TECHNICAL BULLETIN No. 238 JANUARY, 1936

New York State Agricultural Experiment Station

Geneva. N. Y.

SEEDLESSNESS IN GRAPES

A. B. STOUT

PUBLISHED BY THE STATION
UNDER AUTHORITY OF CORNELL UNIVERSITY

ABSTRACT

IN considering the character of fruits and seeds of grapes it is necessary to recognize that the unit for the production of a seed is the ovule; that the unit for a fruit is the ovary with its complement of ovules; that the flower is a unit in respect to the relative development of the stamens and pistil; and that the unit in vineyard culture is the individual plant of a clone with its clusters of flowers and fruit.

In grapes there are three main types of ovules, *viz.*, abortive at some stage and unable to function in fertilization; functional in fertilization, but the seeds abort during development—a condition here termed *stenospermy;* and able to develop by apogamy or after fertilization into a hard seed which may or may not contain an embryo.

The fruits of grapes are of three main classes, *viz.*, *parthenocarpic,* either obligate, when all ovules are aborted, or facultative in which case seedless fruits develop only when there is no pollination; *stenospermocarpic,* when one or more stenospermic seeds are present; and *seeded,* when a berry contains one or more hard seeds. It is the rule that stenospermocarpic and seeded fruits contain some aborted ovules and that the proportion of aborted ovules in such fruits is quite constant for a clonal variety.

"Seedless" fruits are either parthenocarpic with mere rudiments of seeds or stenospermocarpic. In the typical stenospermic seed abnormalities appear soon after fertilization in the feeble development of sclerenchyma and of other tissues which combine to give shape, size, and character to the seed. The endosperm and embryo remain in a partly developed stage and the largest seeds in mature berries are undersized, abnormal in shape, and soft or pulpy.

The clusters of fruit produced by a plant may contain berries that are alike and of the types mentioned above, or there may be mixtures of two or even three types of berries. In these cases the expression of vegetative parthenocarpy may be either obligate or facultative. Further complications in partial variations in the character of the fruit involve stimulative parthenocarpy and apogamy. Also, poorly filled clusters of fruit occur (1) when some flowers have rudimentary or aborted pistils, (2) when some ovaries contain only aborted ovules and there is no parthenocarpy, and (3) when pollination is necessary for fruit but some pistils are not pollinated.

Various seedless grapes chiefly of the tender vinifera types were used as pollen parents in crosses with hardy seeded grapes. A rather large proportion of the first generation seedlings bear seeded fruit with at least one hard seed to a berry, but for certain seedlings the fruits contain only aborted seeds of small size. Between these two extremes there are many intermediates in regard to the maximum development of seeds. More than 80 seedlings bear fruits with stenospermic seeds. Several of these are being propagated for trial culture in New York State.

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(Slightly Reduced)

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SEEDLESSNESS IN GRAPES

A. B. STOUT[1]

INTRODUCTION

This bulletin is concerned with the nature of seedlessness in grapes in respect (1) to when and where the abnormal changes or abortions appear in the development of ovules and seeds, (2) to whether or not fertilizations occur, (3) to the determination of the different types or classes of seedlessness in grapes, and (4) to the relation of seedlessness to other sterilities and to the production of fruit.

In its immediate application and interest this study is concerned primarily with the degree to which seeds develop in the mature fruits of seedlings which are to be classed as seedless or near-seedless and which have been obtained in a project[2] in breeding that aims to develop new hardy seedless grapes of merit for culture in a climate such as that of central New York. In this breeding, various seedless grapes, chiefly of the vinifera varieties cultivated in semi-tropical countries, are used as pollen parents with hardy seeded types as seed parents. Of the seedlings grown to maturity many are seeded, but about 80 different seedlings have already been obtained which bear seedless fruits or fruits that have only undeveloped seeds which are usually soft and pulpy. Other seedlings have fruit with seeds that are somewhat more developed and which may be called near-seedless. Some of these seedlings possess considerable merit and the most promising of them are being propagated for trial under cultural conditions.

Morphological and cytological studies were made to determine the degree to which seed-like structures develop in the mature fruits of various seedlings and studies were also made of the early stages in development to determine where the processes of abortion begin and what types of parthenocarpy and seedlessness are involved. For comparison, similar studies were made of the development of normal seeds and of the extent or degree of the abortions in the seedless grapes used

[1]Director of Laboratories, The New York Botanical Garden, New York City.
[2]This research is done by the writer in cooperation with the Division of Pomology of this Station. Reports of the plans of the project, of the methods employed, and of certain of the results have already been published (Stout, 1921B; 1928; 1933). Further details are given in the acknowledgment on page 64 of this bulletin.

EXPLANATION OF FIGS. 1 TO 4

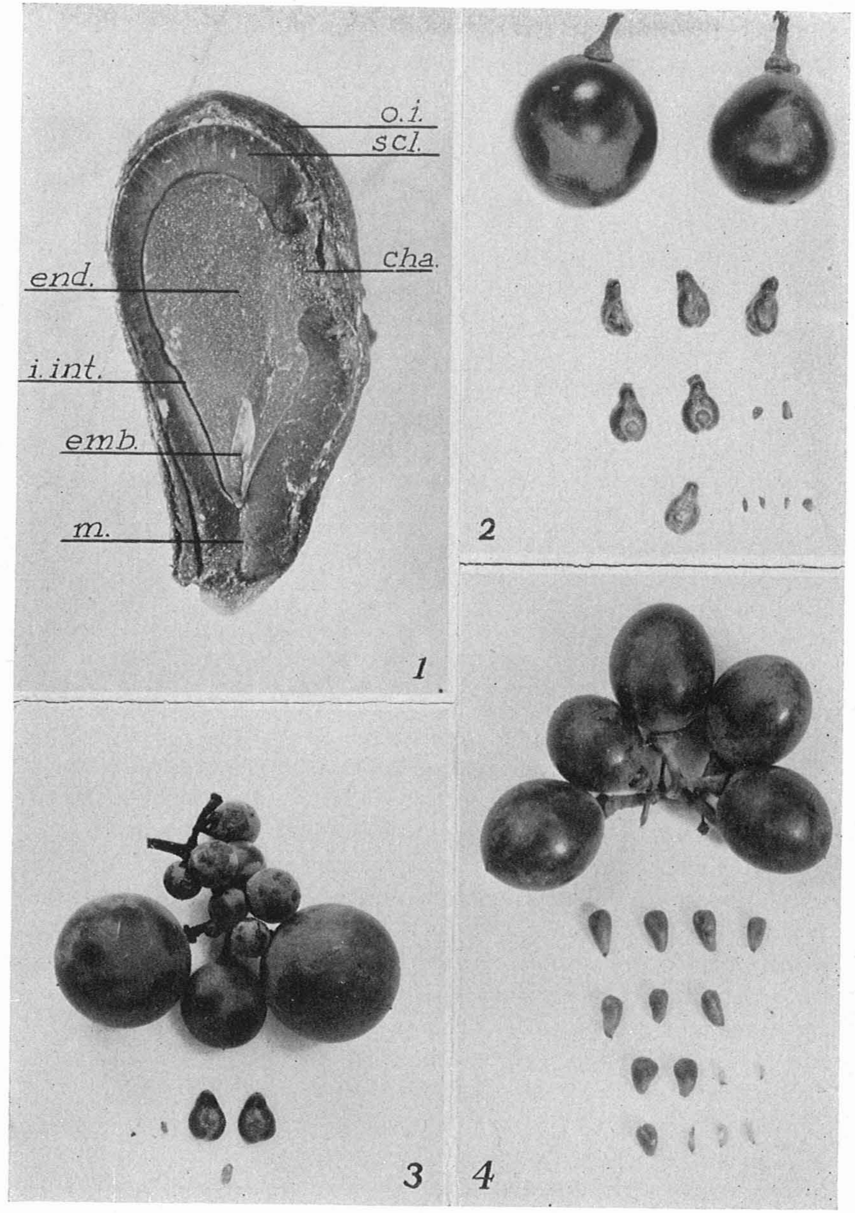
Figure 1 is enlarged to 9 diameters; others are natural size. In this and in following plates the seeds or aborted ovules in a single row came from one berry.

FIG. 1.—A longitudinal section of a normal mature seed of the Concord grape: *emb*, embryo; *end*, endosperm; *i. int*, inner integument with its inner layer of a conspicuous dark-color; *scl*, sclerenchyma which is a bony tissue of thick-walled cells developed from the inner layer of the outer integument; *cha*, break in the sclerenchyma where chalazal pad is located; *o. i*, the outer part of the outer integument which is somewhat fibrous and papery in character; *m*, location of micropyle.

FIG. 2.—Fruits and seeds of seedling No. 13335 which has the Black Corinth grape for its pollen parent. The berries contain as a rule from one to three normal seeds and the ovules which do not thus develop are aborted to small flakes or are not found. The aborted ovules are not able to function in fertilization.

FIG. 3.—Fruit of seedling No. 13201 which has for its parents Ontario by Concord Seedless. This illustrates partial variation which involves facultative vegetative parthenocarpy. The large fruits contain one or more normal seeds which result from pollination and fertilization. Along with these seeds there are aborted ovules and for those this condition is obligate. The small fruits develop when pistils are not properly pollinated; all the ovules abort but for some of them the abortion is merely facultative and due to the fact that there was no opportunity for fertilization.

FIG. 4.—Fruits and seeds of seedling No. 18110. The largest seeds are soft-gritty and endosperm and embryo may be rather fully developed. Figure 39 shows a cross-section of a seed of this grape.



FIGS. 1 TO 4.

as parents in the breeding. As a basis for reporting on these data, a survey of the different types of sterility in grapes will be made in respect to their relation to seedlessness and to fruitfulness.

DEVELOPMENT OF NORMAL SEEDS IN GRAPES

In grapes it is the rule that a normal viable seed is developed from an ovule and its fertilized egg after pollination. Layers of stone cells or sclerenchyma develop in the seed coat and the normal mature seed (Figs. 1 and 38) is hard and bony and not easily crushed. Thus the wild species of *Vitis* and many of the clonal varieties in cultivation bear seeded fruit and the seeds are hard. When the fruit is eaten, the seeds are very noticeable.

The developments concerned with the formation of normal and viable seeds in such cultivated grapes as the Concord and in various of the seedlings studies by the writer seem to correspond closely to the conditions in the vinifera grapes as reported in considerable detail by Berlese (1892)[3] over 40 years ago and more recently by Pearson (1932; 1933).

At an early stage in the development of the ovule the mature macrospore mother-cell is surrounded by two layers of nucellus at which time each of the two integuments consists of two or three layers of cells. The reduction division first gives two cells. Then the basal one divides, making three cells, of which the two at the micropylar end soon collapse and disintegrate, leaving one rather large and elongated cell which is the macrospore. Meanwhile, the nucellar tissue increases to several layers of cells which enclose the macrospore and form a zone between it and the micropyle.

By repeated divisions from the nucleus of the macrospore the eight nuclei of the normal embryo sac are formed. Three lie at the micropylar end and constitute the egg and the synergids; two nuclei fuse rather early and the resulting nucleus lies near the center of the embryo sac which now occupies a somewhat enlarged volume in the center of the nucellus; three nuclei become located at the chalazal end and, as a rule, these soon disintegrate. Meanwhile, there is multiplication of cells in the nucellus. From its epidermal layer a cap (the "calotta epidermica" of Berlese) develops at the mycropylar end and this cap may comprise as many as 10 layers of cells (Fig. 21). Also, several layers of cells form directly in front of the egg apparatus from the sub-epidermal tissue of the nucellus. Thus, after the pollen tubes reach

[3]Refers to Literature Cited, page 65.

the base of the micropyle, they must pass thru some 15 layers of cells which form a solid tissue before they reach the egg apparatus (Fig. 30). The development of this nucellar cap is a noticeable feature in the ovule of grapes. In the region of the micropyle the cells of the inner integument and especially of the innermost layer divide and form a projecting ring or hollow cone of tissue in front of the nucellar cap and the opening thru this may be almost closed by the impinging cells.

The penetration of pollen tubes thru the nucellar cap (Fig. 30) and various stages of fertilization have been observed by the writer in the Concord grape and in certain seedlings which have seeded and seedless fruits. The tubes press their way between the nucellar cells in a slightly sinuous but rather direct course to the egg apparatus where the end of the tube ruptures and releases the sperm cells.

The developments which follow fertilization have been described in considerable detail by Berlese (1892) and also various stages are well illustrated by him. Pearson (1932) summarizes the developments as follows: "In the normal *Vitis* seed a complicated series of changes in the integumentary tissues begins immediately after fertilization, so that within two weeks the fossettes at the sides of the raphe have infolded and the chalaza has grown over into the dorsal region of the ovule. The nucellar cells have enlarged tremendously and have also, for a time after fertilization, continued to increase somewhat in numbers. For at least two, and probably three or four weeks after fertilization the zygote does not divide, but the endosperm nuclei continue dividing slowly. They do not, however, encroach much upon the nucellus until the seed has reached practically its full size".

The above statement applies very well to our observations, except that the nucleus of the fertilized egg may divide somewhat sooner than indicated in the statement quoted above. For the Concord grape the best developed ovules in ovaries collected in late June and only a few days after fertilization contain as many as 20 endosperm nuclei in the embryo sacs in which the fertilized eggs are either undivided or have the walls of the daughter cells only faintly delimited. But some development of a proembryo may soon occur, for not later than 15 days after fertilization at least some of the ovules that are destined to become normal seeds are developed to the degree shown in Fig. 21 for which the following description may be given:

The *sclerenchyma* is rather fully delimited in respect to number, size, and arrangement of its cells, but the cell-walls are not yet noticeably thickened. The several layers of this tissue are formed from the innermost layer of the outer integument. The *inner integument* remains as a relatively thin zone of only three layers of cells, except in the region of the micropyle. Its outer layer becomes flattened and inconspicuous and its cellular nature is lost except near the micropyle. The innermost layer becomes a conspicuous layer of nearly isodiametric cells which appear almost solidly brown in the preparations that are stained with iron haematoxylin. The walls of these cells later undergo thickening. Berlese notes that this layer of cells reacts yellowish brown with potassium hydrate showing presence of tannin which he finds is never lacking in this tissue even before fertilization. This layer early becomes a conspicuous feature of both normal seeds and aborted seeds. The middle layer is present but is less conspicuous. Thus, the three layers of this integument are fully differentiated very soon after fertilization. The *nucellus* at this stage is well developed and the cells of its outer layer are quite distinct from the more enlarged nucellar cells within. They are also very different in character from the adjoining cells of the inner integument. They are densely granular and stain a bluish black, while the cells of the inner layer of the inner integument appear in a solid brown. The *calotta* which forms from this layer is well shown in Fig. 21. The *embryo* at this time is a globular mass composed of rather few cells, perhaps a dozen in all. The *endosperm* is a cylinder of delicate thin-walled cells which extends from the embryo to the chalaza. At this time the *fossettes* of the seed are well defined and the nucellus almost fills the entire inside except for the slender cylinder of endosperm.

Further increase in the size of the seed, the thickening of the walls of the sclerenchyma, and the development of endosperm and embryo now proceed rather rapidly.

Sections of mature normal seeds show a large well-developed embryo and endosperm (Fig. 1). The epidermal layer of the nucellus may persist in part, but its cells are empty and lying against it on the inside there may be a thin layer of compressed cell walls which are remnants of the inner tissue of the nucellus. About the embryo and extending to some distance above it toward the chalaza is a region in which cells are either absent or have little granular material in contrast to the adjacent cells of the endosperm which are packed with granular material. The sclerenchyma, developed from the inner layer of the outer integument, forms the bony part of the seed. The other layers of the outer integuments become a relatively thin covering that in the dry seed is somewhat fibrous and papery.

Thus, in the development of the normal seed in *Vitis* there is, immediately following fertilization, a rapid development of the outer integument and especially of the sclerenchyma. The complete differentiation of the three layers of the inner integument occurs early. The nucellar tissue expands chiefly, if not entirely, by the increase in the size of the cells and it occupies most of the space within the expanding integuments. During this period the proembryo develops slowly into a small globular mass of relatively few cells. Nuclei of the endosperm begin to appear immediately after fertilization. When the integuments and the nucellus reach approximately their maximum size and differentiation, the embryo and the endosperm begin a rapid development. Thus, there is for a time after fertilization a relatively delayed development of the embryo and the slow formation of endosperm, but there is a rapid development of integuments and nucellus.

STERILITIES AND VARIATIONS FROM NORMAL IN THE PRODUCTION OF FRUITS AND SEEDS IN GRAPES

The several types of sterility and unfruitfulness in grapes may be considered in respect (1) to the types of flowers, (2) to the reduction in the number of berries produced in a cluster, (3) to the reduction in the number of seeds in the berries (a) which are produced and (b) which are viable, (4) to the abortions in ovules or seeds which are involved in seedless fruits, and (5) to such other conditions as apogamy, facultative parthenocarpy, and partial variation in the nature of the fruit.

TYPES OF FLOWERS IN GRAPES

The male or staminate type of flower in grapes has a rudimentary pistil. Plants with only staminate flowers produce no fruit. When some flowers in a cluster are male and others are hermaphrodite, a condition that sometimes exists (Stout, 1921A), the number of berries in a cluster in proportion to the total number of flowers is thereby reduced. This is one of the conditions that result in loose and poorly filled bunches of fruit.

The imperfect hermaphrodite type of flower in grapes, as usually recognized, has reflexed stamens, but flowers which have erect-twisted or "crinkled" stamens (Stout, 1921A) may also produce only non-functional pollen. At the time of anthesis many of the pollen grains are shrivelled and more or less empty and those that are plump, or which become plump on sugar-agar media, are devoid of pores. In numerous tests which the writer has made no pollen of this type of flower germinated on the artificial media which yield excellent germination for the pollens of males and of the perfect bisexual flower. Numerous controlled self-pollinations by bagging have been made by the author for plants with reflexed stamens, but no fruits with seeds were obtained. The evidence is that the pollen of the reflexed stamens does not, as a rule, function in fertilization. It is possible that certain of the many seedlings and numerous clones which have this type of flower do produce some viable pollen. Nearly a hundred seedlings which were considered to be from fertilization with pollen of reflexed stamens are reported by Hedrick and Anthony (1915), who state that, "while this impotency may be absolute in many of the varieties, in some at least it is only relative". It is claimed (Ivanova-Paroiskaya, 1930; Negrul, 1934) that the pollen produced in reflexed stamens has some stimulating effect on the development of fruit even tho it does not germinate on the stigma.

It has been found by Dorsey (1914) that abnormalities develop within the pollen of the reflexed stamens of the Brighton grape *after meiosis* and at the time when the generative nucleus is being organized. The processes of abortion and arrested development are here confined to the microspores in the stamens, while macrospores and embryo sacs in the ovules of the same flower may develop normally, and with proper cross-pollination and fertilization there will be the development of fruit containing one or more normal seeds.

A morphologically female type of flower without stamens of any sort has been described by Baranov (1927 B and C) for certain vines that arose as bud-variations from the variety Mourrèdre. This variety usually has flowers of the perfect hermaphrodite type; but nine vines were found that bore flowers in which (1) the petals opened separately and remained attached and persistent on the pistil, (2) the stamens were entirely absent, and (3) the ovaries were 5- to 6-celled with 10 to 14 ovules. Artificial cross-pollination with pollen of hermaphrodite types resulted in a 100 per cent development of fruits. Flowers of this aberrant type have not been observed in any variety or seedling plant grown at the New York State Agricultural Experiment Station.

The perfect hermaphrodite type of flower has a well-formed pistil, which functions in the formation of fruit, and also anthers which usually contain considerable viable pollen. Many of the plants with this type of flower produce fruit to self-pollination. There are wide variations among plants having hermaphrodite flowers in respect to the length, size, and shape of stamens and pistils, and in the number and character of the seeds present in the berries. Also in seedless grapes the pistil may contain ovules that can not develop into normal viable seeds.

The perfect hermaphrodite type of flower is, apparently, not found among the wild plants of the species of *Vitis* native in eastern North America. This type of flower is present in numerous cultivated clones both of the vinifera group and of those most widely grown in New York State. (See discussion by Hedrick, 1908, and by Bailey, 1934.) Baranov (1927A) finds that the so-called "wild" grapes of middle Asia consist of perfect-flowered plants and imperfect hermaphrodites, but he concludes that these plants have been derived as escapes from cultivated grapes. In the Muscadine grapes the origin of perfect-flowered plants is a matter of recent record. A single wild plant of *Vitis rotundifolia* was found (Reimer and Detjen, 1910; Detjen, 1917) which had the large flower clusters and the erect stamens which are typical for staminate plants but which also had functional pistils in some of the flowers. This vine was used in breeding and many seedlings were obtained which had flowers of the perfect-hermaphrodite type. Perfect-flowered seedlings of Muscadine grapes were also obtained (Husmann and Dearing, 1913; Dearing, 1917) among the offspring of imperfect hermaphrodites $\times$ staminate plants and in the progenies of these plants numerous other perfect-flowered plants were obtained.

The above designation of types of flowers in grapes is based chiefly on gross structure. It has been noted especially by Stout (1921A) and by Steingruber (1927) that there are various intermediates or transitional forms between the three main types of flowers in grapes. Further differences are to be recognized especially in the character of the ovary and its ovules in relation to seedlessness.

STERILITY OF HYBRIDITY

Abortions of both pollen and ovules due to hybridity are definitely known for certain hybrids of *Vitis vinifera* and *V. rotundifolia* (Detjen, 1919). These two species possess different numbers of chromosomes and belong to quite distinct sub-genera. They do not hybridize readily; but when hybrids are obtained the pollen of those that bear perfect flowers is a mixture of shrivelled and plump grains and the ovules are so fully aborted that berries seldom mature. Here, the abortions involve irregularities *during meiosis* and not later, as in loss of maleness in unisexualism. For the hybrids which have reflexed stamens there is *also* loss of maleness from unisexualism.

The various clonal varieties of grapes cultivated in New York State are evidently hybrids derived from species which possess the same number of chromosomes. The extent to which sterility from hybridity has appeared in those seedling hybrids that have been discarded during the production of the cultivated clones seems not to have been determined. The seedlings which have survived evaluation and selection are those which are fruitful to the extent that well-filled clusters of berries are regularly produced and it is the rule that at least one seed per berry is developed.

POLYPLOIDY IN GRAPES

Several grapes are known to have 76 chromosomes (Nebel, 1929) or twice the diploid number normal for grapes of the *Euvitis* subgenus. The sets of chromosomes are "balanced" and the plants are fruitful and more robust than the diploid plants of the same type and the fruits are larger. Hybrids between these tetraploid clones and the usual diploid clones may be expected to give some plants that are triploid or that have other unbalanced sets or numbers. These may be expected to exhibit considerable abortion of pollen and of macrospores in ovules which is quite like that seen in hybridity. Sterility of this sort affects the production of fruit by all the individuals in which development of fruit depends on fertilization, but it does not affect the production of fruit by vegetative parthenocarpy.

REDUCTION IN NUMBER OF SEEDS AND IN NUMBERS THAT ARE VIABLE

The typical ovary in the genus *Vitis* is two-celled and each cell has two ovules (Figs. 22, 24, and 25). In at least one wild species, *Vitis labrusca,* as reported by Sturtevant (1890), the number of hard well-developed seeds in mature berries ranges to six. In certain cases some ovaries of the cultivated grapes may have three cells. But for the wild *V. labrusca* and for seeded varieties in cultivation, relatively few berries contain four seeds.

REDUCTION IN NUMBER OF SEEDS

An examination of the mature and ripe berries of such grapes as the Concord and Delaware shows that the majority of the seeds that are formed are quite uniform in appearance and that there is scarcely a trace of the ovules that failed to develop. There are, of course, some seeds that are undersized and immature, but there is not in these fruits a distinct class of soft, pulpy and partly developed seeds of the type seen in such seedless grapes as Sultanina.

Cytological studies were made of the ovaries of several varieties of seeded grapes, especially of the Delaware grape, which have an average of less than two seeds to a berry. These studies show that when the flowers are about to open all four ovules are, with few exceptions, present in each ovary and that they are quite alike in size and in the development of integuments, nucellus, and the early stages of the embryo sac (Fig. 24). Yet it is evident that many of these ovules do not even start the early stages of development into seeds, and *no amount or kind of pollination can increase the number of seeds.* There is some factor or condition which operates very regularly to reduce the number of ovules that are able to function in fertilization and to proceed into the early stages of seed formation. This failure may be termed *ovule abortion.*

The amount of ovule abortion may increase until it includes all ovules in certain ovaries. Then if there is no parthenocarpy, in which case at least one seed is necessary for the formation of a berry, the ovaries which contain only ovules with impotent embryo sacs will fall soon after the flowers shed pollen. The clusters of fruit, in such cases, are poorly filled for only those ovaries which have at least one functional ovule have a chance to develop into berries.

This condition is exemplified in the clonal variety Chasselas Gros Coulard which has been studied by Lazarevsky (1934). He reports that in this grape completely normal embryo sacs are rare and that frequently all the ovules in an ovary contain abnormal embryo sacs. He also reports that the development of the abnormal embryo sacs proceeds normally thru the reduction divisions that produce a macrospore in each ovule and that the further development of the female generation (the macrogametophyte, or the egg and its accompanying nuclei) proceeds to the three-celled stage or to the four-celled stage but stops before the eight nuclei (including the important egg nucleus) are formed. Here, as in the pollen grains of the reflexed type of stamens, the abnormalities appear in the development of the sexual generation *after* normal meiosis. This is quite distinct as to time and stage of action from the abortions *during* meiosis which are associated with hybridity and polyploidy.

Our studies confirm the report of Lazarevsky, mentioned above, that in certain grapes the ovules are all alike up to the point when the egg apparatus is being formed (Figs. 22, 24, and 25). This indicates that the differences in the nature of ovules reside in the sexual or haploid generation and arise after reduction division. It may be considered that the abortions are hereditary and are determined by segregations which give to the macrospores and to the embryo sacs that develop from them these differences in character.

EMPTY SEEDEDNESS

In addition to the abortions of the egg apparatus in ovules there are later embryo abortions which develop within certain of the seeds that become more or less hard and horny. As a result, for many cultivated grapes, the percentage of seeds which will germinate is low. Studies of these non-viable seeds (see especially Olmo, 1934B, page 376) show that a large number of them are almost "empty" but do contain some "shrivelled tissue, occupying a variable portion of the cavity". Such seeds usually float when placed in water, they may display abnormal coloring and they may be somewhat undersized and less hard than are viable seeds.

Thus, in the class of seeded grapes under consideration, there are three types of ovules, *viz.*, (1) ovules in which there is abortion of the egg apparatus and which do not start to develop and of which only mere traces remain; (2) ovules which develop into "empty seeds" due, presumably, to abortion of embryo and endosperm; and (3) ovules that mature into normal viable seeds. It is to be noted that no kind or amount of pollination alters the relative proportion of these classes which is characteristic of a variety. Olmo considers that the cause of empty-seededness is "to be sought in maternal tissue", that the differences in the amount of empty-seededness seen in certain varieties indicates the operation of hereditary factors; and that these factors are recessive. But it is to be noted that there are here at least three classes of ovules, that their distinguishing characters arise in the behavior of the embryo sac or in the developments which come from it, and that these arise after the segregations of reduction.

The relation of these two types of sterility (embryo sac abortion in unfertilized ovules and embryo abortion in the stages of seed formation) to the formation of fruit is evident as follows:

1. In the seeded varieties in which there is no ability to develop fruits by parthenocarpy, any ovary which develops into a berry must possess at least one seed. As soon as any considerable number of the ovaries contain only ovules in which there is embryo-sac abortion, the berries become fewer and the bunches are ragged and poorly filled. Such unproductive seedlings are promptly discarded in making selections for propagation.

2. When there is parthenocarpy of the vegetative type the ovaries which possess only defective embryo sacs will develop as seedless berries and those which possess functional embryo sacs will develop into seeded fruits provided there is adequate pollination (Fig. 20).

3. It will be noted in the following pages that the abortions which give soft, pulpy, seed-like structures provide another class of ovules which may exist along with the classes already discussed and also lead to the mixture of two or more types of fruit in the same clusters.

SEEDLESSNESS IN GRAPES

There are several well-known clonal varieties of the European grape (*Vitis vinifera*) which bear seedless fruits. The fruits of these grapes have either mere rudiments of seeds or soft seeds that are not noticeable when the pulp of the fruits is masticated. Some of these seedless grapes are important in commercial culture for the production of seedless raisins and also to some extent for use as table fruit. These seedless vinifera grapes are *not hardy* in the climate of New York.

Vines which bear seedless or near-seedless berries have appeared as seedlings or as bud sports from certain of the grapes in culture in New York. In most cases these have produced fruit of poor quality or the bunches were poorly filled. One of the best of them has been cultivated to some extent under the name Concord Seedless. Among the large number of grape seedlings grown at Geneva some seedlings have been observed which produce clusters of berries in which seeded fruits and seedless fruits are intermingled.

Several types or grades of seedlessness are to be recognized in grapes in respect to the extent, the degree, and the time of abortion in ovules and in seeds and in regard to whether fruit will be produced with or without pollination and fertilization.

PARTHENOCARPY IN CORINTH GRAPES

There is general agreement among those (Müller-Thurgau, 1908; Susa, 1927; Pearson, 1932; 1933) who have previously studied the currant grapes, Black Corinth (including Panariti), Red Corinth and White Corinth that these clonal varieties produce seedless fruit without fertilization. To this condition in plants Noll (1902) applied the term *parthenocarpy*. Later a further distinction was made and (1) the terms *vegetative* (Winkler, 1908) and *autonomic* (Fitting, 1909) were applied to a parthenocarpy in which there is no pollination and (2) the terms *stimulative* (Winkler) and *aitionomic* (Fitting) were applied to a parthenocarpy when the fruits develop after a stimulus such as that of the growth of pollen tubes or of parasitic fungi on or in the pistil.

POLLEN OF CORINTH GRAPES

The Corinth grapes have flowers of the hermaphrodite type and the anthers contain some viable pollen. The pollen grains of Black Corinth and White Corinth are rather uniform in size and appearance and the percentage of observably aborted and shrivelled grains is low. In germination tests on agar-sugar media made by the writer, however, only about 3 per cent of the grains of these varieties germinated and produced fine long tubes. Susa (1927), however, obtained a germination of from 10 to 20 per cent for the pollen of White Corinth. In our breeding work the pollens of Black Corinth, of the clone Panariti, and of the white Corinth have all been used in controlled cross-pollinations on seeded varieties and numerous viable seeds were obtained from which many seedlings that are certainly true hybrids are being grown. Since there is at least some viable pollen in the flowers, there is opportunity for self-pollination in these varieties. But pollination, either self- or cross-, does not change the berries from seedless to seeded. The parthenocarpy is obligate.

OVULES OF WHITE CORINTH AND RED CORINTH GRAPES

Pearson (1932; 1933) reports that the White Corinth and Red Corinth grapes have an extreme type of defective ovule; that the ovules have only one integument, which is the outer one; that there is no embryo sac; and that the nucellus often becomes extended until it protrudes far beyond the integument. The studies made by the writer are in accord with this report, except that embryo sacs in various stages of development were found in some of the ovules (Figs. 27 and 28). The cross sections of the ovaries of this grape collected at the time of anthesis show, as a rule, large ovules that are almost alike. There is only one integument; a tongue of protruding nucellus bends downward along and against the outer wall of the ovary (Fig. 27). In many ovules no traces of an embryo sac are found and the tissue is solidly filled with vegetative cells. In some ovules cavities of irregular shape are present in the nucellus, but without trace of sporogenous tissue or of embryo sac. In some cases, however, several well-defined nuclei of an embryo sac were present in the nucellus. Usually these are in the protruding tongue (Fig. 27) but sometimes they are located in the main body of the nucellus. But no embryo sacs were observed which appeared to be normal and functional.

OVULES OF BLACK CORINTH GRAPES

In the Black Corinth grape, according to Pearson, the integuments of the young ovules are present and normal but the egg apparatus is usually missing or is in various stages of degeneration at the time of anthesis. The pollen tubes penetrate into the ovary in profusion yet "they seldom enter the micropyle and only rarely penetrate the nucellar cap". Occasionally seeds of noticeable size are produced. Pearson reports that for one very unusual cluster of this grape which she studied "there were 200 seeds; 170 were tested by placing in water and only 66 of these floated; 60 of those which sank were planted and 16 germinated". In the material of Black Corinth studied by the writer the cross-sections of ovaries collected at time of anthesis showed, as a rule, four equally well-developed ovules in which some cytoplasm and nuclei of an embryo sac were present.

Harmon and Snyder (1936) observed a branch on a vine of the Panariti grape (said to be identical to the Black Corinth) which bore two clusters of seeded berries and they found that propagations from this branch bore only the seeded fruit. They also report that a survey in California of commercial vineyards of this grape revealed mixtures of large seeded berries and small seedless berries in certain clusters, bunches of exclusively seeded and exclusively seedless berries on the same branch, and vines that bore only one type of fruit. They report that the average of seeds in the seeded fruits was 2.44 and that the seeds were viable. It was concluded that these conditions indicate that mutation from the seedless to the seeded type of fruit had occurred.

FORMATION OF FRUIT IN CORINTH GRAPES

Thus, in the Corinth grapes there are two grades of imperfect ovules. In the White Corinth and the Rose Corinth the ovules lack one integument, the nucellus protrudes and it is often composed of vegetative cells only. In the Black Corinth the ovule is rather fully formed and the embryo sac is present in some degree of development but it is evidently rarely able to function in fertilization.

There seems to be some question as to whether the seedless fruits of the Corinth grapes are produced by vegetative parthenocarpy or by stimulative parthenocarpy. Pearson (1932) states that "it does not seem likely that the growth of the pollen tubes thru the style has anything to do with the set of Black Corinth berries". In discussing the results of pollination tests for parthenocarpy Pearson makes the following statements: "Of thirty clusters emasculated and bagged, not one set, but this does not seem significant in view of the fact that the 47 which were bagged either without emasculation or after having been emasculated and pollinated, did not set either. Since temperatures are often 10° C. higher inside bags than outside, it seems likely that, although normal Vinifera berries set well inside the bags, the entirely seedless vegetatively parthenocarpic berries are too sensitive to such adverse conditions in the hot summer weather of Davis. Four clusters on ringed shoots which were emasculated set perfectly. It has not been proven, however, that in the absence of the nutritional stimulus of ringing the stimulus of pollination is not necessary in the Black Corinth. Ringing increases the set of vegetatively parthenocarpic berries in other varieties".

H. P. Olmo, in a recent letter to the writer makes the following statement as to whether the Corinth grape will produce fruit without any pollination whatever. "I know of no evidence to show that this is so. Even though fertilization does not take place, it appears probable from some of the results I have obtained that a stimulus of pollination might be necessary to get a normal set".

That the fruits of the Corinth grapes are produced by parthenocarpy is certain, but whether this is vegetative or stimulative is not now fully apparent.

The summary of studies by Oinoue (1925) seems to suggest that the ovaries of the White Corinth as well as those of the Sultanina show growth responses to the stimulus of pollen-tube growth. But the statement that "the so-called seedless grapes are not due to the parthenocarpy but to the incomplete fecundation" which he calls "quasi-fecundation" certainly does not apply to the definite cases of vegetative parthenocarpy which occur in numerous grapes.

Numerous F_1 seedlings have been obtained in our breeding work from the use of pollen of Black Corinth, Panariti and White Corinth on various seeded grapes hardy in New York. With the possible exception of one plant all of these seedlings, thus far grown to fruiting age, have borne seeded fruits; but in some cases the seeds are "brittle" and in one seedling it has been determined that there is facultative parthenocarpy. The one plant mentioned above fruited for the first time in 1935. The clusters of fruit were few and were composed of a few large berries, in which the largest seeds were soft-gritty, and numerous small and entirely seedless berries. The F_2 progenies of the best of these seedlings are being grown in which at least some plants which bear only seedless fruit may be expected and also certain of these plants

are being used in selective breeding in the attempt to obtain the entirely seedless type of fruit in large sizes.

DEVELOPMENT OF FRUIT AND SEEDS IN SEEDLESS GRAPES SULTANINA, SULTANINA ROSE, AND BLACK MONUKKA

FLOWERS

The flowers of the seedless grapes Sultanina, Sultanina Rose, and Black Monukka are of the perfect hermaphrodite type and the pollen is highly viable. Of Sultanina pollen about 90% gives excellent germination in tests on agar-sugar media. The percentage of germination is also high for the pollen of Sultanina Rose and Black Monukka. These varieties are able to function as pollen parents.

POLLINATION

It seems certain that it is the rule that these grapes set fruit best when there is proper pollination. There are, however, some minor differences in respect to the conditions involved in the abortion of seeds and in the production of fruit. Goodspeed (1920) and Pearson (1932; 1933) both report that the fruits of Sultanina are produced, for the most part, only after proper pollination with viable pollen. Susa (1927) reports that relatively few berries start to develop on Sultanina when there is emasculation and no pollination and that these fruits are smaller than normal size. Negrul (1934) reports that Sultanina and Black Monukka will produce at least some berries with empty seeds when flowers are emasculated and not pollinated. For the Black Monukka grape the tests reported by Susa (1927) indicate that self-pollination is effective in giving sets of fruit. Pearson found that the emasculated and unpollinated flowers in the clusters of two shoots that had been ringed set fruits, but the berries were smaller than those of pollinated clusters and their seeds were more rudimentary. Evidently some of the ovaries of this variety may develop by vegetative parthenocarpy, at least after the ringing of a vine. There seems to be some tendency to vegetative parthenocarpy, but this is not to be depended upon for a good set of fruit and also it seems to yield not only smaller but fewer fruits.

THE OVULES

Pearson (1933) reports that in Sultanina, Black Monukka, and Sultana abnormalities in the integuments of the ovules exist at the time of anthesis. The inner integuments (Fig. 29) are elongated at their tubular tips in which the cells are less meristematic than in normal seeds and the micropyle is much larger than in the normal ovules. The writer has made studies of numerous cross-sections of the ovaries of this type of grape. Such preparations show all the ovules of an ovary in the same section. Such sections (Figs. 22 and 25) reveal that it is the rule that all four ovules are present and that at the time of pollination they are very much alike in the size and the development of integuments and nucellus. There are also nearly always embryo sacs in some stages of development and at this time it is not always possible to determine with any degree of certainty which ovules are able to function in fertilization and which are not. This observation also applies to the ovules of seeded grapes.

FERTILIZATION

The different studies regarding fertilization in these grapes are not in full agreement. Müller-Thurgau (1908) states that the ovules of the Sultanina grape are not fertilized, altho the pollen tubes may enter them. Goodspeed (1920) found that in nearly every flower examined the ovules contained some normal embryo sacs and that pollen tubes were found entering the micropyles and he concludes that, "There seems to be no doubt that successful pollination (possibly followed by fertilization) is one of the factors essential for the seeding of fruit in the Thompson Seedless grape".[4] Oinoue (1925) states that "the ovule of seedless grape is not degenerated but the fecundation stops suddenly soon after, and consequently seeds could not be formed" and that "the so-called seedless grapes are not due to the parthenocarpy but to the incomplete fecundation" which is called "quasi-fecundation". Pearson (1933) states that in the grapes Sultanina, Sultana, and Black Monukka abnormalities are frequent at the time of anthesis in the elongation and protrusion of the inner integument and that abnormal embryo sacs are frequent. She is, however, emphatic that "there is also a very high percentage of normal appearing embryo sacs and that in nearly every ovary two or three embryo sacs have pollen tubes entering them within two or three days after anthesis" and also that in such ovules the endosperm starts to develop. Pearson notes that the developments in the seed coat are abnormal; the middle layer of the external integument develops abnormally, and its inner layer develops into sclerenchyma very feebly. But embryos were found in 3 of the 10 largest seeds which were examined. Pearson "believes that as

[4]The Thompson Seedless grape is the same as the Sultanina.

a rule, at least one normal embryo sac has been developed and fertilized in each normal Sultanina berry".

Our own studies of Sultanina, Sultanina Rose, and of seedlings which have similarly aborted seeds indicate that pollen tubes enter some ovules in many of the ovaries, that fertilization occurs in some ovules, but that the embryos fail to develop altho they may remain apparently alive until the fruit is mature. There are, however, a large proportion of embryo sacs that are abortive and apparently unable to function in fertilization. Some of the ovaries contain only ovules which either do not function in fertilization or which do not proceed into even the earlier stages of seed development. The exact distinction between a stimulative parthenocarpy with abortion of all the embryo sacs and the fertilization of at least one ovule followed by early seed-abortion is difficult to make and possibly of little significance in this case.

THE BERRIES AND THEIR ABORTED SEEDS

The largest of the mature fruits of these grapes (Sultanina, Sultanina Rose, and Black Monukka) may contain one or more seed structures of considerable size (Figs. 5 and 6), but it is the rule that these are soft, somewhat misshapen, and much smaller than the normal viable seeds of ordinary seeded varieties. There are only minor differences in these varieties in the extent to which seeds, and especially the sclerenchyma in them, may develop. For all these varieties some berries have only remnants of ovules and even the largest of the berries usually have some such rudiments. Special studies were made of the seed-like structures in mature berries and also various stages of the early developments were studied. For Sultanina the writer has studied mature berries obtained in the New York markets and also fruit developed on vines growing at Geneva and at New York City. In certain clusters of fruit all the berries, even the largest, contained only small seeds that were thin-walled and empty. In other bunches many of the largest berries had from one to three soft seeds of considerable size and cytological preparations of these seeds showed that, in some of them at least, a small globular mass of cells representing the embryo was present (Fig. 34). But in many of the seeds studied no endosperm was found and there were only traces of a nucellus (as in Figs. 33, 40, and 45). The sclerenchyma was composed of several layers of cells, especially about the micropylar end, and the walls of these cells were somewhat thickened. The smaller and more rounded berries contained only small rudiments of seeds or only aborted ovules.

EXPLANATION OF FIGS. 5 TO 9

(All figures are natural size.)

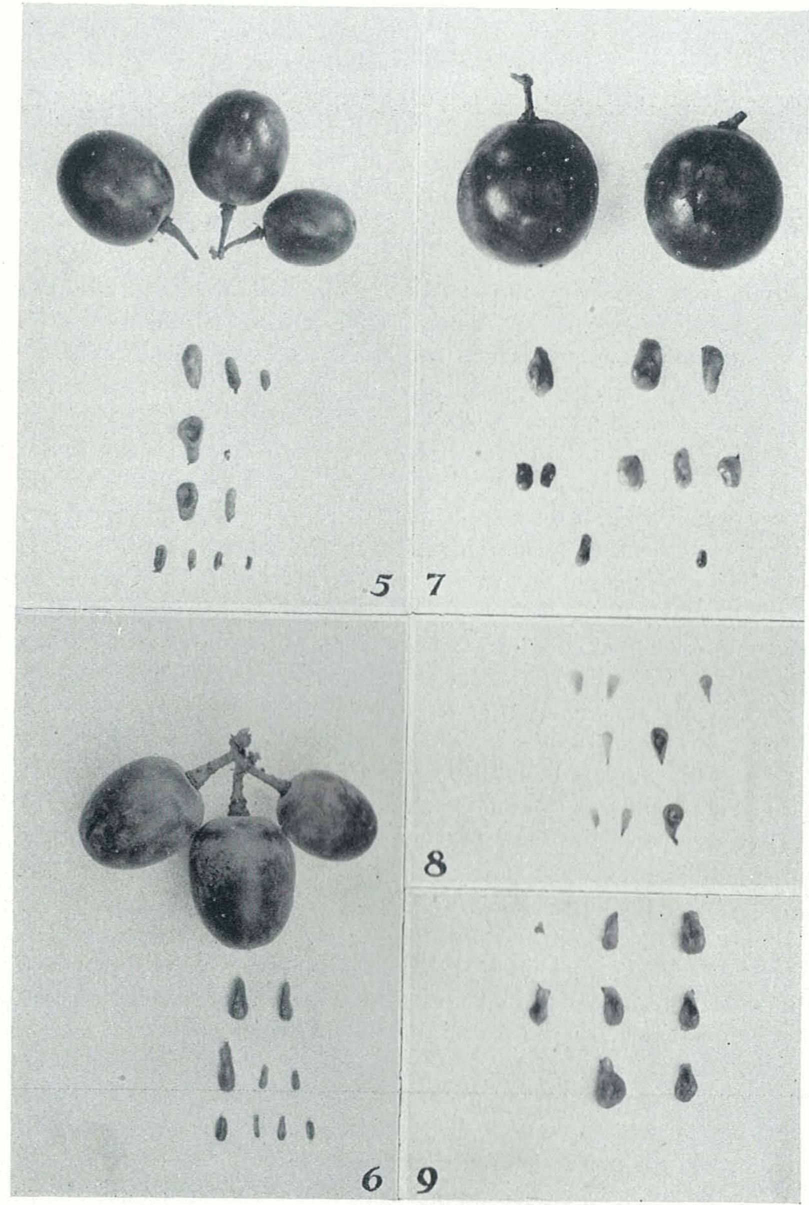
FIG. 5.—Fruit and seeds of the Black Monukka grape. The largest seeds are abnormal in shape and structure, soft, and somewhat shrunken. The size decreases until there are mere rudiments of aborted ovules. The remains of some of the ovules are not identified in dissection.

FIG. 6.—Fruit with stenospermic seeds and aborted ovules of Sultanina Rose.

FIG. 7.—Fruit and seeds of seedling No. 12583 which is rated as one of the best of the seedlings which bear stenospermocarpic fruits. The seeds are often of large size but they are soft and pulpy. The berries are of large size (for seedless grapes), red in color, and of excellent quality.

FIG. 8.—Typical stenospermic seeds of seedling No. 15194 which has for its immediate parentage Golden Muscat x No. 10918. The largest seeds are much shrivelled and are noticeably constricted at the apex. The smaller ones are almost white and devoid of any contents.

FIG. 9.—Stenospermic seeds of Stout Seedless (No. 10918) of which sections are shown in photomicrograph in figures 35, 36, 37, 42, 43, and 46.



FIGS. 5 TO 9.

For Sultanina Rose, the largest seeds in mature berries (Fig. 6) obtained on vines grown at the Experiment Station and at the New York Botanical Garden were soft, somewhat misshapen, and smaller than normal viable seeds of ordinary seeded varieties. The prepared sections of such seeds (Figs. 33, 40, and 41) usually show several layers of radial cells in the region of the greatest development of sclerenchyma, but the cells remain somewhat thin-walled and the layer is relatively thin and irregular in thickness. The configuration of this zone is also irregular and the lobing seen in cross-sections of normal seeds is usually almost lacking. The three layers of the inner integument are present and especially the cells of its innermost layer are quite normal in appearance. In some of the largest seeds immature embryos are present as small globular masses of cells which had ceased development before there was differentiation of cotyledons and radicle. The cells of such undeveloped embryos often contain nuclei and the cell contents appear to be normal and living even when the berry is fully ripe. In such seeds nucellar tissue is usually present as a lining of the enlarged embryo sac; near the chalaza the cells of the nucellus are enlarged and mostly devoid of contents; and about the median portion of the embryo sac the nucellus is a relatively thin zone of compressed and collapsed cells in which the contents have mostly disappeared. The interior of the embryo sac is mostly without cellular structure and the immature embryo lies in the micropylar end of this cavity with some granular material scattered about. In the later stages of the development the nucellus forms the outer wall of a sac which is probably filled with liquid or at least with non-cellular material. Either the endosperm tissue was not formed in the seeds examined or it had disintegrated.

In some of the larger seeds and in many of the seeds of medium size, there is no trace of either embryo or endosperm; the sclerenchyma is feebly developed; and often the inner integument is separated from the outer integument and collapsed upon the nucellus which together form an irregular-shaped strand that extends from the micropylar end to the chalaza. This collapse evidently takes place at some time after the embryo sac reaches its maximum size. In most of these smaller seed-like structures there is little differentiation and development of tissues and no appreciable nucellar tissue.

For the Black Monukka grape the conditions in the ovules are reported by Pearson to be very similar to those in Sultanina and Sultana. Some embryo sacs are abnormal but others appear to be normal at the time of anthesis; in nearly every ovary some ovules are entered by pollen tubes; and in many cases the endosperm nucleus soon starts a series of divisions. The processes of seed-coat formation break down, but some of the best-developed seeds may be more stony than are the aborted seeds of Sultanina. Occasionally, a seed with an embryo in some stage of development will be formed and Ross (cited by Pearson, 1933) reports that four such seeds obtained from Black Monukka germinated. The writer has made studies of the structure of the largest seeds in mature berries of this variety. In Fig. 5 is shown the range in size of mature fruits borne by vines grown at the Experiment Station and the appearance of the seeds which they contained. In Fig. 45 is shown the cross-section of one of the larger seeds obtained in the mature berries and is fairly representative of such seeds. There is almost no development of the innermost layer of the outer integument into sclerenchyma, but the outer layers develop into a considerable amount of pulpy tissue. The inner integument is present and its inner layer is quite normal as shown by the dark stain which it takes. The seed had enlarged somewhat, but the lobing is irregular and the nucellus had eventually collapsed and become a rather thin layer of fibrillar material as shown in the section. In no case for the seeds studied was an embryo or a well-developed endosperm found in a seed of Black Monukka, but as noted above such seeds have been observed.

The seedlessness in the grapes Sultanina, Sultanina Rose, and Black Monukka involves in part a complete failure in the development of ovules that are not fertilized and in part the abortion of embryos and seeds after fertilization. There are many ovules that have defective embryo sacs that can not function in fertilization, a condition also seen in seeded grapes. Berries which contain only remnants of undeveloped ovules are evidently the results of either vegetative parthenocarpy or stimulative parthenocarpy. The experimental evidence indicates that pollination is necessary before there is a good set of fruit and that vegetative parthenocarpy is not strong in these particular grapes. It is a fact, however, that berries may contain some partially developed, immature, and aborted seeds and the evidence indicates that fertilization may have occurred in the ovules. Possibly all ovaries are able to develop by stimulative parthenocarpy and that when fertilization is possible and does occur there is merely an additional stimulus which results in somewhat larger berries.

EXPLANATION OF FIGS. 10 TO 14

(All figures are natural size.)

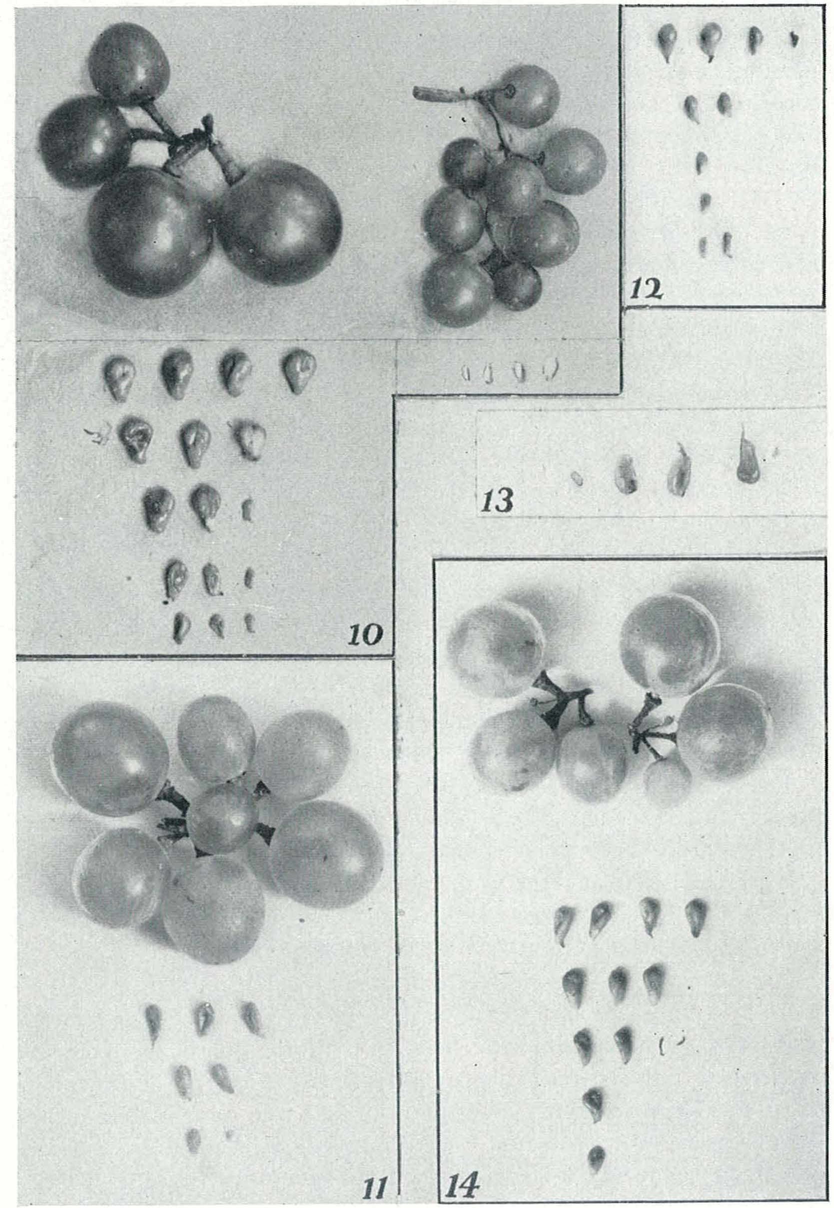
FIG. 10.—Fruits and seeds of seedling No. 12559. At left, typical stenospermocarpic fruits which form to self- and cross-pollination in the vineyard. Seeds from stenospermocarpic fruits; the largest are soft-gritty with partly developed endosperm and embryo. At right, fruits developed by vegetative parthenocarpy when pollination is prevented by emasculation and enclosure of flower cluster in a paper bag. Shows the size to which ovules may develop in parthenocarpic fruits.

FIG. 11.—Typical fruits and complement of seeds of seedling No. 17461 obtained from the cross Melton by Black Monukka. The largest seeds are noticeably undersized and they are soft and pulpy. The clusters are large and well filled and the fruit is excellent in quality.

FIG. 12.—Stenospermic seeds from five fruits showing the range of sizes for the seedling No. 17404. Evidently the aborted ovules are too small to be located by dissection.

FIG. 13.—Typical seeds from a berry of seedling No. 12551. The fleshy seeds are irregular in shape, somewhat gritty, and black areas of the inner tissue show in small areas on the surface.

FIG. 14.—Fruits and stenospermic seeds of seedling No. 17191, obtained from crossing seedling No. 8536 by Stout Seedless (No. 10918). The small berries are produced by parthenocarpy. The aborted ovules in all the berries are so rudimentary that they are usually not found in the ripe fruit.



FIGS. 10 TO 14.

TERMINOLOGY

The term parthenocarpy was applied by Noll (1902) to the development of seedless fruits without the occurrence of fertilization. The formation of what may be called seedless fruits, in which soft, pulpy and partly matured seeds develop from ovules in which fertilization has occurred, can not correctly be called parthenocarpy. Negrul (1934) calls this condition in grapes *parthenospermy*, but in a recent letter to the writer he states that he will not continue to use this term in this relation. Gardner, Bradford, and Hooker (1922) note that seedlessness in various fruits involves embryo abortion rather than merely parthenocarpy. There appears to be no single term in use that applies to this condition, which is well exemplified in certain grapes. The writer suggests the term *"stenospermy"* for the formation of aborted and partly formed seeds after there has been fertilization or at least entrance of pollen tubes into ovules, and for the formation of fruits in which there is stenospermy he suggests the term *"stenospermocarpy"*. If the formation of fruit depends on stenospermy, the stenospermocarpy is obligate.

In grapes the small rudiments that develop from ovules in many of the vegetatively parthenocarpic fruits and also from some of the ovules in stenospermocarpic fruits and in seeded fruits can not, it seems to the writer, properly be termed rudimentary seeds. In this bulletin they are usually called aborted ovules; and it is to be recognized that this condition may be either obligate or facultative.

The term *"phenospermy"* has been applied by Goodspeed (1915) to the formation of empty seeds "either with or without pollination" in parthenocarpic fruits of *Nicotiana*. Such seeds were numerous and some of them were as large as the normal seeds of the same plant, but seeds which contained traces of endosperm or which contained endosperm and embryo were not classed as phenospermic.

Possibly in *Vitis* the entirely empty seeds could be called phenospermic in contrast to the stenospermic seeds in which there are traces of embryo, of endosperm, or of both. In the formation of the fruits in grapes here called stenospermocarpic, pollination and the fertilization of at least some ovules are necessary, but as a rule some ovules in the same ovaries do not function in fertilization and abort fully or form only very rudimentary structures. Of the stenospermic seeds which appear to develop after a stimulus of pollination or fertilization the precise distinction of those that are entirely empty from those that

have traces of endosperm or embryo would be difficult and perhaps of little value.

CONCORD SEEDLESS GRAPE

The berries of this grape are smaller than those of the Concord from which it evidently arose as a bud sport. The largest seeds in the ripe berries are noticeably smaller than the normal seeds of Concord. They are soft or somewhat brittle and pale green with a conspicuous black area on the dorsal face of the seed (Fig. 15). The larger berries have from one to four of the larger seeds; the smaller berries usually contain smaller and more rudimentary seeds.

Some of the largest of the seeds contain an embryo and an endosperm in various stages of development. The fossettes may be rather well defined and the sclerenchyma is rather continuous but relatively thin and composed of few layers of cells (Fig. 16). The majority of the smaller seeds contain only a shrivelled and collapsed nucellus. A large blackened area develops in the tissues of the integuments and appears to involve the excessive formation of tannin. On the surface of the seed it usually covers an area below the position of the chalazal pad toward the outer lip of the integuments. Cross-sections (Fig. 16) of the seeds reveal, however, that the blackened tissue may extend around the embryo sac in the inner layers of the outer integument. Certain seedless grapes of the seedlings obtained by breeding have somewhat similar black blotches in the seeds.

Studies of the young ovules show that at least some of them are apparently normal in respect to integuments, nucellus, and embryo sacs. Sections of later stages show some ovules that are enlarging more rapidly than others. Ovules may reach considerable size but contain only traces of endosperm and embryo.

The stamens of this grape are erect, the anthers dehiscent, and the pollen is highly viable. Several clusters of flowers were emasculated while in bud and left enclosed in paper bags. No fruit developed which seems to indicate that pollination is necessary for the production of fruit by this grape.

The Concord Seedless grape has been used as a pollen parent in numerous crosses with seeded grapes. All the F_1 progeny have borne seeded fruit only. All of the F_2 generations that have been grown also bear seeded fruits.

The grape called "Hubbard Seedless" has been grown and studied by Mr. Fred E. Gladwin of the Vineyard Laboratory of this Station, who has stated to the writer that this grape is the same as the Concord Seedless.

APOGAMY AND POLYEMBRYONY IN GRAPES

APOGAMY

Apogamy, or the development of viable seeds *without fertilization*, is reported by Negrul (1934) to occur in certain grapes. He states that apogamy without even any pollination may occur in the varieties Taokvery, Tagoki, Aligoti, Plavai, and Muscat of Alexandria. This condition may well be termed *vegetative apogamy*. Negrul calls it *faculative apogamy* which seems to imply that a single ovule may develop in at least two different ways.

Five varieties having imperfect flowers (Nimrang, Taokvery, Katta-Kourgan, Doroi-rose, and Tomaiosa) are reported to show *induced apogamy*. According to Negrul's results, when the flowers of these grapes are emasculated and left unpollinated they either develop no fruit or the berries are seedless. When allowed to self-pollinate, they produce at least some berries with viable seeds. It is assumed that the pollen in these varieties is not viable or if it is viable that there is "self-sterility" but that the pollen does exert some sort of influence on the pistils which induces the apogamous development of embryos. In other words, when a plant which has imperfect flowers produces viable seeds or seeds with endosperm after self-pollination, it is considered that fertilization did not occur. In the self-pollination tests made at the Experiment Station certain plants having flowers with the reflexed type of stamens have produced a few berries with seeds, but in these cases it was not certain that some accidental cross-pollination did not occur which resulted in fertilization.

POLYEMBRYONY

Negrul (1934) has reported that each seed of the variety Nimrang produces two seedlings and he also classes this variety as producing apogamous seeds which are induced by pollination. No cases of polyembryony have been observed in the grapes studied at Geneva.

PARTIAL VARIATION IN THE CHARACTER OF BERRIES

There are conspicuous cases in grapes in which the clusters of fruit are mixtures of two or more types of berries in respect to the seedless and the seeded characters. There is what de Vries (1901, page 37)

called "partial variability" among the homologous parts, which in this case are the fruits and the ovules of the individual. In the above discussions of seedless grapes it has been noted that at least three main types in the development of ovules are to be recognized and that frequently two types of fruit are produced in respect to the behavior and development of the entire complement of ovules. There are several rather definite conditions to be recognized in the partial variations seen in the fruits of grapes.

FACULTATIVE PARTHENOCARPY AND PARTIAL VARIATIONS

It has long been known that certain grapes produce only seedless fruit of the Corinth type when there is no pollination but when pollination and fertilization occur seeded fruits develop. When some of the flowers in a cluster are not properly pollinated both kinds of fruit are produced in the same cluster (Fig. 3). The flowers are alike and of either the perfect hermaphrodite type or the type with reflexed stamens, and the ovaries are alike in that they are all able to produce fruit with one or more seeds. The relative number of seedless and seeded berries vary from cluster to cluster and from year to year depending on the irregularities of pollination and either type of fruit may be produced experimentally by preventing or providing pollination. Thus it is to be recognized that the two different characteristics, *viz.*, (1) seededness and (2) vegetative parthenocarpy, may exist in the ovules and ovaries of one individual vine in which case the parthenocarpy is facultative.

This situation is most readily recognized when it exists in a plant that has the imperfect type of flower, such as the vinifera grape Hunisa, the Brighton, and certain seedlings. These, it is believed, have no good pollen grains that are viable and hence the flowers are not able to effect proper self- or close-fertilization. When the male-sterile flowers are properly and fully cross-pollinated and fertilization results, their ovaries develop into large-sized seeded berries; when they are not pollinated, the berries develop but they are small-sized and seedless.

Facultative vegetative parthenocarpy may also exist in seeded grapes that have perfect hermaphrodite flowers; but ordinarily, such plants are self-pollinating and the berries are usually seeded. Only when conditions result in faulty pollination and small seedless berries and large-seeded berries occur in the same cluster do these plants exhibit parthenocarpy.

Facultative stimulative parthenocarpy may be present in grapes that

EXPLANATION OF FIGS. 15 TO 20

(Figures 16 to 19, inclusive, are of free-hand sections.)

FIG. 15.—Fruits and seeds of Concord Seedless grape. The seeds are soft and pulpy and there are large conspicuous black areas on the surface of the dorsal side toward the apex, even on seeds of small size.

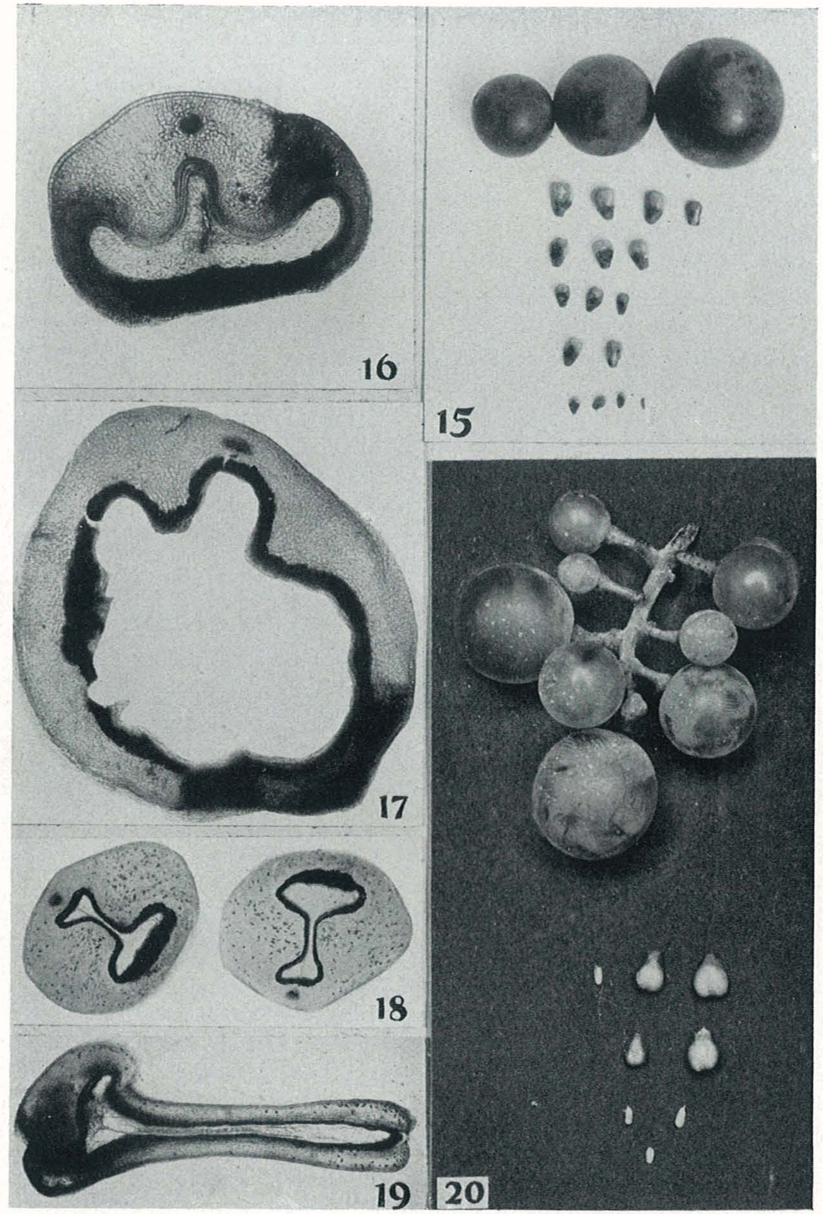
FIG. 16.—Median cross-section of a seed from ripe berry of the Concord Seedless grape. The fossettes are partly formed, sclerenchyma is feebly developed and the nucellus fills the inside except for the cavity of the embryo sac. The blackened area in which there is an excessive development of tannin extends thruout a considerable portion of the integuments and also in the vascular bundle. × 38.

FIG. 17.—Cross-section of a large stenospermic seed of the Stout Seedless grape. There is a zone of excessive tannin in the inner tissues of the outer integument next to the sclerenchyma, and this extends to the surface in a portion of the dorsal side. × 20.

FIG. 18.—Cross-section of a stenospermic seed of seedling No. 14076 showing the zone of blackened tissue and the relatively large amount of pulpy tissue. × 9.

FIG. 19.—Longitudinal section thru a stenospermic seed of seedling No. 12614. Shows the fibrous remains of the nucellus and a much shrivelled and aborted condition. × 9.

FIG. 20.—Illustrating partial variation in fruits involving parthenocarpy which is obligate for the ovules which become seedless fruits. Fruit and seeds of a seedling No. 9130 used in breeding. The small berries have only aborted ovules and their parthenocarpy is obligate. The large berries contain (a) aborted ovules, (b) soft undersized aborted seeds, and (c) larger semi-hard seeds of good size and shape, some of which will germinate.



FIGS. 15 TO 20.

are able to produce a seeded fruit from every ovary, but this will not be expressed unless there are certain stimulative pollinations which do not effect fertilization.

Facultative vegetative parthenocarpy may also exist in connection with stenospermocarpy. This has been observed and determined experimentally for certain of the seedlings obtained in breeding for seedlessness (Fig. 10). In this case the vine has perfect flowers and self-pollination results in berries that bear stenospermic seeds; but emasculation in the bud and prevention of pollination results in the development of only small seedless fruits of the Corinth type. Usually the clusters of fruit on such plants have some small parthenocarpic berries intermingled with stenospermocarpic ones.

OBLIGATE PARTHENOCARPY AND PARTIAL VARIATIONS

When seeded berries and parthenocarpic berries are developed in the same clusters and the ovaries which give the latter can develop in *no other way*, the parthenocarpy of those berries is evidently obligate. In such a case the most adequate pollinations which provide opportunity for the fertilization of every ovule do not change the nature or the number of the parthenocarpic fruits.

This condition has been observed in certain seedlings of which two were studied especially. The flowers of these vines have well-formed pistils and erect stamens and about 50 per cent of the pollen germinates on sugar-agar media. Some berries were of small size and completely seedless (Fig. 20) and the larger berries contained one, two, or rarely three fairly well-developed seeds some of which were viable. Pollination did not reduce the number of the seedless fruits.

The conditions in such cases seem to be as follows: All ovaries are able to develop as parthenocarpic fruits; when an ovary contains only ovules in which there is embryo-sac abortion, it becomes a seedless fruit; ovaries which contain one or more ovules which can function in fertilization may mature as seeded fruits. The parthenocarpy in such cases may be either vegetative or stimulative. If it is stimulative, pollination is necessary for the production of the seedless fruits.

According to Harmon and Snyder (1936) mixtures of seeded fruits and seedless fruits in clusters of the Panariti grape may be a result of mutation in which ovules which are able to develop into viable seeds arise from the usual type of Panariti in which there is abortion of all ovules in the ovaries.

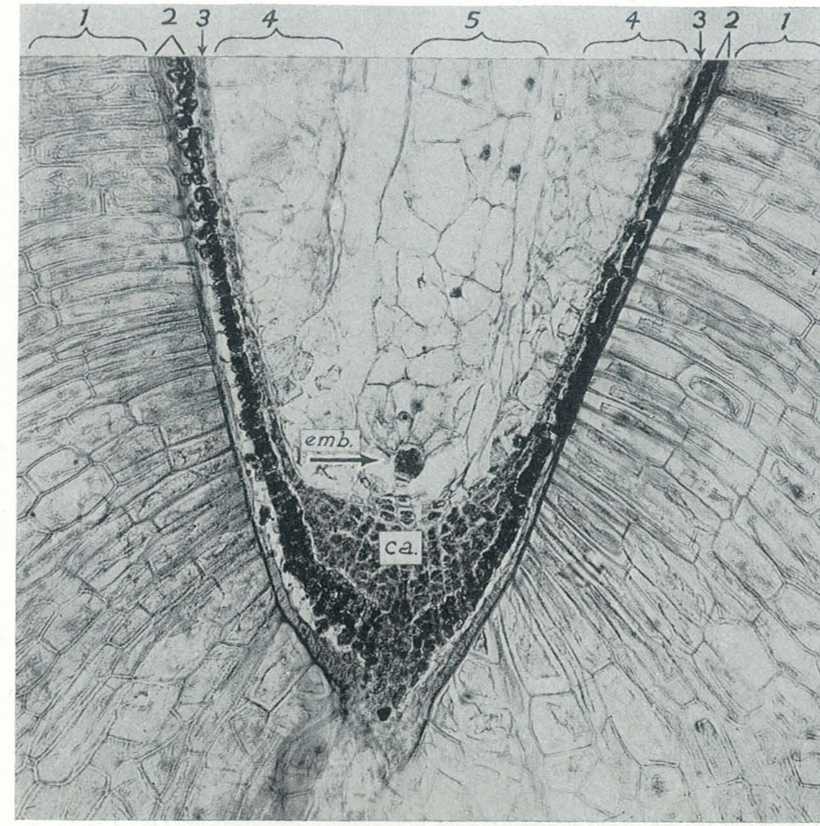


FIG. 21.—Photomicrograph showing in longitudinal section a portion of a normally developing seed of the Concord grape about 2 weeks after fertilization. 1, *sclerenchyma* which is already rather fully formed in amount and in the size of cells but walls not fully thickened. 2, *inner integument;* inner layer composed of large nearly isodiametric cells that stain densely black and more than one cell thick in the region of the micropyle; the outer two layers are much compressed except near the micropyle. 3, the *inner layer of the nucellus* is clearly differentiated and at the micropylar end it forms several layers of the *"calotta"* (ca) in front of the embryo sac. 4, the body of the *nucellus* consists of rather large thin-walled cells. 5, cylinder of *endosperm* extending from proembryo to the chalaza which is not shown here. The *proembryo* (emb.) here shown consists of several clearly defined cells. This illustrates the extent of the rapid developments in the integuments which occur immediately after fertilization while the embryo and the endosperm develop slowly. × 170.

EXPLANATION OF FIGS. 22 TO 28

FIG. 22.—Cross-section of ovary of Stout Seedless (seedling No. 10918) about time of anthesis. All four ovules are nearly alike in size and general development. Some can function in fertilization and become stenospermic seeds, others cannot so function and these will abort as ovules. × 25.

FIG. 23.—Cross-section of two ovules of Stout Seedless about 2 weeks after anthesis. The ovules have enlarged since the time of anthesis. The sclerenchyma is developing feebly; the nucellus is much expanded; endosperm is either absent or feebly formed at other levels. × 25.

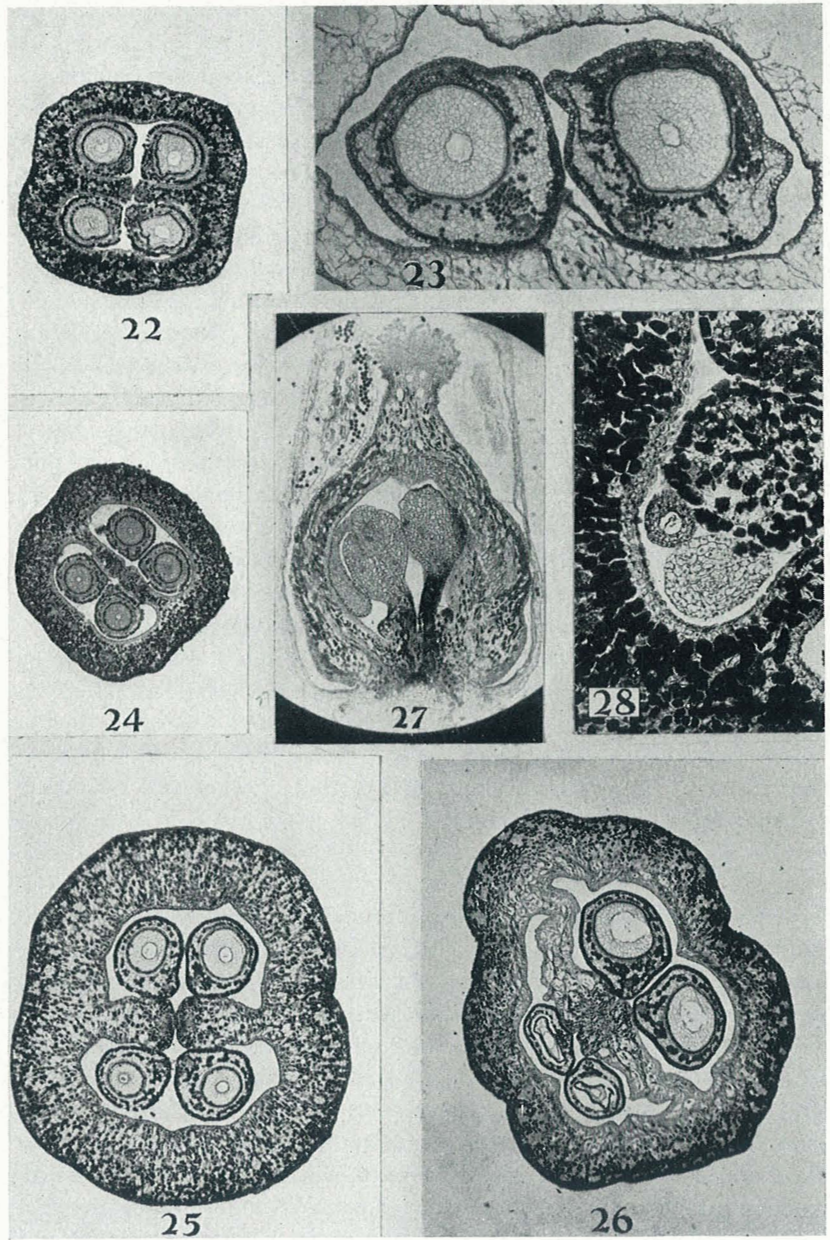
FIG. 24.—Cross-section of a typical ovary of the Delaware grape at time when the flower opens. All four ovules are very fully formed and quite alike but it is certain that some are unable to function in fertilization. Reduction of seed is very noticeable and constant in this grape and it is not changed when the most complete pollinations are provided. Ovule abortions occur during the late stages in the development of the embryo sac. × 25.

FIG. 25.—Cross-section of an ovary of Sultanina Rose shortly after anthesis showing that the four ovules are present and rather fully formed as to size and general structure. × 25.

FIG. 26.—Cross-section of an ovary of Black Monukka about 10 days after pollination. Two of the ovules are aborting; two are enlarging and would probably become stenospermic seeds. × 25.

FIG. 27.—Longitudinal section thru pistil of White Corinth showing a typical abnormal ovule. There is only one integument and the nucellus protrudes as a cylinder of tissue which bends downward against the wall of the ovary. × 20.

FIG. 28.—A portion of a cross-section of an ovary of the White Corinth grape showing cavity in the protruded nucellus in which at least some cells of an embryo sac may form. × 35.



FIGS. 22 TO 28.

MIXTURES OF PARTHENOCARPIC AND STENOSPERMOCARPIC FRUITS

It has already been noted in the discussion of the grapes Sultanina, Sultanina Rose, and Black Monukka that two types of berries are usually present in the clusters of fruit. There are fruits that are entirely seedless and there are those that have some stenospermic seeds (Fig. 14).

That the presence of small entirely seedless fruits along with stenospermocarpic fruits in a cluster may involve somatic mutation has been suggested by Olmo (1934 A). In one vineyard of Sultanina grapes five vines were found which bore mostly small-sized spherical fruit in which the partly developed or stenospermic seeds were absent. Usually, the clusters of fruit were poorly filled. Pollination with viable pollen did not change the nature of the berries, but occasionally a larger berry of the type which has abortive seeds was formed. In fact, about 3 per cent of the berries were of this type. Other vines bearing the same kind of fruit were found in other vineyards. According to Olmo there is in this condition mutation to an obligate vegetative parthenocarpy. If this is the case the smaller fruits should be developed without any pollination and a test for this would reveal whether there has been a mutation in respect to parthenocarpy or a change in the amount of embryo-sac abortion with the parthenocarpy remaining the same.

At any rate the mixture of smaller, entirely seedless berries with larger berries that have either some normal seeds or stenospermic seeds is very frequent. The different types of development in ovules are also seen in individual berries.

FURTHER COMBINATIONS IN PARTIAL VARIATION

Certain cases of partial variations in the character of ovules, ovaries, and fruits may involve apogamy, according to Negrul (1934). Negrul reports that the grape Taokvery, which has the imperfect hermaphrodite type of flowers, develops berries with empty seeds when there is no pollination and that when there is self-pollination the berries may have empty seeds, or seeds with an endosperm, or seeds with an embryo. Negrul believes that the pollen of the imperfect hermaphrodite flowers can not function in fertilization but can exert a stimulative influence, hence he lists the variety Taokvery as exhibiting autonomic parthenocarpy, facultative apogamy, and induced apogamy.

The variety Muscat of Alexandria has perfect hermaphrodite flowers, and according to Negrul, it will produce *after emasculation* and *without any pollination whatsoever* (1) some berries with no seeds, (2) some berries with empty seeds, (3) some berries with seeds which have an endosperm, and (4) some berries with seeds which contain embryos. Self-pollination also gives the same range of developments. It has long been known that this grape is one of the varieties which will produce a large proportion of parthenocarpic fruits when there is imperfect pollination—a condition called "Millerandage" in France (Bioletti, 1921). According to Negrul, there is in this grape (1) facultative parthenocarpy, both of the vegetative and the stimulative types; (2) abortion of some of the embryos that arise from apogamy; and (3) maturity of some embryos by apogamy. Also, this grape can be used as a seed parent in cross-breeding (especially Snyder, 1935) and it is certain that fertilization may occur at least in certain ovules. It is not clear to what extent these different developments are possible or facultative for individual ovules or to what extent they are obligate.

FRUIT AND SEED DEVELOPMENT IN SEEDLINGS OBTAINED IN BREEDING FOR SEEDLESSNESS

Numerous seedlings have already been grown to the fruiting age which have various seedless grapes as a pollen parent and of these several progenies have two generations of seedless plants as pollen parents. (See earlier reports by Stout, 1928; 1933.) These have been studied as to the type of flower, the character of the fruit, and the extent to which seeds develop in the mature berries. Tests for the presence of apogamy and for induced parthenocarpy by emasculation and controlled pollination have thus far been made for only a few plants. The seedling vines have been subjected to self- and cross-pollination under vineyard conditions, controlled self- and cross-pollinations have been made for some plants which are being used in further breeding, and studies have been made to ascertain the degrees of seedlessness that exist when there is adequate pollination.

A large proportion of seedlings bear seeded fruits with at least one seed of the normal type in each berry. Some of the seedlings bear fruits all of which are fully seedless to the degree that the largest seeds are only small and much aborted. Between these two extremes there are many intermediates in respect to the maximum development which any of the seeds make. The clusters of fruit produced by various seedlings also display partial variation; that is, there are mixtures of berries of different sizes which differ in the complement of seeds and in the grades of their development.

SEEDLINGS WHICH BEAR SEEDED FRUIT

The berries of grapes may be classed as seeded when they contain one or more seeds that are of such size and development in respect to the hardness of the outer integument that they are at least noticeable if not normal.

FRUIT WITH NORMAL SEEDS

In one class of seedlings all berries, with rare exceptions, contain at least one apparently normal seed (Fig. 2). The formation of fruit seems to be dependent on the development of seed and as no cases of apogamy have been observed in these plants it may be considered that pollination and fertilization are necessary for the production of seed. The number of seeds per berry, judged by the number present in the greatest number of berries, differs for various plants and may be one, two, or three. Possibly plants may be found in which this number is four but thus far none has been noted. As a rule, when the seed complement in a berry is less than four, the other ovules abort to mere traces quite as shown in Fig. 2, but in certain cases stenospermic seeds are present along with normal seeds and aborted ovules.

A second class of seedlings produces seeded berries and seedless berries in the same cluster and such plants possess parthenocarpy or stenospermocarpy. In several cases of facultative vegetative parthenocarpy the clusters were frequently mixtures of large-seeded berries and small seedless berries (Figs. 3 and 20), especially when the plant had flowers with reflexed stamens.

EMPTY SEEDEDNESS

Seeds which are hard and which appear to be well developed may be abnormal in respect to their contents. The mature berries produced by one seedling which was studied contained large hard seeds, but the majority of them had no endosperm or nucellus, or only traces of them, and the sclerenchyma extended inward and encroached on the space normally filled with endosperm. In a few of the seeds an embryo was found, but it was not fully developed. It is possible that for some seedlings all of the best-developed seeds are empty or not viable, altho they are of normal size and shape.

SEEDS WHICH CRUSH

Seeds which crush readily between the teeth characterize certain seedlings. Such seeds may appear to be normal as to size, form, and contents.

BRITTLE SEEDS

Brittle seeds or seeds which break quite easily into fragments, are produced by certain seedlings. Frequently there are also seeds that are somewhat shrivelled or greenish in color, or even decidedly soft and of the type classed as stenospermic.

PAPERY SEEDS

For certain seedlings the largest seeds produced are of large size, but the integuments are tough, "papery", and flexible. The seeds are mostly empty and the walls can be compressed. The tissue which becomes sclerenchmya in normal seeds is in these seeds flexible and tough.

EXTREME PARTIAL VARIATIONS

Extreme partial variations occur in which aborted ovules and stenospermic seeds are present along with one or more grades of development listed above. The distribution of these may be such that some berries contain only fully aborted ovules, others have both aborted ovules and stenospermic seeds, and others have the most fully developed seeds, together with one or more of the lower classes. In some cases there is evidence of a parthenocarpic development of the seedless fruits.

SEEDLINGS WHICH BEAR FRUITS WITH STENOSPERMIC SEEDS

The seeds which are here classed as stenospermic are only partly developed in some one or more features. They are often misshapen or constricted at the micropylar end. Usually, they are greenish in color and pulpy or soft in which case there is feeble development of sclerenchyma but often there is an excessive development of the rest of the outer integument. Many stenospermic seeds contain nucellus, embryo, and endosperm, either alone or together, in various degrees of relative development (Figs. 1 to 14).

Of the seedlings which have already fruited, over 80 bear fruit in which the most fully developed seeds are to be classed as stenospermic; but in respect to the maximum development of seeds and the propor-

EXPLANATION OF FIGS. 29 TO 37

FIG. 29.—From a seedling (No. 12570) which produces stenospermocarpic fruit. The embryo sac is apparently normal and able to function in fertilization. The inner integument protrudes noticeably beyond the tip of the outer integument which is an abnormal feature frequent in certain types of seedless grapes. × 50.

FIG. 30.—Also of seedling No. 12570. Longitudinal section showing course of a pollen tube thru the calotta. The pollen tube is coiled and enlarged in the micropyle where it first comes in contact with the calotta; its course thru the calotta is somewhat sinuous; and the end of the tube is in contact with the egg apparatus. The outlines of the egg and synergids are faintly indicated but their nuclei are at a slightly different level. The dark-colored inner layer of the inner integument is well shown and it is fully differentiated at the time of fertilization. × 180.

FIG. 31.—From seedling No. 12600. Cross-section at apex of a stenospermic seed from an almost mature fruit. Shows some sclerenchyma, the fully differentiated layers of the inner integument, considerable nucellus, and the cavity of the embryo sac with a small compact ball of cells of the undeveloped embryo. × 25.

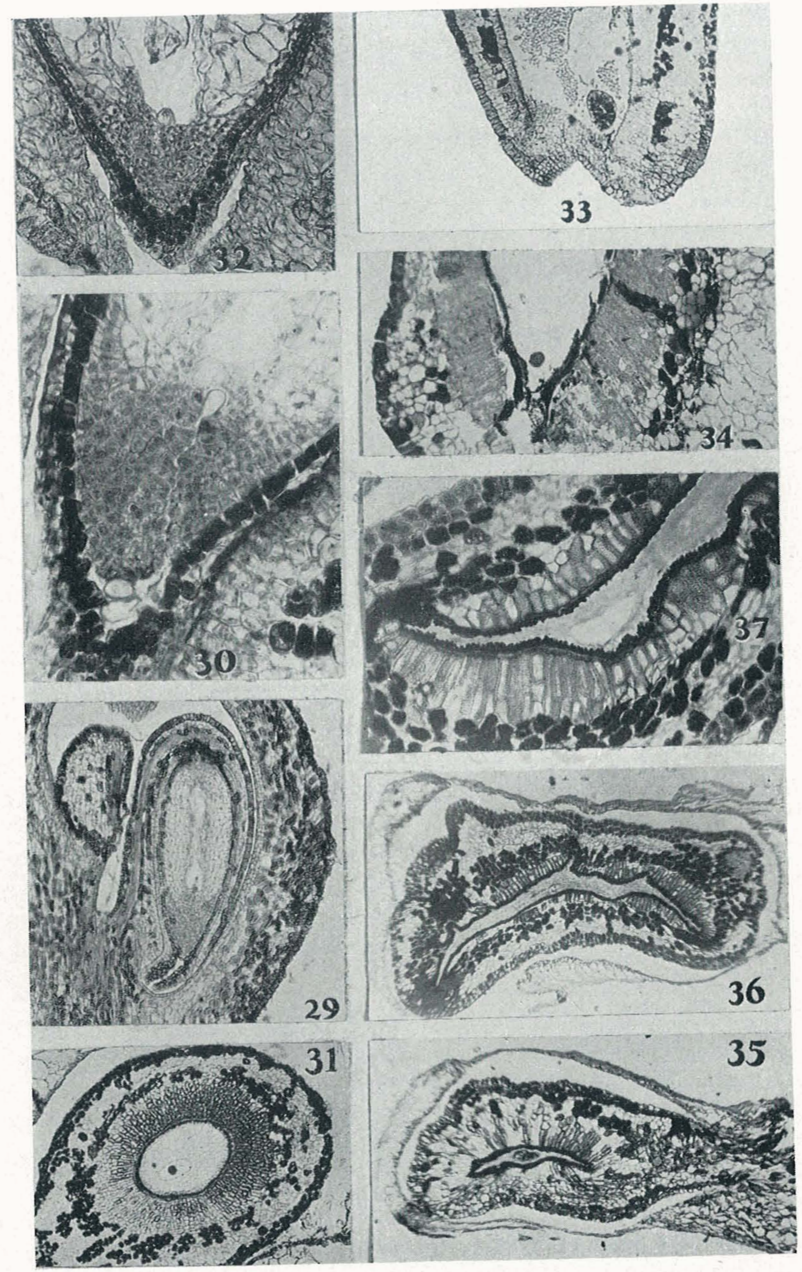
FIG. 32.—From Stout Seedless (No. 10918). Apex of ovule about 4 weeks after time of pollination. Shows a rather large fertilized egg lying against the inside of the calotta. Under the microscope with high magnifications it is seen that this is a group of several cells with thin separating walls. × 120.

FIG. 33.—Apex of a stenospermic seed of the Sultanina Rose grape from a mature berry showing ball of tissue comprising the undeveloped embryo, and granular material surrounding it. × 35.

FIG. 34.—Portion of the apex of a stenospermic seed from a mature berry of the Sultanina grape showing especially the ball of tissue comprising the aborted or undeveloped embryo. × 40.

FIGS. 35, 36, and 37 are cross-sections of a medium-sized stenospermic seed from a ripe berry of the Stout Seedless grape. Fig. 35 is at the apex (× 30) and shows the aborted embryo surrounded by fibrous remnants of nucellus. Fig. 36 (× 30) is a medium section of the same seed showing the collapsed and fibrous remains of the nucellus, the scant development of sclerenchyma, the distribution of tannin cells in the pulp, and the irregular shape of the entire structure.

FIG. 37.—A portion of Fig. 36 more enlarged. Shows the amorphous nucellus, absence of endosperm, the black compact cells of the inner layer of the inner integument, the relatively thin tissue of the sclerenchyma, the cells of which are somewhat thickened. × 80.



FIGS. 29 TO 37.

tion of such seeds in the berries, there is much variation among the seedlings.

GRITTY STENOSPERMIC SEEDS

The largest seeds in the largest fruits (Fig. 10) of one seedling (No. 12,559) obtained from a cross between seedling No. 8,536 (Goff × Iona) × Sultanina) are of nearly normal size and shape, but they are green in color and soft except for some grittiness which is noted when the berries are masticated. Endosperm and traces of embryo may be found in a few of them, but others are almost empty and collapsed and on some there is a black area in the region of the chalaza. The berries of smaller size contain smaller and more rudimentary seeds that are often fully soft. Clusters of flowers in bud were emasculated and enclosed in paper bags and these produced well-filled bunches of small-sized seedless berries, showing that the plant also has the character of vegetative parthenocarpy. The undeveloped ovules in these parthenocarpic fruits are small empty sacs quite like the aborted ovules in the stenospermocarpic fruits (Fig. 10).

Another seedling (No. 18,110; originating from a cross between seedling No. 10,902 [No. 8,396 (Delaware × Goff) × Concord Seedless] × Sultanina) which has gritty seeds (Figs. 4 and 39) was studied. The berries contain from one to four rather plump seeds that are soft-gritty. A cross-section of one of the seeds is shown in Fig. 39. The sclerenchyma is present but forms a much thinner layer than in normal seeds and outside of this there is the excessive development of soft tissue characteristic of stenospermic seeds. Endosperm is well developed and an embryo is often present, but the outline of the sclerenchyma and of the outside of the seed is rounded with little evidence of fossettes. Nearly every berry has at least one gritty seed, and when there are less than four such seeds, the other ovules are much aborted.

SOFT OR ONLY SLIGHTLY GRITTY STENOSPERMIC SEEDS

For various seedlings the largest and most completely developed seeds are either soft or only slightly gritty. In this group are included the seedlings that are considered to be most promising for culture. In respect to seedlessness some of them are very nearly like the Sultanina and Sultanina Rose. In respect to the size and shape of the stenospermic seeds, the extent of the development of endosperm and embryo, and the proportions of completely aborted ovules, there is considerable variation. Several seedlings may be cited as typical of this class.

Seedling No. 12,597.—For this plant which has the same origin as No. 12,559, nearly all berries have at least one large-sized seed which contains an embryo and endosperm in advanced stages of development, but the seeds are soft or merely firm and slightly brittle. No doubt under special culture many of the embryos would germinate. In this plant the abnormal developments appear to be confined chiefly to the outer integument.

Seedling No. 12,551.—At least some of the largest seeds (Fig. 13) in the mature fruits of this seedling (parentage same as No. 12,559) contain both endosperm and embryo; the sclerenchyma of the seed coat is poorly developed; the mature seeds are soft, or slightly brittle, and green in color with conspicuous black areas showing on the surface of some of them. Numerous of the more rudimentary seeds contain only a shrivelled nucellus and are practically empty.

Seedling No. 12,572.—This grape, which has the same origin as No. 12,559, produces no hard and fully normal seeds. The largest seeds in mature berries are yellowish green in color and may contain an embryo and endosperm in various degrees of relative development. Special study was made of the condition in the ovules about six weeks after anthesis at the time when the sclerenchyma in normal seeds is rather fully formed. In Fig. 44 is shown a median cross section of such an ovule. The sclerenchyma is feebly developed and only at a few points does it consist of more than one layer of cells. The inner integument is normal in appearance, which is a condition quite general for the larger stenospermic seeds of seedless grapes. The outer layer of the nucellus is clearly differentiated as a single layer of compact cells. The endosperm fills the space within the nucellus. The outer portion of the outer integument is strongly developed as fleshy tissue with a considerable number of tannin cells in the inner portion.

Seedling No. 15,302.—This seedling was obtained by crossing Ontario with Sultanina. The clusters of fruit are large and well filled. The berries are amber in color, meaty, vinous, and of excellent flavor and quality. The aborted seeds range from one to six per berry; they are rather large, but are green and soft or slightly gritty. The smallest berries have one or two stenospermic seeds quite like the seeds in the larger berries. In the mature berries which were examined no rudiments of ovules were found or identified. All the seeds were quite alike in appearance.

Seedling No. 10,918.—This grape was named Stout Seedless (Howe, 1929), and was obtained by crossing seedling No. 9,135

EXPLANATION OF FIGS. 38 TO 46

(All of these figures are magnified about × 25 except Fig. 43 which is × 75. Fig. 39 is of a hand section, the others are of microtome sections.)

FIG. 38.—Cross-section near the middle of a normally developing seed, and above level of the embryo, about 4 weeks after pollination. The sclerenchyma is very fully formed; the outer pulpy tissue is relatively thin and contains numerous tannin cells; and the shape of the mature seed is already determined. The endosperm is developing and the nucellus is being absorbed. The outer layer of the nucellus is shown against the inner layer of cells of the inner integument which appears as a conspicuous black layer.

FIG. 39.—Median cross-section of a gritty stenospermic seed from a ripe berry. Several layers of sclerenchyma cells surround a well-developed endosperm; the outer pulpy tissue is somewhat excessive in amount; fossettes are not formed and the seed is nearly circular in outline.

FIG. 40.—Longitudinal section of a stenospermic seed from a ripe berry of the Sultanina Rose grape. Shows the feeble development of sclerenchyma. The inner integument is collapsed except at the chalaza. Neither endosperm nor embryo is present.

FIG. 41.—Median cross-section of a stenospermic seed almost identical to that shown in Fig. 40. Shows zone of tannin cells surrounding the thin zone of sclerenchyma and the dark-colored layer of the inner integument which has collapsed upon and with the shrivelled nucellus.

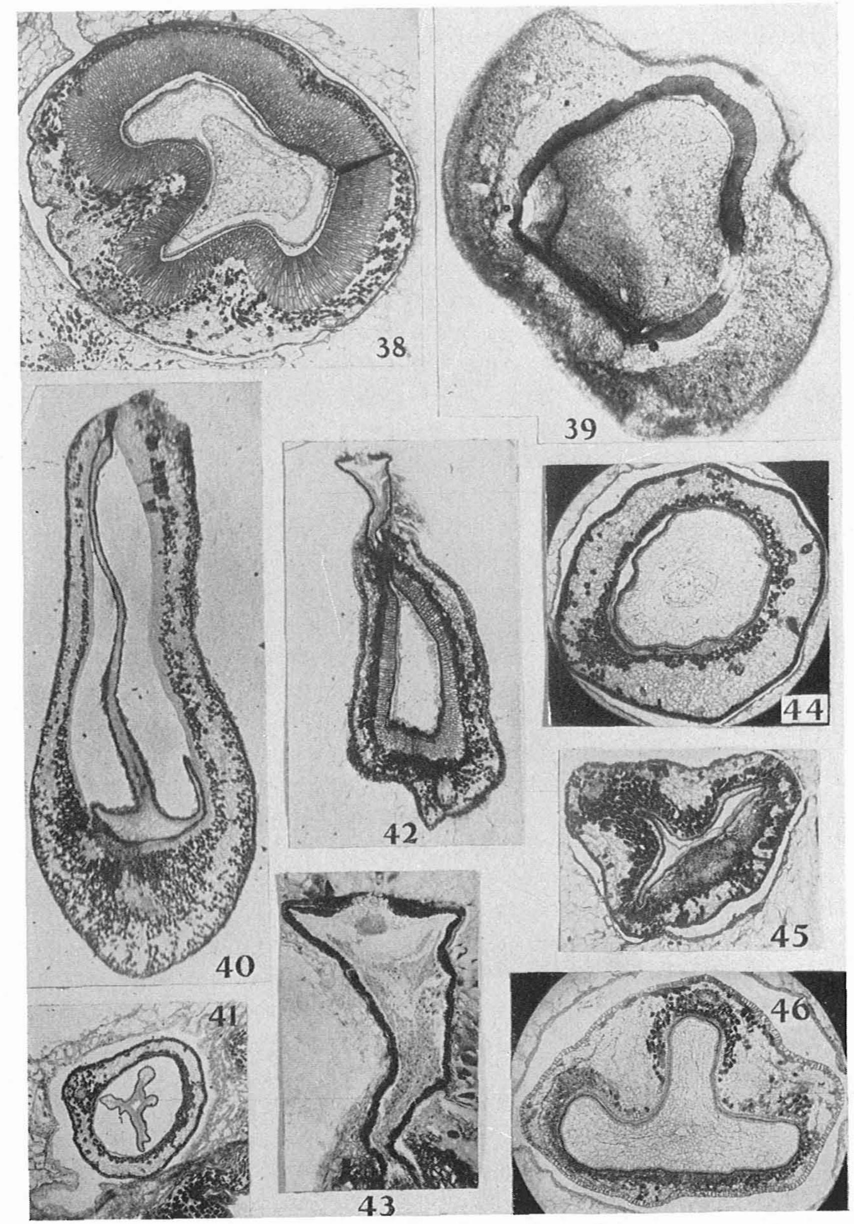
FIG. 42.—Longitudinal section of a medium-sized seed from a ripe berry of the Stout Seedless grape. There is considerable sclerenchyma surrounded by a rather dense zone of tannin cells in the pulpy tissue. The inner integument protrudes far beyond the lip of the outer integument. Cells, most of which are endosperm, are scattered in the main cavity.

FIG. 43.—Shows an enlarged view of the protrusion seen in Fig. 42. The three layers of the inner integument are involved in the protrusion. A ball-shaped mass of cells constitute a living embryo but its parts are not developed nor differentiated. Some remnants of the nucellus are somewhat mingled with endosperm cells.

FIG. 44.—Median cross-section of a developing stenospermic seed of seedling No. 12572 about 6 weeks after pollination. The sclerenchyma is feebly developed; the layers of the inner integument are fully differentiated; nucellar tissue is normal and conspicuous in amount; in the center of the ovule is a cylinder of endosperm. The seed is cylindrical in the outline of the cross-section.

FIG. 45.—Median cross-section of a typical medium-size seed of Black Monukka from a ripe berry. Only fibrous traces of the nucellus are seen; there is no endosperm; the sclerenchyma is feebly formed.

FIG. 46.—Median cross-section of a developing seed of the Stout Seedless grape (which has stenospermocarpic fruits) about 4 weeks after pollination. The sclerenchyma is composed of a thin layer mostly one cell in thickness and absent in areas along the dorsal side. The fossettes have appeared. There is no endosperm tissue. The tissues of the nucellus fill the interior of the ovule except for a small cavity in which the embryo sac was located.



FIGS. 38 TO 46.

(Triumph × Dutchess) × Sultanina Rose. The seed parent exhibits partial variation in its fruits with some that are seedless and some that have one or two brittle seeds, some of which are viable. The pollen parent has seedless fruit of the stenospermocarpic type and the fruits of this seedling are also to be thus classed. Since this plant was the first of the seedlings obtained in this breeding project which produces stenospermocarpic fruits, special study was made of the development of the fruits and the seeds and of the requirements for setting fruit.

The pollen produced by seedling No. 10,918 is highly viable and pollination seems to be necessary for the setting of fruit. Clusters of flowers that were emasculated in the bud and enclosed in paper sacks have all failed to produce fruit. Controlled self-pollination has given well-filled clusters of berries.

The berries usually contain from one to three rather large pulpy and soft or slightly brittle stenospermic seeds of irregular shapes (Fig. 9). The size of the seed-like structures diminishes until there are some that are mere flakes. Usually, there are two cells in an ovary, but there may be three with at least five ovules. At the time of anthesis it is the rule that the ovules are all quite alike in appearance and there does not appear to be any noticeable abortions of ovules previous to this time (Figs. 22 and 23). Embryo sacs in some stage of development appear to be present in the ovules and some of them are apparently normal. Pollen tubes were observed passing thru the nucellar cap and also in contact with an egg.

Material collected about one month after pollination showed in certain ovules a small globular embryo composed of a few cells, a narrow central cylinder of rather few endosperm cells, and much nucellar tissue. In Fig. 46 is shown a median cross-section of an ovule at this time which was presumably destined to become one of the larger seeds. The sclerenchyma is almost absent and is mostly composed of a single layer of rather thin-walled cells, the nucellar tissue fills most of the space within, and the endosperm is lacking or composed of only a few scattered nuclei.

In mature fruits many of the larger seeds are empty or nearly so; others have some endosperm with or without an embryo in various stages of development. In some seeds endosperm and embryo are present and fairly well developed.

The conditions in many shrivelled seeds are shown in Figs. 35, 36 and 37. In median cross-section it is seen that the nucellus has collapsed and that the endosperm has either not developed or has

disintegrated. The more enlarged view in Fig. 37 shows the amorphous condition of the nucellus, the well-defined inner layer of the inner integument, and the few cells of the sclerenchyma. A cross-section near the micropylar end shows a small shrivelled embryo (Fig. 35).

Black areas develop in the tissues and show on the surface of various seeds of this grape. A study of the cross-sections of such seeds shows that the dense accumulation, presumably of tannin, forms a compact zone in the pulpy tissue about the seed cavity in the region (Figs. 23, 41, 44, and 45) in which many tannin cells are normally found, and that extensions from this zone may reach the surface (Fig. 17). But the spots on the surface are not as large or as frequent as in the seeds of the Concord Seedless (Fig. 16).

One type of abnormality observed in this grape is the extension of the inner integument far beyond the outer one, as shown in Figs. 42 and 43. The seed shown is from a ripe berry; it is undersized, misshapen, and poorly developed, and has some sclerenchyma and considerable endosperm tissue. The inner integument which protrudes thru the micropyle in the form of a sack may readily be traced by its dark-staining inner layer. Endosperm tissue fills most of this protruded part. At the extreme apex there is a group of disintegrated cells of the nucellar cap and behind this a globular embryo composed of perhaps 50 cells which evidently remained alive but dormant thruout the summer. Behind this embryo is an area that is free of cellular structures and which was probably filled with fluids. Faint fibrous remains of the nucellus exist about this area.

Seedling No. 12,570.—In respect to the quality of the fruit this seedling, which has the same parentage as No. 12,559, is one of the best of those which bear stenospermocarpic berries. The largest seeds are fleshy and green in color; some have a black spot irregularly placed; some are somewhat brittle; none is hard; and a few seeds in mature berries contain traces of endosperm or embryo. The smaller seeds and the tiny rudiments are fleshy, red in color, and devoid of contents. At the time of anthesis at least some of the ovules of this seedling show some protrusion of the inner integument (Fig. 29). This feature of abnormality was observed in much greater degree in some ovules of Stout Seedless. The protrusion of the nucellus is a regular feature in the ovules of White Corinth for which fruit development is by vegetative parthenocarpy. The embryo sacs in some of the ovules are able to function in fertilization. In Fig. 30 is shown the pollen tube in its course thru the calotta to the egg and synergids.

Seedling No. 12,583.—At the present time this seedling (frontispiece) is rated as one of the most promising of the seedlings that yield seedless or near-seedless fruit. The clusters are of good size and well-filled; the berries reach a diameter of ¾ inch, which is rather large for seedless fruits; the color is a rich light red; the fruits are fully ripe at Geneva on October 1; and the quality is excellent. There are wide differences in the size of the seeds (Fig. 7), but the largest are green in color and pulpy with an enlarged but soft outer integument within which there is only a slight development of the sclerenchyma. Some of the seeds of this seedling have embryo or endosperm in various stages of development. The medium-sized seeds are entirely soft and often irregular in shape. The smaller sizes are red in color. In eating the fruit of this seedling one is not aware of the largest of the seeds.

Seedling 12,583 was obtained from a cross between seedling No. 8,536 (Goff x Iona) and Sultanina. The vine is large, vigorous, and productive; the cluster large, shouldered, long and medium compact; the berry large, oval, adherent, and light red; texture melting and juicy; flavor vinous and nearly sweet; quality good; skin medium tough. This seedling has been named Bronx Seedless. (See frontispiece.)

Seedling No. 15,194.—This seedling has for its seed parent Golden Muscat and for its pollen parent Stout Seedless which in turn had the Sultanina Rose as its pollen parent. Thus, the plant No. 15,194 has two generations of seedless parentage in the pollen parents. Of the 14 plants of this series which have fruited, 6 bear seedless or near-seedless berries. The seedling No. 15,194 is ranked as an excellent plant. The clusters of fruit are of fair size, about 6 inches in length, and they are well filled with berries of which many are ¾ inch in diameter. The seeds (Fig. 8) are all shrivelled at the apical end and noticeably wedge-shaped. The largest are greenish and soft or somewhat brittle. As a rule the larger berries have one of the larger green seeds along with one or two small and almost white rudiments of seeds. The sections of the largest seeds of ripe berries show in some cases several layers of sclerenchyma and a small globular embryo but only shrivelled remnants of endosperm or nucellus.

Seedling No. 17,404.—This seedling, derived from Melton by Stout Seedless, has fruit very like that of the pollen parent. The largest seeds (Fig. 12) are green, soft or slightly gritty, noticeably constricted at the micropylar end, and endosperm and embryo are often present.

Seedling No. 14,099.—This plant has for its parents seedling No. 9,130 (Triumph x Delaware) and Sultanina. The seeds in the ripe berries of this plant are of small size and noticeably flattened and shrivelled. Some are almost white in color; a few are dark in color. The largest seeds appear to be empty or to contain only remnants of a nucellus; the outer integument is relatively thin both for the outer pulpy portion and for the sclerenchyma which is poorly developed. The fruits of this plant are very fully seedless, but the berries are rather small. The clusters are of fair size and well-filled.

Seedling No. 14,105.—The largest seeds in the mature fruits of this plant are undersized, misshapen, and somewhat shrivelled. Most of them are dark and almost black at the larger end. Smaller rudiments, some of which are mere flakes, may also be present. The seed parent is seedling No. 9,135 and the pollen parent is Black Monukka.

Seedling No. 17,191.—This seedling (Fig. 14) produces large clusters of medium-sized berries of good quality. It was obtained from a cross between No. 8,536 (Goff x Iona) and Stout Seedless. The largest seeds are fleshy and soft or slightly gritty. Frequently, there is a sunken area and a small dark spot in the region of the chalaza. The larger seeds often contain well-developed endosperm and embryo. There are some small berries but usually these have one stenospermic seed. The fully aborted ovules are tiny or so small they are not found in dissection.

Seedling No. 17,451.—This seedling, obtained from a cross of Melton with Black Monukka, is one of the few plants thus far obtained which produce stenospermocarpic fruits and have flowers with reflexed stamens. The largest seeds (Fig. 11) are shrivelled, green, and soft. The quality of the fruit is excellent.

Seedling No. 17,936.—This seedling has Golden Muscat for its seed parent and Stout Seedless for its pollen parent. The bunches are large and well filled with greenish amber berries most of which are about 9/16 inch in diameter. The seeds in the larger berries range from 1 to 5; they are undersized, shrunken, soft, and greenish in color. Many of the seeds are empty except for traces of nucellus or for small ball-like groups of cells of an undeveloped embryo. A few small berries are present and these have either small rudiments of ovules or none that can be found.

SEEDLESS FRUITS OF THE CORINTH TYPE

Thus far no seedling obtained in this breeding project has borne fruits in quantity which are all exclusively of the Corinth type. Various seedlings which fruited sparsely in 1935 had a large proportion of such berries. In several seedlings there were mixtures of seedless and seeded berries; in others there were seedless and stenospermocarpic berries. In some cases it was determined that there is facultative parthenocarpy.

For several seedlings it seems that the formation of the relatively large proportion of small berries involves obligate vegetative parthenocarpy. These seedlings are considered to be worthless for culture, at least in comparison with the plants which bear the better stenospermocarpic fruits.

The seeded F_1 seedlings of the progeny of Corinth grapes used as pollen parents with seeded grapes are being selfed, cross-pollinated among themselves, back-crossed with the Corinth grapes, and also crossed with various grapes which have stenospermocarpic fruits, including certain plants that also have vegetative parthenocarpy. In the progenies which are being grown some plants which produce parthenocarpic fruits of the Corinth type may be expected.

DISCUSSION AND SUMMARY

SEEDLESSNESS IN GRAPES

There are two distinct types of seedlessness in the "seedless" Vinifera grapes that have already become important in cultivation and which have been used as pollen parents in the breeding project reported in this bulletin.

The Corinth grapes have relatively small-sized and entirely seedless berries. The ovules are abortive and unable to function in fertilization and the fruits are produced by parthenocarpy. In the White Corinth grape the ovules have but one integument; the nucellus protrudes into the cavity of the ovary as a tongue of tissue that is usually entirely sterile and vegetative, altho in some ovules stages in the formation of an embryo sac are found. In the Black Corinth grape the ovules have the two integuments and the embryo sacs develop, but, except in rare instances, these are abortive and evidently do not function in fertilization. The flowers of these Corinth grapes are of the perfect hermaphrodite type. The stamens produce some viable pollen, but the evidence is that pollination of any sort does not change the character of the fruit. The berries are relatively small and this condition limits the value of these grapes for culture as table grapes. But the fruits are dried and sold as "currants" and the culture of the currant-grapes is an important industry in Greece (Husmann, 1920). The Corinth grapes are not hardy in New York State.

The Sultanina or Thompson Seedless grape exhibits a second type of "seedlessness" which is also seen in the grapes Sultanina Rose and Black Monukka, and possibly also in the Sultana. The berries are larger than those of the currant-grapes and many of them possess some rudimentary or aborted seeds which are often of considerable size but which are so soft and pulpy that they are not noticed when the fruits are eaten. There are also, as a rule, some smaller rudiments of seeds and also some ovules that failed to develop to appreciable size. A considerable number of the berries, especially those of the smaller sizes, may contain only the aborted ovules. Pollination seems to be necessary for adequate yields of fruit and fertilization occurs in at least some of the ovules, but abnormalities appear during the early stages in the development of the seeds. The sclerenchyma, which forms the hard bony part of the outer integument of a normal seed, is feebly developed; the embryo is merely a group of undifferentiated cells; the endosperm is feebly developed; the entire seed is misshapen, shrivelled, and soft. The term *stenospermy* is suggested for this condition, and the fruits which contain such seeds may be designated *stenospermocarpic.*

The grapes which have this type of fruit are especially valuable for culture, both for the production of raisins and for fruit for table use. The fruits are larger than those of the currant grapes. In the clones named above the flowers are of the perfect-hermaphrodite type and the individual plants, as a rule, produce full yields of fruit to self-pollination.

Partial variations in the clusters of fruit in which some of the berries are of small size and seedless and others are of large size and seeded are known in certain grapes. In the important vinifera grape Muscat of Alexandria (Bioletti, 1921), small seedless fruits "occur commonly". Either one of two conditions will produce clusters which have mixtures of seeded and seedless fruit. In one condition, all the ovaries are alike and able to develop as seeded fruits, but they are also able to develop by vegetative parthenocarpy which occurs for those pistils which are not properly pollinated. In this case the parthenocarpy is "facultative" and the proportion of seedless fruits varies greatly according to irregularities in pollination. In another condition, the ovaries are unlike in respect to the complement of ovules and those ovaries which develop as seedless fruits have only the abnormal ovules and are able to develop in no other way and their parthenocarpy is "obligate".

The writer has not studied the Muscat of Alexandria or used it in breeding, but the studies by Negrul (1934) indicate that the conditions in the ovaries and ovules involve several grades and types of development including apogamy.

Seedless grapes have appeared as seedlings and as bud sports from seeded grapes of the types grown in New York State. Such seedless grapes are few and in nearly all cases the berries have been small and the clusters so poorly filled that the plants were discarded. One "seedless" grape has been cultivated under the names Concord Seedless and Hubbard Seedless. The seeds of Concord Seedless are abortive and soft, pollination is necessary for the formation of fruits, and the fruits are evidently stenospermocarpic but somewhat different from those of the Sultanina type. The aborted seeds have large black areas which extend thru a considerable part of the tissues of the seed coats and appear to involve an excessive formation of tannin. This grape has been used as a pollen parent in breeding and progenies have been grown into the F_2 generation, but thus far no seedlings thus derived have seedless fruit.

About 80 seedlings have now been developed which bear fruits of the stenospermocarpic type. These were obtained by using the vinifera seedless grapes as pollen parents in crosses with seeded grapes of the types being grown at the Experiment Station. Many of these seedless plants appeared in the first generation grown from these crosses. Also plants bearing seedless fruit appear in the progeny of crosses between the F_1 seedless plants and seeded plants. There is noticeable variation among these seedlings in respect to the degrees of development in the various parts of the aborted seeds, and especially in the tissue that forms the sclerenchyma. Some of these plants are able to develop fruits by vegetative parthenocarpy. If there is pollination, the fruits are stenospermocarpic; if there is no pollination, the fruits are small, entirely seedless, and vegetatively parthenocarpic. A considerable number of these new "seedless" plants possess a combination of desirable characters and seem worthy of trial under cultural conditions. The plants are hardy, at least for the average winter at Geneva; the clusters are well filled and of good size; the fruits are of good flavor and quality; they ripen at Geneva; and the stenospermic seeds which they contain are soft and not noticeable when the fruits are eaten.

SEEDED GRAPES

The fruits of grapes are to be classed as seeded if there is present one or more seeds that are hard. It is the rule that such seeds are of normal size and shape and that they develop from ovules in which fertilization occurred. The development of seeds without fertilization (apogamy) occurs in a few grapes. There are nearly always some fully aborted ovules along with the one or more hard seeds in a berry and the proportion of these is rather constant for any given clone or individual. The most frequent number of seeds per berry may be one, two, or three, or possibly four, according to the particular clone. The group of "seeded" grapes includes individuals which have at least some fully developed seeds, "empty seeds"', or more or less brittle seeds.

A considerable proportion of the seedlings of the progenies which contain seedless plants are seeded. There are also plants with seeds that are somewhat intermediate between the normal seed and the soft stenospermic seed in some one or more structural features. There are also certain seedlings whose clusters of fruit exhibit partial variations, and there are mixtures of seedless and seeded berries. In some of these the expression of parthenocarpy is facultative; in others it is obligate.

SURVEY OF CONDITIONS INVOLVED IN FORMATION OF SEEDS AND FRUIT IN GRAPES

The different developments in the fruits of grapes and their combinations and interrelations admit of the following descriptive analysis.

THE OVULES

Several types of ovules are to be recognized in grapes with reference to the development of seeds.

A. Unable to function in fertilization and unable to develop as seeds of any sort.—First (a) the ovules may be abnormal in structure and composed of only vegetative tissue, as in most ovules of the White Corinth; or (b) abortions may appear during the formation of the egg apparatus, as in the Black Corinth. This last-named type of embryo-sac abortion evidently occurs in *part of the ovules in nearly all grapes*, both seedless and seeded.

B. Able to develop as stenospermic seeds.—In many ovules of this type fertilization occurs and both the embryo and the endosperm start to develop, but abnormalities appear in the formation of the seed coats and the seed becomes defective and abortive in one or more features. There are many ovules in which the endosperm or the embryo is not found and some in which neither seems to start development. It seems probable, in such grapes as Sultanina, Sultanina Rose, Black Monukka, and numerous seedlings here reported upon, that fertilization of both egg and endosperm nucleus is not normal in many of the ovules which develop into aborted seeds of the smaller sizes, but it seems likely that the entrance of the pollen tube into the ovule as far as the embryo sac may provide a stimulus that results in a small-sized seed of the stenospermic type.

C. Able to develop into hard but empty seeds.—Whether or not this type of seed arises from ovules in which fertilization has occurred seems not to have been determined but may perhaps be assumed.

D. Able to develop after fertilization into normal hard seeds with viable embryos.—This is the rule for the one or more hard seeds in the seeded fruits of grapes.

E. Able to develop without fertilization as hard seeds with apogamic embryos.—In rare cases there is polyembryony.

It appears that the conditions listed above are all obligate to the extent that an ovule cannot also function as one of a higher class; but when ovaries are able to develop by vegetative parthenocarpy without pollination, any ovules of classes *B*, *C* or *D* which are present will fail entirely to develop and will appear in the ripe fruits very similar to those of class *Aa*.

It has been considered that somatic mutations in the character of the ovules occur (a) when viable seeds are produced by the Panariti grape (Harmon and Snyder, 1936), in which case the mutation is from aborted ovules to ovules that can produce seeds, and (b) when ovaries which bear mostly parthenocarpic berries arise from the Sultanina grape which produces mostly stenospermocarpic fruits, in which case the mutation is from ovules which can function in fertilization to those which do not.

THE OVARY AND ITS DEVELOPMENT INTO A FRUIT

In grapes the normal ovary is two-celled and each cell has two ovules. Occasionally, there are three cells and more than four ovules. Except for the White Corinth and the Rose Corinth, the ovaries of all grapes are quite alike in that the full complement of ovules are present and developed as far as the stages when the egg apparatus or embryo sac is organized. As a rule the four ovules in any ovary of any of these grapes look alike at the time when the flower opens, but the ovules soon become different in their ability to function in fertilization and

seed formation. The character of the fruit is often determined by the one or more ovules which are not abortive but which proceed to develop either as aborted seeds or as hard seeds.

In respect to the nature of the fruit which develops from an ovary the following classes are to be recognized.

I. *Parthenocarpic and entirely seedless.*—1. When a fruit develops without any pollination it is purely a vegetative structure and there is vegetative or autonomic parthenocarpy. (a) If all the ovules are of class *A* as designated above, the parthenocarpy is obligate. (b) If one or more ovules in an ovary are of the class *B*, a parthenocarpic fruit develops only when there is no pollination and its parthenocarpy is hence facultative. 2. Stimulative parthenocarpy is evidently only expressed in an ovary in which all ovules are of the class *A* or when pollen tubes fail to function in the fertilization of ovules that are capable of fertilization. It seems that some of the berries of Sultanina, Sultanina Rose, Black Monukka, and various seedlings are of this type and that pollination is necessary even when a considerable proportion of the berries are entirely seedless. It may be anticipated that there are cases of complete obligate ovule abortion (as in White Corinth) in which pollination may increase the size of the fruit.

II. *Stenospermocarpic fruits.*—In these one or more of the ovules develop into stenospermic seeds. However, in nearly all ovaries some of the ovules are of class *A*.

III. *Seeded fruits.*—Here one or more ovules develop into hard seeds, but such seeds may be "empty-seeded" or they may be normal and viable and in the latter case the embryos are usually the result of fertilizations altho cases of apogamy are known. In all individuals and cultivated clones, however, the number of such seeds per fruit is usually less than four because some ovules of class *A* or of both *A* and *B* are also present.

THE FLOWER

The flowers are of three main classes, as follows:

1. *Staminate* which has well-developed stamens which yield much viable pollen but in which the pistil is only rudimentary. Such flowers produce no fruit.

2. *Imperfect hermaphrodite* which usually has recurved stamens that produce defective pollen; but the fertile pistil produces fruit of some one of the types noted above. This type of flower is functionally pis-

tillate. A structurally pistillate type of flower has been reported (Baranov, 1927 A and B), but this occurrence seems to be rare in grapes.

3. *Perfect hermaphrodite* which has normal stamens with much viable pollen and a pistil which may develop into one of the following types of fruit, namely, seedless, stenospermocarpic, or seeded.

There are wide differences in the precise character of both stamens and pistils of the flowers of individual seedlings and cultivated clones in each of the above classes.

THE INDIVIDUAL PLANT AND ITS CLUSTERS OF FRUIT

The unit in vineyard culture is the clone composed of plants derived by the vegetative multiplication of a single individual. For the individual, a typical cluster of its flowers and fruit may be regarded as a unit of production.

In respect to the character of the flowers, the flowers of a plant may be (1) all alike and either staminate, imperfect hermaphrodite, or perfect, as noted above; or (2) there may be mixtures of two types of which the mixture of staminate and the perfect hermaphrodite is readily recognized.

In respect to the nature of the berries,

1. *All berries may be alike,* in which case they may belong to any one of the classes or sub-classes indicated above under I, II, and III; or

2. *There may be partial variation* with the clusters consisting of a mixture of two or more types of fruit. This latter case may involve one or more of several conditions, as follows: (a) There may be mixtures of parthenocarpic and stenospermocarpic berries. When some ovaries contain only ovules that are unable to develop into seeds (class *A*) the berries developed from such ovaries are formed by an obligate parthenocarpy and pollination of any kind or degree does not change the nature or the proportion of these particular fruits. But when ovaries contain some ovules capable of developing into stenospermic seeds, those which become parthenocarpic do so because pollination failed for their pistils. In both cases, if the parthenocarpy is vegetative, a complete lack of pollination results in clusters composed of parthenocarpic fruits only.

(b) The clusters of fruit may be mixtures of parthenocarpic fruits and seeded fruits. When proper pollination will change all the berries to seeded fruits, the parthenocarpy is vegetative and facultative. When pollination does not change the proportion of seedless fruits the ovules are of two classes, I and III. If the parthenocarpy is vegetative, the lack of any pollination will in either case, result in clusters composed only of seedless fruit; but if it is stimulative, no fruit will be formed.

(c) The clusters may be mixtures of parthenocarpic fruits, stenospermocarpic fruits, and seeded fruits. In two of the seedlings which the writer has studied that have clusters of this sort adequate pollinations do not change the proportions of berries that are parthenocarpic and stenospermocarpic. Here, as in the case above, lack of any pollination gives only parthenocarpic fruit if there is vegetative parthenocarpy, and no fruit if there is stimulative parthenocarpy.

According to Negrul (1934), further partial variations may be present which involve apogamy, but this condition has not been observed or identified in the clones or seedlings concerned in the studies here reported.

BREEDING FOR SEEDLESSNESS

The production of 83 seedless or near-seedless seedlings in this breeding project has shown that hardy seedless grapes suitable for culture in New York State are to be obtained in the F_1 generation as well as in later generations of crosses between the tender seedless vinifera grapes and hardy seeded types. It is not the intention to discuss here the genetical significance of this fact or to present and discuss the data obtained in the breeding. There are many seedlings of the same sort of breeding which have not yet fruited and it is expected that more seedless plants will appear among these. The best of these seedlings afford excellent material for further selective breeding in the effort to produce still better seedless grapes in respect to quality and size of fruit, productiveness, and hardiness. There is considerable diversity among the best of these seedless grapes which admits of selective breeding.

In grapes there is the possibility of utilizing vegetative parthenocarpy in connection with complete ovule sterility due to hybridity, unisexualism, hereditary abortion, or polyploidy in obtaining fruits of large size. When there is vegetative parthenocarpy the pistils which are completely "sterile" are able to mature as purely vegetative fruits.

In respect to this matter the hybrids between *Vitis rotundifolia* and *V. vinifera* are of special interest. Detjen (1919) obtained a few berries on one such hybrid, but the seeds apparently were unviable for they did not grow. Mr. Charles Dearing in a recent letter to the author states that "all the hybrids between *V. rotundifolia* and *V.*

vinifera proved to be sterile, although they produced some fruit. This fruit proved to be more or less seedless and if seeds developed at all they were not viable". If such hybrids were obtained from vinifera parents that have parthenocarpy, either obligate or facultative, at least some of the sterile hybrids might bear seedless fruit.

At the present time the progress in obtaining new seedless grapes of merit in the breeding here reported has been in the grapes which produce fruit by stenospermocarpy. Several of the seedlings already obtained possess much merit and these are being propagated for trial under general cultural conditions.

CONCLUDING STATEMENTS

1. In grapes it is the rule that the normal viable seeds develop from an ovule and its fertilized egg, but cases of the development of such seeds without fertilization (by apogamy) are known. The embryo and endosperm develop slowly for a period of two or more weeks after fertilization but during this period there is a rapid development and enlargement of other parts of the seed and especially of the sclerenchyma which forms the hard bony tissue.

2. The seedless fruits of grapes are of two main types. In one type, exemplified by the Corinth grapes, the fruits develop without fertilization and are entirely seedless, a condition called *parthenocarpy*. In another class, exemplified by the Sultanina grape, the fruits may contain one or more aborted seeds some of which formed from ovules in which fertilization occurred, a condition here designated *stenospermocarpy*.

3. For the typical stenospermic seed the abortions appear soon after the time of fertilization in the feeble formation of sclerenchyma and of other tissues which combine to give shape, size, and character to the seed. The endosperm and embryo remain in a partly developed state, and the largest seeds in the mature berries are undersized, abnormal in shape, and soft or pulpy.

4. There are at least six types of ovules in grapes, as follows:
   (a) Abnormal and vegetative in structure and embryo sac usually lacking as in the White Corinth and the Red Corinth.
   (b) Slightly abnormal in structure, but the embryo sac is defective and unable to function in fertilization, as in the Black Corinth grape. But ovule abortions occur in some of the ovules in all varieties of grapes.

   (c) Able to function in fertilization but seed stenospermic.
   (d) Able to develop as an empty seed.
   (e) Able to develop as a normal seed when there is fertilization.
   (f) Able to develop without fertilization as an apogamic seed.

5. The type of fruit which develops from an ovary is determined by three main factors, *viz.*, (a) by the character of the different ovules which it contains, (b) whether parthenocarpy is present or not, and (c) whether or not pollination is accomplished.

6. Parthenocarpy is of two types; (a) when entirely seedless fruits develop without fertilization, the condition is called *vegetative* or *autonomic* parthenocarpy; (b) when the formation of a fruit depends upon, or is influenced by, the stimulus of pollen on the stigma or by the growth of pollen tubes in the style or ovary the parthenocarpy is called *stimulative* or *aitionomic*.

7. When vegetative parthenocarpy operates in a plant whose pistils contain only aborted ovules, as in the Corinth grapes, the parthenocarpy is obligate and only seedless fruits are produced under any circumstance. But when it exists in a plant whose ovaries can also develop either as stenospermocarpic fruits or as seeded fruits, the seedless berries form only when there is no pollination or at least no fertilization. In this instance the parthenocarpy is *facultative*.

8. Poorly filled bunches of berries occur in grapes (a) when some of the flowers have rudimentary or aborted ovaries and others have functional pistils, either when there is parthenocarpy or when there is none; (b) when some of the ovaries contain only aborted ovules and there is no parthenocarpy; and (c) when pollination is necessary for the formation of fruit but some pistils are not pollinated.

9. Mixtures of seedless and stenospermocarpic fruits, seedless and seeded fruits, or even of seedless, stenospermocarpic, and seeded fruits may occur in the same cluster. In all such cases there is vegetative parthenocarpy. But the development of the seedless fruits may be *facultative*, in which case the ovaries involved are able to develop as fruits of a higher grade, or *obligate*, in which case the ovaries can develop in no other way. Further complications in partial variations in the character of fruit may involve stimulative parthenocarpy and apogamy.

10. The hybrids thus far obtained between the Muscadine grapes and members of the Euvitis group which differ in the number of chromosomes are so sterile from spore abortions that very few fruits

have been borne by them and these have had seeds that did not germinate. The clonal varieties of grapes now cultivated in New York State are evidently all hybrids between species of the Euvitis group which have the same diploid number of chromosomes and they are highly fruitful.

11. In grapes the entirely seedless fruits of the parthenocarpic class are smaller than stenospermocarpic fruits, and the latter are smaller than seeded fruits. But various factors, such as heredity and the number of seeds per berry, are also involved in determining the size of fruit.

12. In grapes at the present time the small size of the berries which are vegetatively parthenocarpic limits the value of plants with this type of fruit, but it may be possible to obtain by selective breeding plants which produce by vegetative parthenocarpy fruits of much larger size.

13. The most valuable type in the production of new seedless grapes seems, at the present time, to be that with stenospermocarpic fruits. The flowers should be perfect to provide adequately for the necessary pollination and the aborted seeds should at least be soft, if not of small size. It will also be of some value if vegetative parthenocarpy is also present in the plant.

14. Eighty-three seedlings have already been obtained in the project here reported upon which bear seedless or near-seedless fruits of the stenospermocarpic type and some of these are also able to develop fruits by vegetative parthenocarpy. These seedlings are of the first and the second generation of hybrids between hardy seeded grapes and the grapes Sultanina, Sultanina Rose, and Black Monukka. The best of these plants are being used in further selective breeding and several are being propagated for trial culture in New York State.

ACKNOWLEDGMENTS

With the permission of the authorities of The New York Botanical Garden the author of this bulletin, since 1919, has cooperated with the Division of Pomology of The New York State Agricultural Experiment Station in a project which aims (1) to develop by breeding hardy seedless grapes of value for culture in a climate such as that of central New York, (2) to obtain data on the inheritance of seedlessness, and (3) to determine by experimental and cytological studies the nature and scope of the various types of seedlessness in grapes. The greater part of the work of breeding has been performed at Geneva and the many seedlings obtained have been grown and cared for at the Station.

The New York Botanical Garden has provided for most of the expenses involved during the periods of work at Geneva during each spring and autumn. A special contribution of $100.00 to the support of this work was made by the late

Mr. Anton Hodenpyl. A grant of $200.00 from the Committee on Grants in Aid of the National Research Council provided for these expenses in 1935 and also for the assistance of Miss Marylizabeth Wellington during the period of work in June 1935.

The morphological and cytological studies reported in this bulletin were all made in the laboratories of The New York Botanical Garden, but much of the material used was collected at Geneva by Dr. B. R. Nebel. Several thousand slides for the microscopic studies were prepared by Miss Theresa Huth and Mr. James Yearwood who were assigned to this project under the United States Works Progress Administration thru the special interest and consideration of their supervisor. The laboratory work was performed under the immediate supervision of Miss Clyde Chandler, technical assistant to the writer, who also made many of the more difficult of the preparations, made the preliminary studies of the material, and took the photomicrographs. Miss Fleda Griffith, photographer at The New York Botanical Garden, devoted special attention to the preparation of the prints used in the plates. Mr. Charles Roberts arranged the plates as they are here reproduced and Miss Eleanor Clarke made the painting reproduced in the frontispiece. Mr. Roberts and Miss Clarke were supplied by the WPA. The author is also indebted to Mr. Frank Landi and Mr. Victor Shortino for translations of Italian articles and to Mrs. Lena Lewin, a PWA worker, for translation of Russian literature.

To Dr. U. P. Hedrick and Mr. Richard Wellington the writer is especially indebted for continued support of the entire project since its earliest inception and for complete cooperation in the organization and execution of all phases of the work which it involves at Geneva. Various members of the Division of Pomology have, from time to time, assisted in the routine of the breeding. To all those noted above and to all others not mentioned who have contributed directly or indirectly to this bulletin, the writer is pleased to express his obligation and appreciation.

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