. Within a short time geneticists became convinced that a very useful tool had been discovered, because colchicine methods were more effective and more suitable for making polyploids, plants with additional sets of chromosomes, than any formerly used.

Immediate and wide universal interest in colchicine developed among botanists, as shown by the rapid rise in popularity that followed closely upon the announcements of chemical induction of chromosomal doubling.[11, 12, 52, 53, 62] A new era in polyploidy investigations began in 1937, the year the colchicine method was discovered.[36, 72]

Soon the advantages of colchicine became clear. One out of 600 cotton plants treated by "heat-shock" became polyploid (1:600), but colchicine procedures applied to a comparable group yielded 50 polyploids from among 100 (1:2) of the cotton plants surviving the chemical treatment.[8] Similarly the superiority of colchicine was discovered by workers at the chromosome laboratory, Svalof, Sweden, where up to the time colchicine was introduced, elaborate heat-shock machinery, with refrigeration controls, had been used to double the number of chromosomes.[46] Swedish botanists soon discovered that such complicated equipment was no longer necessary.[46] A rapid change-over to colchicine took place.[44, 3, 8, 14, 16, 20, 21, 23, 25, 26, 30, 32, 41, 43, 46, 51, 50, 54, 56, 57, 58, 59, 63, 64, 65, 66, 69, 70, 73, 74] The switch to colchicine in Sweden and elsewhere was so fast that it appeared that the colchicine "fad" in research had arrived.[72, 28]

As we mentioned in Chapter 2, colchicine was not the first chemical to be tried and used for doubling of chromosomes. Other chemicals, heat-shock methods,[10] production of callus tissue,[40] and other

techniques yielded polyploid types.[60] The reason these methods were replaced is found in the two specific advantages demonstrated by colchicine: First, colchicine was very effective for making polyploids with many different species; and second, the drug was applied easily to young growing plants with very little damage being done to them.

There are several noteworthy features of colchicine that account for its effectiveness as a polyploidizing agent. Briefly, colchicine is highly soluble in water; colchicine is not toxic to plant cells even in strong dosages; colchicine is effective in concentrations ranging from 1.0 to 0.01 per cent (1:100 to 1:10,000); and finally, it is soluble in lipoids. Furthermore, the effect obtained during a treatment is wholly reversible. Thus the drug is almost "made to order" for changing diploids into polyploids.

After recovery from treatment the new tissue from treated generations (C_0 = generation) and the progeny of succeeding generations (C_1 = first, C_2 = second, etc.) do not show damage of a hereditary nature. The usual changes associated with multiplication of chromosomes, gigantic characters in leaf, flower, fruit, and seed, are transmitted to the next generations; there is no evidence that "deterioration"[47] sets in after colchicine reaches the protoplasm. While the treated plants may perhaps have wrinkled leaves, distorted stems, and various anatomical malformations, such temporary changes disappear in C_1, C_2, and later cycles.

Gene changes or chromosome repatterning have not been proved,[33, 71] although preliminary tests led to these suggestions. This much is certain: Changes comparable to those produced by X-ray have not been found, and if we choose to use the word *mutation,* it must be clearly stated that colchicine does not cause gene mutations. Only in the broad sense of *mutation,* which includes chromosomal doubling, may we use the term in connection with colchicine as a producer of mutations.[24] If the definition is limited to *gene changes* and *chromosome repatterning* (inversions and translocations), colchicine does not cause mutations. Hence it is incorrect to classify colchicine with *mutagens,* such as p-acetamidotropolone, a 7-carbon compound which appears to cause chromosomal breakage.[71]

More knowledge about the meaning and use of chromosome numbers in relation to species relationship formation is desirable. Every experimenter before commencing a project with colchicine should know the drug is not a chemical fertilizer; it is not a phytohormone; it is not a weed killer; it is not a vitamin; it is not a mutagen; and finally, colchicine is not merely one more organic substance on the present long list now at the disposal of many persons interested in plants.[29] The drug has specific and limited uses; therefore, reports giving directions to spray a field with colchicine or to soak the soil as one would with fertilizing agents, are completely erroneous.

In this chapter and the next four chapters the future possibilities,[70] limitations, and accomplishments are given. Miracles were predicted in the numerous writings in praise of colchicine, but there often followed a serious disillusionment for those not informed in polyploidy and cytogenetics.[27] A wave of great enthusiasm for colchicine in some quarters was succeeded by a loss of interest. Totally discounting colchicine, however, is quite wrong.

11.2: Terminology

In the rapidly expanding field of cytogenetics, new terms are constantly being added, while others are modified as more information is acquired. The two terms, *auto-syndesis* and *allo-syndesis,* have been used with exactly opposite meanings by two groups. Now each time the terms are used, an explanation must accompany the usage. When autopolyploidy and allopolyploidy were first pointed out by Kihara and Ono in 1926,[43] the distinctions were based on materials at hand. When many more examples came into consideration, the differences were not as specific as one might desire for a classification. Terms and their meanings often introduce added confusion. The terminology and definitions used here have in large part been adapted from Clausen, Keck, and Heisey.[18] Extensive work on terminology has been done by Stebbins.[66]

Ploidy, in recent usage, means *fold* (from the Greek *ploos*) and a combining form *like* (*oid*). Thus the prefixed word *polyploidy* means *many-fold.* This refers to the number of sets of chromosomes for a particular plant or animal. *Monoploid* refers to those cells or individuals with one set; *diploid,* twofold; *triploid,* threefold; *tetraploid,* fourfold. Then *autoploid* means self-fold; *amphiploid,* both-fold.

Polyploidy describes a serial relation of numbers in multiples starting from some basic number. If the number is 7, then the polyploid series would read 21, 28, 42, for triploid, tetraploid, and hexaploid, respectively.

Autoploidy is an abbreviated form of the term *autopolyploidy* and will be used for those polyploids formed by multiplication of sets of chromosomes within the limits of a species. Admittedly, the range is wide, and complications arise in classification because the autoploid with four homologous sets will differ from the one derived from two subspecies, that is, the doubled intraspecific hybrid.

Amphiploidy embraces the polyploids derived from the additions of two distinct species. A sterile hybrid *AB* upon doubling becomes the amphiploid *AABB.* If the number of species included increases beyond two, a polyploid-amphiploid condition obtains.

Segmental allopolyploid is an amphiploid which shows characteristics of autoploids with respect to pairing of chromosomes, resemblance to parents, and fertility; yet the amphiploid exhibits enough difference between the genomes contributed by the parents to fall within the scope of amphiploids. Segmental types are important for practical and theoretical reasons. Our discussion of the segmental allopolyploid will be included in Chapter 12 (The Amphiploids).

Genome designates the set of chromosomes derived from a species; the term may be used to express a relationship between species. Extensive use has been made of genomes since many interspecific hybrids have been made and doubled with colchicine. Among species of *Gossypium* the genome concept is related to geographical distribution of species. The genomes of *Triticum* refer to generic contributions. The original term was introduced by Winkler in 1920.

Dysploidy refers to a series of polyploids in nature whose basic numbers are not multiples. A dysploidy is superimposed upon an amphiploid series. A good example is found among the Cruciferae, where basic numbers 5, 6, 7, 9, 11 fall at levels of diploid, tetraploid, and hexaploid status.

Aneuploidy is a condition in which chromosomes are added or lost from the diploid set of chromosomes. Aneuploids may or may not represent balanced genotypes. The loss or addition may be found at polyploid levels. For example, the nullisomic is essentially aneuploid.

Cryptic structural hybridity[66] designates a chromosomal differentiation in very small segments that does not readily find expression in configuration at metaphase of meiosis. Pairing of chromosomes may be bivalent and apparently normal, for the segments that are differentiated are so small that no opportunity is afforded for abnormal configurations during synapsis. For these reasons a structural hybridity of this nature may be indistinguishable from the genetic hybridity.

11.3: Cataclysmic Origin of Species

The origin of a new species by gene mutation or chromosomal repatterning (inversions or translocations) is a slow process and requires a long time. Surprisingly, there exists in nature, alongside these slower processes, a very rapid method that can catapult a new species into existence within a generation or two.[7] This sudden origin is called "cataclysmic evolution."[24] By this process a new plant is separated at once from its immediate parents and is destined to occupy new environments different from either, or both, of its progenitors (Fig. 11.1).[73]

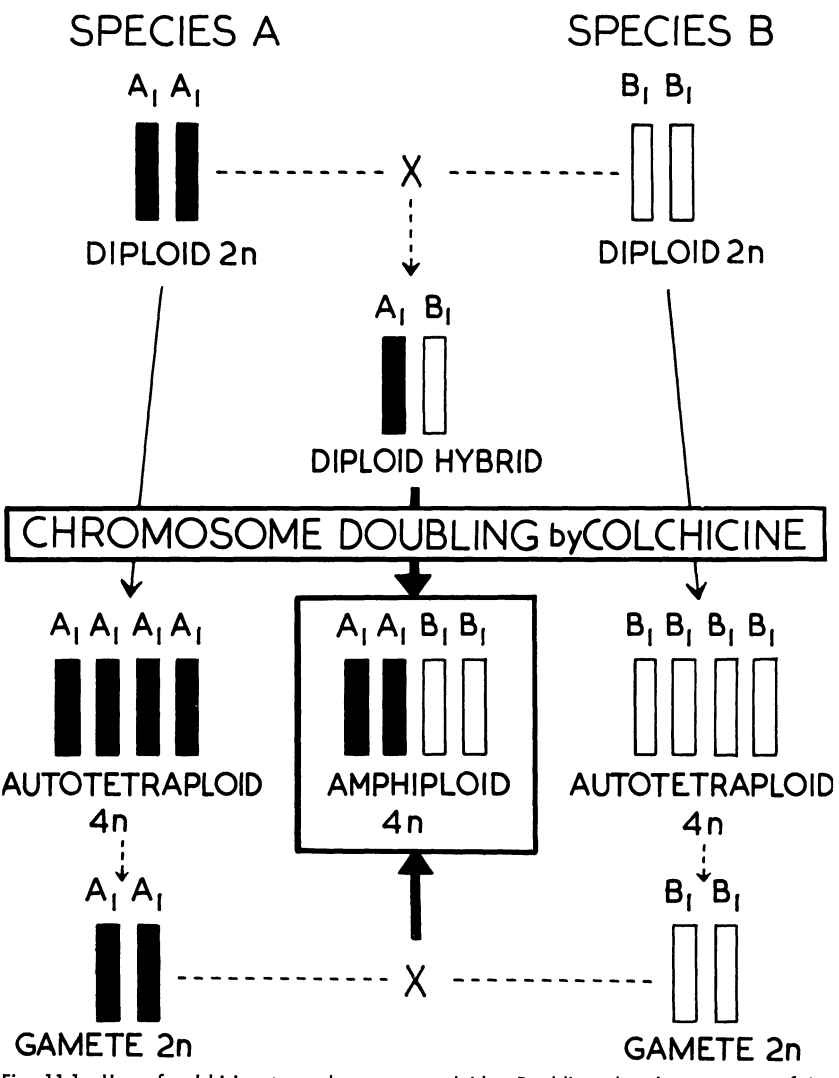

Fig. 11.1—Use of colchicine to make autotetraploids. Doubling the chromosomes of interspecific diploid hybrid. Amphiploids made by hybridizing two autotetraploid species. (After Wexelsen)

This kind of evolution was formulated as the $A \times B$ hypothesis by Winge in 1917 before any examples were well known, although the doubling of *Primula kewensis* was on record.[66] According to the $A \times B$ hypothesis, a polyploid series with a basic number of 7 would read 21, 28, and 42; or triploid, tetraploid, and hexaploid, respectively. These can originate as follows: A triploid, sterile hybrid arises from the hybridization between the diploid, $2n = 14$, and a

tetraploid, $4n = 28$; upon doubling of the 21-chromosome triploid, a hexaploid (42-chromosome) species originates.[49] In this way species hybridization, followed by doubling of the chromosomes, fulfils the principle of the Winge hypothesis. Among the wheats (Triticinae) there is an excellent chance to show how this mode of evolution accounts for speciation as well as the production of mankind's most valuable economic crop species, hexaploid wheat, (42-chromosome *Triticum aestivum* L.).[49] However, on a purely numerical basis and without a knowledge of the only known case to support his assumption, the $A \times B$ hypothesis was outlined to explain the origin of species with high chromosomal numbers. The data which Winge needed were published by Digby for *Primula kewensis*.[66]

The facts of cataclysmic evolution became clearer, for new tetraploids were discovered[34] or synthesized continuously from 1926. These include Müntzing's synthetic *Galeopsis tetrahit*;[51] *Primula kewensis*, arising under culture at Kew Gardens;[66] Karpechenko's *Raphanobrassica*,[24] a doubled intergeneric hybrid between radish and cabbage. Finally *Spartina townsendii*,[18] a new polyploid of recent historic times, is a new species which invaded a habitat not previously occupied. The mud flats along the channel coastline of England abound with this new species, but records show that prior to 1870 no plants were present in this area.[18]

Two important conclusions emerge from the numerous studies dealing with polyploidy and evolution. (1) Polyploid species are abundant in nature; by one estimate as many as 50 per cent of the flowering plants are in some duplicated form. (2) Valuable economic crop species (food, fiber, and others) are polyploid, e.g., bread wheat, cotton, oats, sugar cane, tobacco, grapes, berries, nuts, and many other horticultural and floricultural species. In the first instance our problem may be called cataclysmic evolution in nature; in the second, evolution under domestication.[48]

Polyploid agricultural species originated through the years in nature without man's guidance, but under his hand and through his selection they may have become quite different species than if left to natural processes of selection. When man eliminates certain types and nurtures the environment for his choice plants, the situation is not comparable to nature's elimination process and selection that goes on competitively without cultivation. Nevertheless, the problems of evolution in nature and under domestication[48] are very closely interrelated. That is why closer integration of theoretical and practical work seems advisable in polyploidy research. Increasing the information about the origin of polyploids in nature improves our position in the planning of a new hybridization program.[66] Furthermore, the data from countless selections by the practical breeder could be valuable for analysis with purely theoretical objectives in mind.[67]

When colchicine was discovered as a tool for doubling the chromosomes, it was believed by many that evolution was about to be speeded up out of proportion to anything known. The tool, colchicine, did in fact remove a serious bottleneck[66] in permitting a doubling of the species hybrid by a new and more efficient method than ever before available. Many newcomers to the ranks of new species have been produced; this is evident if we compare our list of amphiploids produced since 1937 with the list made before that date. There is no doubt of a speeded-up tempo, but unless one possesses a broad and deep knowledge of cytogenetics, he will fail to see that the expected "miracles" have been forthcoming. The introduction of a new variety of wheat by ordinary standards requires about 15 years.[66] To produce a new polyploid variety is as difficult, if not more so.

11.4: Classification of Polyploids

The two principal classes of polyploids are (1) autoploids derived from homozygous diploids, e.g., tetraploid maize,[60] and (2) amphiploids, like *Raphanobrassica*,[24] resulting from hybridization. These two types are not difficult to distinguish. They are extremes with the autoploid carrying four sets of homologous chromosomes *AAAA*, and the amphiploid, two diploid sets *AA* and *BB*. The difficulties in classifying polyploids arises when dealing with examples between the different types, that is, polyploids with both autoploid and amphiploid characteristics.[66] There are many cases — and more are being made continuously — that are intergrading types and, as such, are not easily classified into the autoploid or the amphiploid category.

Problems of classification in polyploidy are similar to those in other systematic studies. For example, everyone agrees on which individuals of the species belong to the Mammalia and the Spermatophyta; however, among the microorganisms a classification problem has new difficulties. Since the bacteria are so widely studied in relation to human disease, the medical bacteriologists find it illogical to group them with the fission fungi, or Schizomycetes, of the plant kingdom. As a matter of fact, some bacteria do have plant and animal characteristics, and so present a distinct problem in classification. Likewise in polyploidy, the borderline cases have characteristics that are both autoploid and amphiploid. As colchicine increases the number of polyploids, the intergrading types are increasing at the same time.

The artificially induced hexaploid *Phleum nodosum*, created by colchicine,[55] may be used as an example of the disagreement on classification because the true nature of its autoploidy is in dispute. When all the evidence is carefully reviewed in this case, the complex-

ities of classification become very real. These are problems requiring further study which cannot be resolved entirely in this review. There are other cases. In fact, the group between the autoploid and amphiploid provides the most interest and perhaps the greatest opportunity for practical and theoretical work in polyploidy. Even though one cannot decide definitely on the classification, there is no need for concern, for he may utilize the opportunities presented by these intergrading polyploids without classifying them.

One way to explore this group has been opened by an inquiry into the special kind of polyploid called the "segmental allopolyploid."[66] Good reasons were given to justify the establishment of this special group. Some types of polyploids have segments of chromosomes so closely associated that pairing is between the two parental genomes, and therefore they cannot be considered as strictly amphiploid; but in other segments, there is enough differentiation to prevent pairing of the chromosomes that originate from the different parents. Viewing the chromosomes segment by segment, instead of as whole chromosomes or even whole genomes, gives one a more critical picture of the basis for borderline types between the autoploid and the amphiploid. Theoretical and practical aspects are greatest among the polyploids that fall between the unquestionable autoploid and amphiploid.

Pairing of chromosomes is of limited value in classifying the polyploids even though this cytological method is one way to point out the difference between the autoploid and the amphiploid. Some diploid species hybrids may show pairing at the diploid level, but this does not necessarily happen. On the other hand, complete lack of pairing at the diploid level does not insure total bivalents at the polyploid stage.[42] Less and less reliability is being placed on pairing of chromosomes as a measure of homology and a means of distinguishing the autoploid from the amphiploid. As more examples come into view, the case for pairing is increasingly complicated. Other factors must be considered.

Sterility and fertility characteristics may separate the amphiploid from the autoploid. The latter is invariably less fertile than the diploid, and the amphiploid changes from a sterile condition to a fertile one upon doubling of the chromosomes. In reviewing many cases, one can find wide variation in degree of sterility among the autoploid and the amphiploid cases. Actually, the causes of sterility are so complex that this relationship is of little help in trying to classify the two types. Yet basically, sterility may be closely related to some basic cytogenetic mechanism.

The best solution to the classification problem appears to be the chart developed by Clausen and his colleagues[18] on which they place the amphiploids in a relative position depending upon a series of

characteristics that place the type closer or farther from one of the two classes. Table 12 of their work is worth considerable attention for those interested in the classification of polyploids. As would be expected, the known polyploids form an intergrading series from the extreme autoploid to the amphiploid, which is a completely diploidized type. Colchicine-induced polyploids cause increasing intergradation as more and more examples appear.

For purposes of reviewing the colchicine-induced polyploids, resorting to taxonomic authority has served a very useful purpose. If the polyploid has been a product of doubling a species hybrid involving accepted species, then the type is considered amphiploid, while the diploids made tetraploid are autoploid. Admittedly the system is artificial and does not delve into the real problem that makes a polyploid what it is. However, with the view of handling large amounts of data and many polyploids, this method of classification is simpler. At no time has the basic feature of the segmental allopolyploid or its significance been overlooked. Those characteristics that are peculiar to the segmental allopolyploid are important practically and in certain evolutionary aspects.

11.5: Principles of Polyploid Breeding

Within five years, from 1938 to 1942, examples of all the major agriculture species of Sweden were converted into polyploids.[46, 69, 1] In other places throughout the world vast numbers of polyploids were created at about this same time. Colchicine accounted for many of the new polyploids, but few of these could be used in agriculture.[73, 65, 49, 54, 56, 57, 63, 35, 62, 44, 19, 21, 22, 30, 32, 3, 5, 8, 9, 15, 16] This may come as a shock to practical agronomists. A re-examination of the principles basic to polyploid breeding was needed. Since so much material was at hand, polyploids were used to test a number of points about chromosome doubling as a method of plant breeding. The principles enumerated below have been stated directly as such or indirectly through the work of a number of investigators.

The application of colchicine permitted the production of large numbers of polyploids from diploids. One would expect these new polyploids to replace the standard diploid varieties.[60] However, artificially induced polyploids are, at the beginning, "raw" polyploids without exception.[46] Such types are generally unselected, so the task of plant breeding has only begun after the polyploid has been made.[46] Too many investigations disregarded the principle of raw polyploids and tested the tetraploids against the selected diploids. Naturally, the tetraploids failed to measure up to diploids in all-around performance. What is even more surprising is the condemnation of colchicine when tetraploids, apparently as raw polyploids, failed to

outperform the best diploids. Statements that colchicine causes "harm"[47] to the plants are also difficult to understand.

A second principle well known to practical breeders is the use of large populations. If one starts with a few plants, his project is doomed before a start has been made. Two qualifications should be stated in this respect. The self-fertilized species should be used with more strains and fewer plants from each, while the cross-fertilized types demand many plants, but these can be taken from fewer strains. In both instances, large numbers of tetraploid genotypes must be made as the material for future selection work.[46] Naturally, a few plants cannot serve as a substitute for mass production.

Each successful tetraploid must eventually have genotypical balance. Through selection the relation between plant and its environment must be brought into an adjustment.[18] Practical breeders are acquainted with the need for the all-around performance of more than one characteristic. It is not enough to acquire disease resistance, or some other quality, to the exclusion of those equally as important.[67] The new tetraploids are no exception in this respect. The transfer of a specific gene for disease resistance must not be permitted at the expense of the whole genotype which may be thrown out of balance – that is, if success in a practical way is anticipated. Therefore, the opportunities for selection begin with the polyploid, and the difficulties are also started as we shall learn in subsequent sections.

The genetic traits of the polyploid are an accumulation of those contributed by the diploid. It does not follow that a very good diploid will always give rise to the best polyploids. But there is this rule to be observed that a polyploid, like the diploid, is a plant with genetic traits that segregate and respond in selection according to the same rules as the diploid.

In judging the chromosomal numbers of natural species, there is a law of optimal numbers above or below which the maximum performance or adaptation cannot be expected. The polyploid series of *Phleum* is a good example.[46] Those types with best characteristics as polyploids were found in the numbers 6×7, and 11×7. One cannot expect to achieve success by doubling a tetraploid, so the diploid species are needed for a start. Chromosomal doubling of natural tetraploids in cotton from 52 to 104 chromosomes creates very weak and poor plants; obviously this exceeds the optimum number.[8] There is, however, another point to be remembered: If the number of diverse genotypes can be increased during the process of doubling high numbers with plants having good fertility, vigor and growth are possible. Merely stating that the numbers cannot be above a certain value is too limiting. In nature the natural polyploids are combinations of two or more genomes that can be recognized. For example, the hexa-

ploid wheat combines three genomes, and after this process the optimal number of 42 seems to be attained.

Cross-fertilizing, or allogamous, species are more promising as a group than the self-fertilizing types. This general rule seems to hold for a large number of plants included in the Svalof experiments. Some qualification needs to be made, for the sampling was not as extensive as might be desired. The changes from incompatability to compatibility upon doubling the number of chromosomes is an involved genetic problem, not merely a result of the tetraploid nature, but consisting of a combination of events that create the changes.[16]

The autoploids are almost without exception less fertile than the diploids.[60] Therefore, seed and fruit yields, if dependent upon seed production, will at once suffer in the polyploid stage, at least before selection can be done to rectify the situation. The sterility barrier is by-passed when a hybridization is included with the doubling; then the degree of fertility generally improves, but not always. The principle of reduced fertility after polyploidy from the diploid should always be considered by every one starting a new project. Then the changes that might be induced by selection in the later generations can be considered along with the sterility-fertility relations. Granted that fertility levels can be raised by selection, the danger of introducing other changes constantly attends the selection processes.

The part of the plant to be used for economic production becomes a first consideration, for the root and shoot yields will not be influenced by sterility. Vegetatively propagated plants are a new problem. They need not pass through the reproductive cycle that is so critical to a polyploid at many levels. Perennial plants are favored, and plants that produce propagating shoots like the grasses are immediately more favorable than the strictly seed-producing annuals.

A principle of transfer of characteristics from one species to another has been mentioned frequently in polyploidy work. Among many species the favorable traits are prominent in the wild species. There is at once a desire to introduce this character into the valuable commercial species. A notable case is the mosaic resistance transfer in tobacco.[17] This problem is discussed in greater detail later, but it should be noted that the transfer of such a trait is in effect a problem of polyploidy breeding. On a plan in blueprint stage, the idea appears relatively simple, but now it is well known that accomplishment is quite difficult. One of the greatest obstacles in transfer is the introduction of undesirable traits along with the desirable ones being sought.

Combining the good features of two diploid species into the amphiploid is another aspect of how hybridization and the doubling of chromosomes offer opportunity for future programs of selection. A

new species such as the *Cucurbita moschata* × *C. maxima* amphiploid combines good traits from two diploids. A new species of economic potential is apparent. However, interspecific segregations in the fifth and sixth generations show that a lack of uniformity can be expected (cf. Chapter 12). Such variation is not what the breeder hopes for in a true breeding variety. By transfer of whole genomes into a hybrid the characters of the polyploid can be influenced. If in later generations there is pairing between the two genomes that originated with the two species, the chance for segregation is good. If the segregates are undesirable and if the interchange is so great that the original type is lost, all the transfer is circumvented by the after-breeding effects. Transfer in *Gossypium* has presented a very difficult problem, that of introducing the good characters and maintaining all the original traits of the cultivated varieties. In spite of the problems, the principle of transfer is basic in polyploid breeding.[61]

The advantages balanced against the disadvantages are necessary for a final evaluation.[51] No tetraploid within a certain species may be expected to surpass the diploid in all respects. Therefore, the desirable traits balanced against the unfavorable ones should be calculated to see whether the new result is in favor of the tetraploid or the diploid. Triploid sugar beets are not perfect, but there is the important fact that the triploids can be grown to a larger root size before the percentage of sucrose decreases than is the case for the diploids.[58] In this way the triploid has an advantage over the diploid, while for seed production, germination, and growth problems the triploid is sometimes at considerable disadvantage beside the diploid. Tetraploid rye offers another notable example of balancing two sets of characters.[51]

All plants arising from treated generations may not be totally tetraploid. The diploid cells may be found mixed with the tetraploid, and a mixoploid condition may persist.[37] Or the layers of cells may differ one from the other, so that the shoot apex is stratified with respect to its ploidy.[23] These are called periclinal chimeras discussed in Chapter 14 (The Aneuploids).[13] From the point of view of polyploid breeding the mixoploids and chimeras are very important problems. The reversion of polyploid to diploid is sometimes explainable on the basis of a chimera, or sometimes it may arise from cross-breeding.

Stabilizing the polyploid by selection and by preventing the reversion to the diploid or through segregation, to some inferior type is a problem that confronts the plant breeder after the polyploid has been produced. The first and second generations may be quite uniform, but later generations less so. Or the first generation may have defects that yield to selection in later generations. The effectiveness

of selection between diploid and amphiploid is one of degree and speed rather than absolute difference. Genetic types can be isolated more quickly in diploids than in polyploids if one can base his evidence on a specific character and extend the idea to a whole set of characters.* Selection as a result of interspecific segregation creates a good opportunity for making wholly new lines.[66]

Regardless of the plant, whether diploid or tetraploid, the testing methods are important to success in measuring the gains made, in keeping the good qualities, and in raising the standards if possible. In tetraploid rye the testing side by side of diploid and tetraploid is impossible, and consequently an adjustment must be made by a yield factor with another plant.[51] This at once complicates evaluation of the polyploid against the diploid. There are many other problems of testing peculiar to certain plants, and tetraploids are involved because the success of the polyploid may depend upon the mode of testing rather than the qualities of the polyploid itself.

The list of principles is not complete in the above survey, but a start has been made. More information is needed before the additional principles of polyploidy breeding can be described in greater detail.

11.6: The Scope of Research

Colchicine increased the frequency of induced polyploids beyond that possible with any other method known up to 1937. This discovery had two major effects upon research in the plant sciences all over the world. (1) Polyploidy, already a subject of study, was increased immediately. (2) New programs were started because greater reliability could be placed upon this technique and much time could be saved in converting the diploids into polyploids. The net result of these two developments has been an unusually great expansion in research with polyploidy in many nations.[44, 54] In fact, a detailed review of all work with colchicine goes beyond the permissible allotment of space in this review.

One might single out specific cases where certain scientists have had an exceptional influence upon polyploidy and greater than average progress has been made accordingly. For example, the personal interest that Vavilov took in polyploidy led to great activity in cytogenetics in Russia.[70] In Sweden, Nihlsson-Ehle made special efforts to organize laboratories such as the chromosome laboratory at Svalof and other institutes in that country.[46] These and other special institutes[43] throughout the world were at work on problems in polyploidy before colchicine became known as a tool for creating poly-

*See Reference No. 103 in Chapter 12.

ploids. When colchicine appeared to be useful, its future possibilities were expressed in several American papers[70] published by Chronica Botanica in 1940. A broad view was taken at this time.

The progress made in Sweden from 1937 to 1947 was rapid. Scientists from every nation observed the scope of this work as a result of demonstrations made before two international congresses, the genetics meeting of 1948 and the botanical meeting of 1950. Obviously, the discovery of colchicine in 1937 appeared at a very favorable time in the history of plant sciences in Sweden. A large amount of work was done in Russia from 1937 to 1947, but less attention has been given to this contribution.[74] Already in 1945, Professor Zebrak reported in a lecture at the University of California that numerous polyploids in the *Triticum* group had been made, perhaps not exceeded elsewhere in the world.[74] The extensive report on the situation in biological sciences in Russia made in 1948 gives a general survey of the status of research with polyploidy before 1947. After 1948 the use of colchicine was apparently not encouraged in Russia.[47] There can be no doubt that Vavilov had an important influence on the use of polyploidy as a research method.

Japanese geneticists have made direct and special contributions to practical and theoretical phases of polyploidy.[54] The triploid watermelon, triploid sugar beet, tetraploid radish, and tetraploid melon have been put into agricultural practice since 1937.[54] Much progress has been made at the Kihara Biological Institute, Kyoto, where a number of workers have been able to make their contributions. Furthermore, the influence of this laboratory was directed to other institutes in Japan. Polyploidy has been a familiar subject, and there has been close integration of theoretical and practical problems under the direction of one group of workers.[43]

Accomplishments in the field of polyploidy by three nations, Sweden, Russia, and Japan, are quite out of proportion to the relative number of scientists, and particularly of geneticists, in each country. In this respect, the progress made in the United States is far behind these others if one compares the total work in plant sciences in relation to the progress made in the area of polyploidy. Therefore, one cannot understand why colchicine and polyploidy are thought to be tools owned solely by America. They are not. In fact, no nation can claim a priority in the use of colchicine and in progress made by its application to polyploidy. The records of the Seventh International Genetics Congress show some unbalance, but by the time the Ninth Congress was held, there was an equalization, so that no single group has dominated the program of colchicine and problems in polyploidy. Historically the situation has been clarified since the early period of work with colchicine.

There is another aspect in the scope of research with colchicine that tends to be overlooked. Scattered throughout the world, special institutes were at work on species whose background was recognized to be polyploid, such as *Gossypium*,[8, 15, 67, 35] *Nicotiana*,[35] *Triticum*,[49, 74] *Solanum*, and others. Theoretical problems and the practical importance of polyploidy were well known before 1937. One outstanding case is the British Empire Cotton Research Station at Trinidad, British West Indies, where diploid and tetraploid *Gossypium* was studied in detail (cf. Chapter 12). Soon after colchicine became known, it was applied to the sterile hybrids on hand.[67] The drug was merely incidental to the whole project, and many polyploids were made as a matter of routine in the larger program. For these reasons research with colchicine did not get prominent notice in their publications.

The application of polyploidy breeding in *Nicotiana* began before colchicine was discovered. After 1937 the number of polyploids for this genus was increased.[17] A transfer of disease-resistant traits from one species to another is an example of polyploid breeding and a contribution of experimental genetics.[17]

Breeding programs with forage species,[4] *Triticum*,[49] fruits, and flowers are under way in many places. The state and federal stations in the United States alone represent a large program.[22] Polyploidy is included in many of these programs. Public and private institutions throughout the world have put colchicine to work.

A complete list of research centers and projects using colchicine would be large. The bibliography and list of polyploids indicate the international character of such research.

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