# On the Mutable Genes of Pharbitis, with Special Reference to their Bearing on the Mechanism of Bud-Variation.

By

**Yoshitaka Imai.**

With Plate XIII and six Text-figures.

CONTENTS.



## Introduction.

The genes, generally speaking, are relatively stable. Those, however, with low stability are called mutable genes. As the majority of the mutable genes mutate automatically, they are automutable. The contrary case, though of rare occurrence, is exomutable, the recurring mutations being affected by the stimulation of other genes. Mutability may be influenced by the different stages of ontogeny, including gametophytic generation, as well as by changes in other environmental factors. Further, there have been found some genes that affect mutability. The direction of the recurrent mutations is generally definite, changes sometimes occurring in two or more directions. In certain cases, mutations arise only in so-

[Jour. Coll. Agric., Vol. XII, No. 3, 1934.]

matogenesis, in others either only in gametogenesis or in both cases. The mutable nature of the genes is of importance in studying the mechanism of bud-variations, or in that of the formation of periclinal chimeras. The purpose of this paper is to describe the behavior of mutable genes in *Pharbitis*, extending the discussion to the general problems above stated.

## List of Mutable Genes in *Pharbitis*.

In *Pharbitis Nil* twenty (possibly more) mutable genes, situated in seventeen loci, were investigated, including those unpublished. The following list contains these loci with short remarks.

1. contracted (*ct*): Whole plant body contracted, reverting to normal; most of the contracted strains however are constant (IMAI 1927a, and present paper).

In the former publication (IMAI 1927a), I presented data on the mutability of gene contracted, giving somatic reversion as well as seminal mutants that were heterozygous normals. Further new data proved the lability of the contracted line, two progeny plants

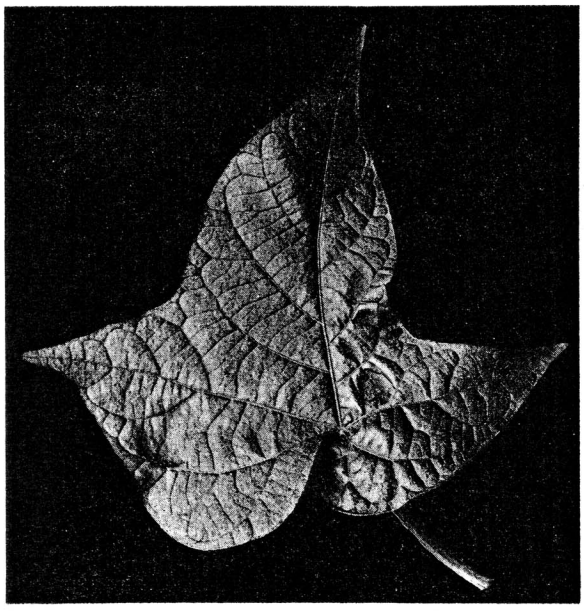


Fig. 1. Sectorial leaf, half normal (left) and half contracted (right). Crumpling of leaf is due to gene crumpled-1.

being the mosaic of contracted and normal. One of the specimens showed a sectorial chimera, partly contracted and partly normal, sometimes bearing mosaic leaves (Fig. 1) and flowers. But the other was contracted, bearing periclinal branches with contracted 'skins' and normal 'cores' (Fig. 2). On account of the coarse texture of the inner mesophyll, the leaves of the chimerical branches

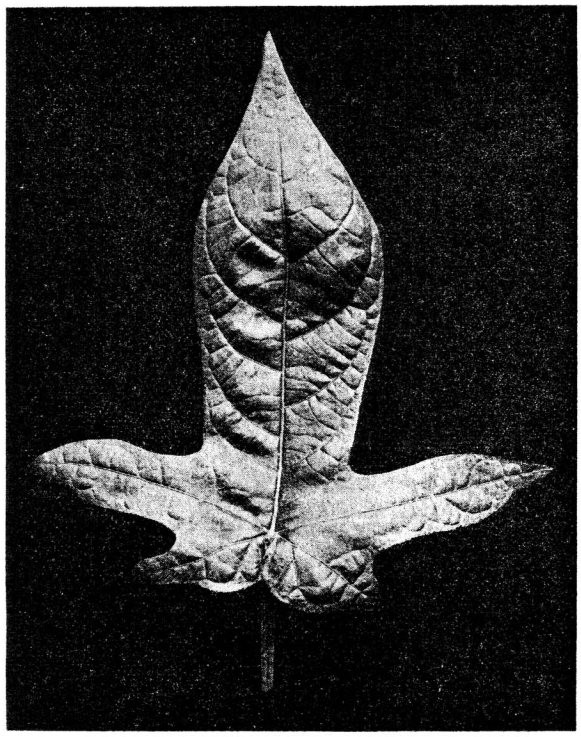


Fig. 2. Periclinal leaf with contracted 'skin' and normal 'core'. Note crumpling of leaf due to heterogeneous tissues, but not to gene crumpled-1.

are somewhat crooked and crumpled. The data presented in Table I were obtained through sowing seeds collected from the latter mosaic plant.

TABLE I.

| Character of branch | Normal | Contracted | Total |
|---|---|---|---|
| Contracted | 0 | 21 | 21 |
| Periclinal | 1 | 24 | 25 |

The results accorded with our expectations, proving the periclinal structure of the branches that had contracted sub-epidermal tissues. The mutable contracted line however was lost before making further studies. BLAKESLEE's dwarf *Portulaca* (1920) is also mutable, reverting to normal.

2. cream (*cr*): Creamish flowers with a few normal-colored stripes, due to recurring somatic mutations: seminal mutants also occur (IMAI 1925a, 1927a).

3. delicate (*dl*): Leaves with sharp-pointed lobes and less protruding shoulders, mesophyll thin, flowers split, fertility low; sometimes mutable, reverting to normal (IMAI 1927a).

4. dotted (*dt*): Dotted flower, reverting to self-colored condition (TERAO and U 1930).

5. duskish (*dk*): Composed of a graded series of multiple allelomorphs. Roughly speaking, they are near-stable-plain ($dk^{ns}$), unstable-plain ($dk^{us}$), ordinary-ruled ($dk^{or}$), highly-ruled ($dk^{hr}$), and normal. Near-stable-plain and unstable-plain result in plain duskish flowers, sometimes with a few normal-colored spots; ordinary-ruled and highly-ruled produce variegated flowers with different amount of variegation. Near-stable-plain rarely reverts to normal, others transform mutually and to normal (IMAI 1931, and in press c).

6. flecked (*fl*): White flower with normal-colored flecks, frequently reverting to self-colored condition (IMAI 1931, and present paper).

7. lobeless (*ls*): Lobeless leaves, complete sterility, reverting to normal (TERAO and U 1930).

8. miniature-inconstant ($mi^i$, probably allelomorphic to constant miniature): Organs miniature, flowers split, bloom in small cluster, complete sterility; reverts to normal (TERAO and U 1930).

9. pine-inconstant ($pi^i$): Cotyledons very small, leaves filiform, flowers split very narrowly, reverting to normal and mutating to constant pine (IMAI 1927a; U 1930).

The *phantastica* form of *Antirrhinum* has also 'nadelartig' leaves, frequently reverting to normal (BAUR 1924, 1926a). Another slender leaf form of this plant, *graminifolia*, seems also to be mutable (SCHIEMANN 1926).

10. pipy (*pp*): Flowers tubular in form, at times reverting to normal (IMAI and KANNA, unpublished data).

11. purple (*pr*): Purple flower, rarely reverting to normal

condition (IMAI, present paper).

A few remarks on the behavior of gene purple will now follow. Purple, which is a recessive gene producing a purple flower, results in a red flower in cooperation with gene magenta. Close examination of purple or red flowers reveals at times the occurrence of blue (on purple flowers) or magenta (on red flowers) flecks, rarely forming their mono-chromatic flowers. By selfing six plants that bore purple flowers with some blue stripes, I raised 272 seedlings, 4 of which later bloomed blue flowers and the rest purple, excepting one mosaic. The occurrence of normal variants is 1.5 percent. Two of these normals were examined for their progeny, which consisted of 78 normal and 24 purple flowers. The normal variants are therefore due to mutations from recessive to dominant in their origin. The mosaic plant for blue and purple flowers gave rise to 42 purple plants from purple flowers, whereas 35 blue and 13 purple plants were obtained from blue flowers, showing the occurrence of a vegetative mutation in the mother plant. Reverted blue flowers, when they became homozygous, bred true to type in the offspring. On the basis of these data we arrived at the conclusion that gene purple is of mutable nature, reverting to normal.

12. speckled (*sp*): Speckled flowers, rarely reverting to self-colored (TABUCHI, unpublished data).

13. willow ($m^w$): Willow leaves, corolla splits into narrow petals, pistilate organ nearly completely sterile, mutating to maple (IMAI 1925b, 1927a) and rarely to normal (TABUCHI, unpublished data).

14. wrinkled (*wr*): Corollas wrinkled, somewhat star-shaped, fertility generally low, rarely reverting to normal (IMAI 1927a).

15. xanthic (*x*, formerly called golden): Yellow cotyledons and leaves; dies after germination without extending leaves; sometimes with green patches due to somatic reversions (IMAI 1930b; TERAO and U 1930).

16. yellow-inconstant ($y^i$): Yellowish green cotyledons and leaves, frequently with green patches; revert to normal (IMAI 1927b, 1930a, and present paper; MIYAZAWA 1929, 1932).

17. yellowy (*ye*): Young leaves yellowish green and finely mottled with green dots, green mottling extends later, become nearly green leaves, sometimes reverting to normal (IMAI 1930b, and present paper).

The mutable genes so far known in *Pharbitis* act as recessives to the prototypic normal, from which they arose by sporadic mu-

tations in the course of evolution under cultivation. The recurring mutations occur both vegetatively and seminally. In some of my previous papers, I hesitated to regard the anthocyanin flecks in the corollas as results of mutation, but recent experiments cause me to believe that they are so. That is, in such flowers as cream, flecked, purple, and possibly dotted, the anthocyanin flecks are evidently due to recurring somatic mutations that occurred in the late cell-generation of the corollas. In speckled, however, the variegation, general speaking, is a pattern character, sometimes mixed together with a few flecks due to somatic mutations. The duskish variegation seems to depend on plasmic manifestation under the control of certain genes (IMAI, in press c). All mutable genes so far found in *Pharbitis* exhibit both vegetative and seminal mutations. With the exceptions now to be stated, the recurring mutations take place as reversions from the recessives to their dominant prototype. The willow, however, generally mutate to maple, which forms multiple allelomorphs with normal and willow. In this set of allelomorphs, maple behaves as recessive to normal and dominant to willow. Therefore the recurring mutation occurs to the gene of the second rank in the allelomorphic series, but not to the prototype. In the duskish graded allelomorphs, some complicated reversible changes arise, frequently from recessive to dominant and *vice versa*. This is the only case found in *Pharbitis* in which the recurring mutations take place also from dominant to recessive. Of these twenty mutable genes, yellow-inconstant, yellowy, and flecked will be treated further in the following sections.

## Two Yellow-inconstant Stocks.

Gene yellow, which results in mono-chromatic yellowish green leaves (known as 'yellow leaves'), is located in the yellow chromosome (IMAI 1933, IMAI and TABUCHI 1933). Yellow is allelomorphic to yellow-inconstant and normal (IMAI 1930a), the former manifesting yellowish green leaves, frequently with green patches and the latter self-green leaves. The order of dominancy is normal—yellow-inconstant—yellow. Of these multiple allelomorphs, yellow-inconstant is mutable, frequently changing to its dominant condition, but never to its recessive state, while the others are highly constant. The origin of yellow-inconstant is old and its history can

be traced back about 120 years (IMAI 1927b). Its strain, which is still being maintained, has been the subject of my investigations. MIYAZAWA (1929, 1932) also studied the same character of this plant, but its origin is different; it was obtained under his observation as a new mutant character in a hybrid progeny. Both forms being mutable, they bear yellow leaves with green patches and show mosaics of yellow and green as well as green mutants in their progeny. The appearance of the green and mosaic plants in the two forms of yellow-inconstant, however, differ greatly in their frequency. With the object of making comparative studies of them, we exchanged seeds, I receiving seeds of the new yellow-inconstant from Professor MIYAZAWA, to whom I wish to express my hearty thanks for his generosity. To assist identification, we shall designate the old line by yellow-inconstant-1 and MIYAZAWA's by yellow-inconstant-2. In 1930 four plants were obtained from his seeds, one green and three yellow-inconstant-2. These yellow-inconstant-2 plants bore yellow leaves with some green patches, but scarcely conspicuous enough to be set down as mosaics. On selfing they gave the progenies shown in Table II.

TABLE II.

| Pedigree No. | Green | Mosaic | Yellow-inconstant-2 | Total |
|---|---|---|---|---|
| 4 | 88 | —— | —— | 88 |
| 1 | 4 | 13 | 13 | 30 |
| 2 | 14 | 21 | 26 | 61 |
| 3 | 34 | 86 | 43 | 163 |
| Total | 52 | 120 | 82 | 254 |
| Percentage | 20.5 | 47.2 | 32.3 | 100 |

The proportion of the green and mosaic plants is very high compared with that observed in my own strain. The mosaic plants showed variegation of green and yellow, frequently showing chimerical arrangement of the two parts in a sectorial or a periclinal form. In Table III are given additional data obtained from the succeeding generation.

Of seven green plants, one proved to be homozygous and the others heterozygous. In the segregating green pedigrees, the fre-

quency of the yellow-inconstant-2, including the mosaics, was very

TABLE III.

| Pedigree No. | Green | Mosaic | Yellow-inconstant-2 | Total |
|---|---|---|---|---|
| Offspring of green plants | | | | |
| 1—9 | 55 | — | — | 55 |
| 1—3 | 14 | 1 | 0 | 15 |
| 1—10 | 38 | 2 | 2 | 42 |
| 2—3 | 16 | 3 | 0 | 19 |
| 2—6 | 96 | 13 | 7 | 116 |
| 3—1 | 14 | 2 | 3 | 19 |
| 3—2 | 32 | 4 | 6 | 42 |
| Total | 210 | 25 | 18 | 253 |
| Percentage | 82.9 | 10.0 | 7.1 | 100 |
| Offspring of mosaic plants | | | | |
| 2—1 | 39 | 3 | 2 | 44 |
| 2—2 | 33 | 24 | 13 | 70 |
| Total | 72 | 27 | 15 | 114 |
| Percentage | 63.1 | 23.7 | 13.2 | 100 |
| Offspring of yellow-inconstant-2 plants | | | | |
| 1—1 | 6 | 11 | 9 | 26 |
| 1—2 | 8 | 16 | 7 | 31 |
| 1—4 | 17 | 19 | 10 | 46 |
| 1—5 | 5 | 10 | 8 | 23 |
| 1—6 | 2 | 4 | 3 | 9 |
| 1—7 | 13 | 27 | 10 | 50 |
| 1—8 | 18 | 26 | 23 | 67 |
| 2—4 | 9 | 17 | 17 | 43 |
| 2—5 | 3 | 14 | 4 | 21 |
| Total | 81 | 144 | 91 | 316 |
| Percentage | 25.6 | 45.6 | 28.8 | 100 |

low compared with that observed in the corresponding case of the yellow-inconstant-1 (17.1 percent against 23.6 percent), the difference being mainly due to the higher mutation ratio in the former. In the yellow-inconstant-1, the mosaic plants, on account of low mutability during post-embryonic somatogenesis, may continue the

condition of the sectorial and periclinal forms rather typically throughout their growth. But since in the case of MIYAZAWA's yellow-inconstant, the vegetative mutations at times occur even in the post-embryonic somatogenesis, they naturally interfere with and complicate secondarily the original chimerical arrangements of the heterogeneous tissues, the green parts of which are due to mutations that occurred during embryonic development. Should, in addition to this, a somatic rearrangement of the tissues take place in the plant ontogeny, the mosaic nature will be more complicated. Such mosaic plants generally give a higher ratio of green variants in their progeny. Reviewing the chlorophyll variegation in the ever-sporting races, CHITTENDEN (1927) wrote: "Perhaps the most peculiar fact about these *variegata* varieties is that the green sports described all appear to be solid green. From our knowledge of other sports we should expect the periclinal condition to be the most frequent, and the solid green branches to appear only very rarely." Thorough observation however disclose some periclinal sports on other vatiegated races as well. For instance, in the variegated *Mirabilis* some sectors with yellow-over-green chimerical leaves were observed. Such periclinal chimeras are observable in all variegated tri-histogenic plants carrying mutable genes.

The offspring of the yellow-inconstant-2 recorded in 1932 contained 25.6 percent green, 45.6 percent mosaic, and 28.8 percent yellow-inconstant-2. These figures are nearly the same as those obtained in the preceding generation (20.5 percent, 47.2 percent, and 32.3 percent respectively). The average figures calculated on the basis of their total numbers (133 green, 264 mosaic, and 173 yellow-inconstant-2) is 23.3 percent green, 46.3 percent mosaic, and 30.4 percent yellow-inconstant-2, which roughly agree with MIYAZAWA's data[1] published in 1929 and 1932 (giving 11.8 percent, 58.8 percent,

1) MIYAZAWA's data were as follows:

| Year | Green | | Mosaic | | Yellow-inconstant-2 | | Total |
|---|---|---|---|---|---|---|---|
| 1927 | 67 | 14.0% | 295 | 61.6% | 117 | 24.4% | 479 |
| 1929 | 49 | 10.8% | 253 | 55.7% | 152 | 33.5% | 454 |
| 1930 | 4 | 5.0% | 48 | 60.0% | 28 | 35.0% | 80 |
| 1931 | 61 | 4.9% | 186 | 14.9% | 1001 | 80.2% | 1248 |

The 1931 data show a very high percentage of yellow-inconstant-2 and a remarkably low percentage of mosaic. The deviation seems so remarkable that we cannot ignore it and there must be some causes at work to bring it about. Excluding therefore 1931 data from the calculation, we get a total of 120 green, 596 mosaic, and 297 yellow-inconstant, which works out to 11.8 percent, 58.8 percent, and 29.4 percent respectively.

and 29.4 percent respectively). The difference in frequency of the green and mosaic variants between the two yellow-inconstant strains is evident, as stated in my former paper (IMAI 1930a). To enable more accurate comparison, new data covering the yellow-inconstant-1 are given in Table IV.

TABLE IV.

| Number of pedigrees | Green | Mosaic | Yellow-inconstant-1 | Total |
|---|---|---|---|---|
| 35 | 120 (5.2%) | 212 (9.3%) | 1958 (85.5%) | 2290 |

The following is a comparison of the newly obtained percentages with those estimated for the yellow-inconstant-2:

| Stock | Green | Mosaic | Yellow-inconstant |
|---|---|---|---|
| Yellow-inconstant-1 | 5.2% | 9.3% | 85.5% |
| Yellow-inconstant-2 | 23.3% | 46.3% | 30.4% |

The large surplus of green and mosaic in the new strain induces a remarkable deficit in the yellow-inconstant. This difference is due to unlike mutability of the two stocks.

In 1933, some $F_2$ derived from crosses between yellow-inconstant-1, yellow-inconstant-2, and yellow were placed under observation, the data obtained being collected in Table V.

TABLE V.

| Cross | Green | Mosaic | Yellow-inconstant | Yellow | Total |
|---|---|---|---|---|---|
| Yel.-inc.-1 × yellow | 3 (0.8%) | 16 ( 4.3%) | 351 (94.9%) | | 370 |
| Yel.-inc.-2 × yellow | 4 (1.2%) | 46 (14.2%) | 274 (84.6%) | | 324 |
| Yel.-inc.-1 × yel.-inc.-2 | 8 (4.5%) | 106 (60.2%) | 62 (35.3%) | — | 176 |

The proportional distribution of the green, mosaic, and yellow-inconstant in these crosses is graphed in Fig. 3, in comparison with the two homozygous yellow-inconstant strains.

The crossing of yellow-inconstant-1 with yellow diminished the proportions of green and mosaic, which however may be attributed to segregation of the yellow. Theoretically, 25.0 percent of $F_2$ are yellow segregates, which are fully constant, and the rest homozygous

and heterozygous yellow-inconstant-1, including their mutants. The

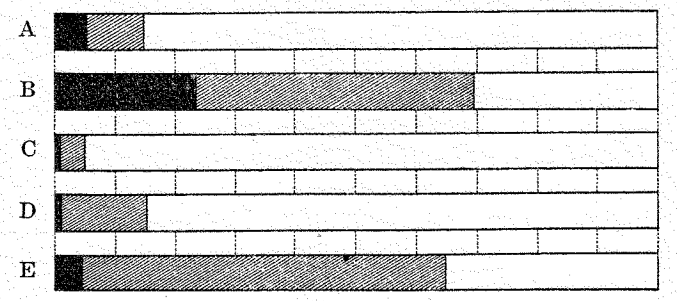



Fig. 3. Indicating the proportion of the green (black part), the mosaic (shaded part), and the yellow-inconstant (white part) in the offspring of yellow-inconstant-1 (A) and yellow-inconstant-2 (B), and in $F_2$ of cross yellow-inconstant-1 × yellow (C), of cross yellow-inconstant-2 × yellow (D), and of cross yellow-inconstant-1 × yellow-inconstant-2 (E).

expected percentages are therefore

| Green | Mosaic | Yellow-inconstant-1 | Yellow |
|---|---|---|---|
| 3.9 | 7.0 | 64.1 | 25.0 |
| | | 89.6 | |

The theoretical percentages show some surplus in the green and in the mosaic which, if reliable, may indicate the heterozygous effect of yellow-inconstant-1 on the rate of mutability. In the case of crossing yellow-inconstant-2 × yellow, the theoretical percentages of the respective segregates are

| Green | Mosaic | Yellow-inconstant-2 | Yellow |
|---|---|---|---|
| 17.5 | 34.7 | 22.8 | 25.0 |
| | | 47.8 | |

In the production of green and mosaic, the theoretical figures are much greater than the observed. This seems to be due to the fact that the mutability of yellow-inconstant-2 is greatly lessened in the hybrids that are heterozygous for yellow. The theoretical percentages obtained through similar treatment with data of crossing yellow-inconstant-1 × yellow-inconstant-2 is as follows:

| Green | Mosaic | Yellow-inconstant |
|---|---|---|
| 18.8 | 37.0 | 44.2 |

The calculation for these percentages, which was made on the assumption that yellow-inconstant-2 is dominant to yellow-inconstant-1, or that a high rate of mutability is dominant to a low, results in figures that deviate considerably from those actually observed. The percentages of the green produced is smaller, while those of the mosaic is larger compared with what were expected. It is however too early to draw any final conclusions from these data regarding the behavior of yellow-inconstant in the hybrid states.

As have been pointed out elsewhere (IMAI 1927b, 1930a; MIYAZAWA 1929, 1932), young seedlings may at times change their cotyledonous characteristics in later growth. Table VI also shows such changes in certain out-bred cultures.

TABLE VI.

| Cross | Cotyledonous seedling | Full-grown plant | | | Total |
|---|---|---|---|---|---|
| | | Green | Mosaic | Yellow-inconstant and yellow | |
| Yel.-inc.-1 × yellow | Green | 3 | 1 | 0 | 370 |
| | Mosaic | 0 | 8 | 5 | |
| | Yellow | 0 | 2 | 351 | |
| Yel.-inc.-2 × yellow | Green | 4 | 0 | 3 | 324 |
| | Mosaic | 0 | 8 | 22 | |
| | Yellow | 1 | 12 | 274 | |
| Yel.-inc.-1 × yel.-inc.-2 | Green | 8 | 0 | 0 | 176 |
| | Mosaic | 6 | 52 | 40 | |
| | Yellow | 0 | 8 | 62* | |
| Total | Green | 15 | 1 | 3 | 870 |
| | Mosaic | 6 | 68 | 67 | |
| | Yellow | 1 | 22 | 687 | |

* All yellow-incostant.

The proportion of individuals that change their forms (in heavy type) is 2.2 percent in the cross of yellow-inconstant-1 × yellow, 11.7 percent in cross yellow-inconstant-2 × yellow, and 30.7 percent in cross yellow-inconstant-1 × yellow-inconstant-2; the proportions being greater in cases of higher mutability. The mosaic cotyledons quite frequently produce yellow-inconstant leaves, while cases in which

this is reversed alco occur at times. The other alternations, including some extreme cases in which yellow cotyledons produce purely green leaves or *vice versa*, are rather rare. In these alternation cases, the genetic types of growing points that are situated between two cotyledons differ from those of the cotyledons. The chances for differentiation are heightened in cases of higher mutability.

The frequency of the seminal mutants (majority of the green) in the two yellow-inconstant stocks differs considerably. The vegetative mutations also show conspicuous differences in their frequencies, while the times at which mutations occur also differ somewhat. In the yellow-inconstant-1, as was pointed out elsewhere (IMAI 1930a), vegetative mutations frequently take place during embryonic development, rarely during post-embryonic somatogenesis, and frequently again at a late stage of cell-generation of the leaves. Mutations occurring during embryonic development should generally give rise to mosaic cotyledons; and although the majority of them do bear mosaic or yellow-inconstant leaves, sometimes they produce green or yellow cotyledons owing to excessive growth of the mutated or prototypic areas. Mutations occurring in a late stage of cell-generation produce fine green spots in the leaves. Although the yellow-inconstant-1 plants sometimes bear leaves without such fine green spots, these spots may be found in other leaves (IMAI 1930a). No luxuriant plants that did not have spots throughout the plant growth have so far been found. This characteristic distinguishes them from the ordinary yellow. In MIYAZAWA's yellow-inconstant strains, mutations occur more frequently during embryonic development than during a late stage of cell-generation. The mutability however is somewhat high during post-embryonic somatogenesis. On account of the low rate of mutability in the late stages of cell-division in the leaves, plants with no spotted leaves may occur in the yellow-inconstant-2, remaining throughout the plant ontogeny as false yellow, that is, crypto-yellow-inconstant-2. In the manifestation of characters, the green-variegated *chlorina* leaves of *Mirabilis* (CORRENS 1910) are quite analogous to the yellow-inconstant forms of *Pharbitis*. The mutable *chlorina* stock gives green patches and branches due to somatic mutations as well as seminal mutations in the progeny. As will be referred to later, green mutated tissues also occur in the leaves of the yellowy stock of *Pharbitis*. The xanthic seedlings of this plant have vivid yellow cotyledons, on

which green patches frequently appear through somatic mutations. An analogous case, probably, was recently reported by KONDÔ and KASAHARA (1933) in breeding experiments with *xantha* seedlings from heterozygous normal rice. The total observed (sum of all the available data) was 5301 green, 1433 *xantha*, and 170 variegated, or 76.7 percent, 20.8 percent, and 2.5 percent respectively. Gene *xantha* is mutable in this case, reverting to normal. The green stripes on the *xantha* seedlings (or variegated) are due to somatic mutations, only 10.61 precent of *xantha* being variegated. In *Antirrhinum* (BAUR 1924; KUCKUCK and SCHICK 1930), some mutable chlorophyll characters have been reported, for example, *albostriata*, *flavostriata*, and *bicolor*. On the leaves of these mutants, green patches and sectors due to recurring mutations occur on the white, yellow, or yellowish white background. I have obtained *xantha*-leaved seedlings in my culture of *Hordeum* through sporadic mutation. This form very frequently exhibits somatic mutations, giving green stripes and shoots. On selfing the stock, three forms, green, variegated, and xanthic seedlings appeared in various proportions. The occurrence of the green and xanthic is due to recurring mutations from xanthic-inconstant to its dominant normal and to its recessive, the constant xanthic, which is incapable of growing beyond the seedling stage. The *albovariabilis* form of *Capsella Bursa pastoris*, reported by CORRENS (1919), may be cited in connection with these instances. In his case, a series of graded genes with different modes of mutability seems to occur in giving that complex *albovariabilis* character (IMAI, in press c). Here mutations frequently take place reversibly between unstable white and unstable green. In a variegated strain of *Commelina* now under our observation, the case seems to be analogous to CORRENS'. ANDERSSON (1930) showed that in *Polystichum* mutations in certain ontogenical stages of gametophytes regularly arise from green to white and later from white to green. All these ever-sporting cases cited above seem to be due to the mutable genes that are responsible for the chlorophyll characters.

### Gene Yellowy.

The yellowy leaves, which are yellowish green at the young stage and have fine green mottling, later turn almost green. The

yellowy character made its appearance by mutation under my observation in 1928 (IMAI 1930b). The locus of the gene however has not yet been determined. In addition to the green mottled pattern, the leaves sometimes have pure green patches. On selfing, the yellowy gives rise to a mixed population in the subsequent generation. A summary of these observations is given in Table VII.

TABLE VII.

| Number of pedigrees | Green | Yellowy (not-green) | Total |
|---|---|---|---|
| 5 | 20 | 115 | 135 |
| Percentage | 14.8 | 85.2 | 100 |

The yellowy stock threw out 14.8 percent of green mutants. The yellowy seedlings, including mosaic, grew up as follows:

TABLE VIII.

| Y | Mosaic | | | | | | | Total |
|---|---|---|---|---|---|---|---|---|
| | P | Y+G | Y+P | Y+R | G+P | Y+G+P | Y+G+R | |
| 79 | 1 | 22 | 4 | 2 | 1 | 4 | 2 | 115 |

Y = yellowy, G = green, P = periclinal, R = reversal.

The full-grown plants contained 68.7 percent of plain yellowy, the remaining 31.3 percent being mosaics. Of these mosaics, one showed periclinal arrangement of tissues, that is yellowy-over-green. Other mosaics showed sectorial chimeras (yellowy and green) or compound chimeras with sectorial and periclinal situations (yellowy and periclinal; yellowy and reversal; green and periclinal; yellowy, green, and periclinal; yellowy, green, and reversal). The reversal branches are green-over-yellowy, while the green ones bear leaves entirely of green mesophyll. The green and reversal bud-variations gave rise mostly to green seedlings, segregating the yellowy ones as recessives; the periclinal branches produced mostly yellowy seedlings, as expected. These results indicate the mutable nature of gene yellowy. Genetically, gene yellowy has generally the same appearance as yellow-inconstant.

In crossing yellowy with normal, the $F_1$ plants obtained bore

green leaves, while in $F_2$, monogenic segregation occurred as shown in Table IX.

TABLE IX.

| Character of $F_2$ | Pedigree number | Green | Yellowy | Total | Percent of yellowy |
|---|---|---|---|---|---|
| | 9 pedigrees | 527 | —— | 527 | 0.0 |
| Green | 1 | 72 | 20 | 92 | 21.7 |
| | 4 | 73 | 24 | 97 | 24.7 |
| | 5 | 12 | 2 | 14 | 14.3 |
| | 6 | 35 | 11 | 46 | 23.9 |
| | 8 | 52 | 16 | 68 | 23.5 |
| | 11 | 31 | 9 | 40 | 22.5 |
| | 12 | 45 | 15 | 60 | 25.0 |
| | 13 | 80 | 23 | 103 | 22.3 |
| | 15 | 60 | 12 | 72 | 16.7 |
| | 16 | 70 | 13 | 83 | 15.7 |
| | 17 | 111 | 16 | 127 | 12.6 |
| | 18 | 89 | 38 | 127 | 29.9 |
| | 19 | 74 | 11 | 85 | 12.9 |
| | Total | 804 | 210 | 1014 | 20.7 |
| Yellowy | 1 | 8 | 16 | 24 | 66.7 |
| | 2 | 2 | 9 | 11 | 81.8 |
| | 3 | 3 | 9 | 12 | 75.0 |
| | 4 | 1 | 3 | 4 | 75.0 |
| | 5 | 0 | 7 | 7 | 100.0 |
| | Total | 14 | 44 | 58 | 75.9 |

The heterozygous green $F_2$ segregated out yellowy seedlings, including mosaics, with an average of 20.7 percent. This low ratio is attributed to reversions of gene yellowy. The yellowy $F_2$ gave rise to 24.1 percent green mutants, besides many mosaics.

## Variability of Flecked Flowers.

Gene flecked, which is located in the contracted chromosome (IMAI 1933), constitutes a set of multiple allelomorphs with normal and white-2b. White-2b results in a perfectly white flower, flecked in a white flower with fine flecks (stem green, frequently with color-

ed flecks), while normal manifests a self-colored flower in which anthocyanin pigment is distributed uniformly over the corolla. White-2b and normal are fully constant, whereas flecked is a mutable gene, giving at times self-colored mutants and mosaics of various chimerical forms. Since the variability in the flecked was described in an earlier paper (IMAI 1931), some additional facts recently observed will now be given.

The previous experiments were made with a stock presented by the late Mrs. T. SHIMIZU. Afterwards some cross-breeding experiments as well as some pedigree cultures were made with this strain. For the present, however, reference will be made only to the data collected in connection with the culture of the yellow-inconstant-2 stock received from Professor MIYAZAWA. This strain was originally used in the investigation of yellow-inconstant-2, and as the strain carries another mutable gene flecked, pedigree cultures were also observed for flecked.

The four plants raised in 1930 bore flecked flowers (Pl. XIII, fig. 1), and their offspring were cultivated the following season, the data being collected in Table X.

TABLE X.

| Pedigree number | Self-colored | Flecked | Mosaic | | | | | Total |
|---|---|---|---|---|---|---|---|---|
| | | | I | F+N | F+I | F+II | F+N+III | |
| 1 | 3 | 24 | 0 | 0 | 2 | 1 | 0 | 30 |
| 2 | 1 | 59 | 0 | 0 | 0 | 0 | 1 | 61 |
| 3 | 1 | 160 | 1 | 1 | 0 | 0 | 0 | 163 |
| 4 | 0 | 15 | 0 | 0 | 0 | 0 | 0 | 15 |
| Total | 5 | 258 | 1 | 1 | 2 | 1 | 1 | 269 |
| | | | | | 6 | | | |
| Percentage | 1.9 | 95.9 | | | 2.2 | | | 100 |

The culture contained 1.9 percent of self-colored mutants and 2.2 percent of mosaic variants, the remainder being flecked. The mosaic plants exhibited chimerical arrangements of tissues differing in phenotypes. Of six mosaic plants, one invariably bore self-colored flowers on its green stems (shown in Table X as Type I; Pl. XIII, fig. 3), one had green stems with flecked flowers and colored (colored means anthocyanin unless otherwise stated) stems with

colored flowers (Type F+N; for the stem and flower of Type N see Pl. XIII, fig. 2), two had green stems with flecked and self-colored flowers (Type F+I), another had green stems with flecked flowers and colored stems with fringed flowers (Type F+II; for the stem and flower of Type II, see Pl. XIII, fig. 4), while the last one comprised three forms, green stems with flecked flowers, colored stems with colored flowers, and green stems with flecked flowers having colored rays (Type F+N+III; for the stem and flower of Type III, see Pl. XIII, fig. 5). As will be dealt with fully later, the three simple mosaics, Types I, II, and III (the last two occurred in compound forms in this case), are periclinal chimeras. Type I is a flecked, covered with a self-colored epidermis; Type II, a flecked with a self-colored sub-epidermal tissue; and Type III, a flecked with a self-colored innermost component. Although the former two types were described in my previous paper, the last type is new. Out of six mosaic plants, one had green stems and invariably bore self-colored flowers. This plant is evidently a periclinal chimera, vegetative mutation having occurred early in its embryonic somatogenesis. By pronounced development of the mutated parts, the original flecked tissue had no chance to bloom flecked flowers. In other mosaic plants, the distribution of the original (flecked) and the mutated or periclinal parts differed widely, sometimes so much that one type existed as a small part of them, bearing one or two flowers, the predominant parts being the other type. With the exception probably of Type F+N, these forms of mosaics, while being sectorial chimeras, are at the same time periclinals. I raised succeeding generation of self-colored and flecked plants, including these six mosaics, the results being shown in Table XI.

Two self-colored plants segregated flecked as recessives and 10 flecked plants gave 4.7 percent self-colored, 6.0 percent mosaic, and 89.3 percent flecked, the respective proportions roughly agreeing with those obtained in the preceding generation. By totaling the results from the two generations, we have 23 self-colored, 29 mosaic, and 601 flecked (653 in all) from the flecked mothers, or 3.5 percent, 4.4 percent, and 92.1 percent respectively. In the flecked stock sent by Mrs. SHIMIZU, the data for three years comprised 227 self-colored, 38 mosaic, and 5737 flecked, or 3.8 percent, 0.6 percent, and 95.6 percent respectively (IMAI 1931). Although the self-colored mutants have nearly the same frequency of occurrence, a considera-

TABLE XI.

| Mother plant | Pedigree number | Self-colored | Flecked | Mosaic | | | | | | | | | | | Total |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | N | I | II | III | N+III | F+N | F+I | F+II | F+III | F+N+III | F+II+III | |
| Self-colored | 1—1 | 21 | 5 | | | | | | | | | | | | 26 |
| | 1—6 | 7 | 2 | | | | | | | | | | | | 9 |
| | Total | 28 | 7 | | | | | | | | | | | | 35 |
| Flecked | 1—2 | 3 | 28 | | | | | | | | | | | | 31 |
| | 1—3 | 2 | 12 | 1 | | | | | | | | | | | 15 |
| | 1—5 | | 22 | | 1 | | | | | | | | | | 23 |
| | 1—7 | 2 | 47 | | | | 1 | | | | | | | | 50 |
| | 1—8 | 1 | 65 | | | | | | | | | 1 | | | 67 |
| | 2—1 | 3 | 38 | | | | | 1 | 1 | 1 | | | | | 44 |
| | 2—2 | 3 | 61 | 1 | | | 2 | | 1 | | 2 | | | | 70 |
| | 2—3 | 1 | 17 | | 1 | | | | | | | | | | 19 |
| | 2—4 | 3 | 32 | 1 | 1 | 2 | | | | 2 | 1 | 1 | | | 43 |
| | 2—5 | | 20 | | 1 | | | | | | | | | | 21 |
| | Total | 18 | 342 | 3 | 4 | 2 | 3 | 1 | 2 | 3 | 3 | 2 | | | 383 |
| | | | | | | | | | 23 | | | | | | |
| | Percentage | 4.7 | 89.3 | | | | | | 6.0 | | | | | | 100 |
| Type I | 3— 1 I | | 18 | | | | | | 1 | | | | | | 19 |
| Type F+N | 3— 2 F | 1 | 4 | | | | | | | | 1 | | | | 6 |
| | 3— 2 N | 20 | 14 | | 1 | | | | | | 1 | | | | 36 |
| Type F+I | 1— 4 F | 1 | 39 | | 1 | | | | | | | | | | 41 |
| | 1— 4 I | | 5 | | | | | | | | | | | | 5 |
| | 1—10 F | 4 | 13 | | | 1 | | | | 1 | | | | | 19 |
| | 1—10 I | | 22 | | | | | | | | 1 | | | | 23 |
| Type F+II | 1— 9 F | | 29 | | 1 | | | | | | 1 | | 1 | 1 | 33 |
| | 1— 9 II | 22 | | | | | | | | | | | | | 22 |
| Type F+N+III | 2— 6 F | 5 | 36 | | | | 1 | | 1 | | | | | | 43 |
| | 2— 6 N | 19 | 7 | | | | | | | | | | | | 26 |
| | 2— 6 III | 3 | 43 | | | | | | | | 1 | | | | 47 |

ble difference is shown in the rate of occurrence of the mosaic variants, a much higher proportion being recorded in the new strain. This rather remarkable difference seems to me however to be due mainly to difference in the exactness with which observations were recorded, the new data being more accurate in the countings of the mosaics.

Some new combinations of mosaics appeared in this generation (see Table XI). Of the mosaic plants, Types I, II, and III exhibited almost invariably only one periclinal type each, while under Type N are collected plants with colored stems and self-colored flowers that were grown from seedlings with colored hypocotyls having some green parts. These plants are of mosaic origin, whose mutated parts had developed to a marked extent during somatogenesis. A special mosaic plant (Type N+III) was also recorded in a pedigree, No. 2—1, its mosaic origin being probably much more complicated. As the original part had no chance to bloom flecked flowers, the two mutated types developed into nearly the whole plant body. The other compound mosaics with sectorial and periclinal conditions, together with those cited above, are regarded as having developed from mosaic embryos.

Table XI contains records of the progeny of certain mosaic plants, the data being separate for each type of bud-variation. Since Types I and III gave results similar to those of flecked, they did not transmit their acquired characters to the offspring, whereas Types N and II, which resulted mostly in self-colored segregates, showed accompanying genic changes in their germ cells. In No. 1—9, the flecked flowers gave 4 mosaic and 29 flecked, while the fringed flowers bred true to 22 self-colored. In this case, double mutation, which occurred either singly or simultaneously, contributed to the origin of the fringed bud-variation.

Additional proof of the genetic constitution of the mosaics was obtained last summer when examining the progeny of certain mosaics that showed only one particular type predominantly, the data recorded being shown in Table XII.

The observation concerned the color of the hypocotyls in the seedling bed. Types N and II segregated into colored and green hypocotyls, the former of which will bear colored stems and self-colored flowers in the majority of cases, whereas most of the latter will have green stems and flecked flowers. Types I and II however

TABLE XII.

| Types of mosaic | Number of plants | Colored | Green including mosaic | Total |
|---|---|---|---|---|
| N | 2 | 128 | 34 | 162 |
| I | 4 | 12 | 238 | 250 |
| II | 2 | 215 | 60 | 275 |
| III | 3 | 9 | 214 | 223 |

gave mostly green hypocotyls, indicating that they had accompanied no genetic change in their germinal track.

The flecked plants at times bear flowers with broadly colored stripes, rarely the larger parts of the flowers being colored. When the flecked plants have more colored parts, resulting in at least a few colored flowers, the colored parts can almost always be traced to the base of the plant body; this is done easily and accurately when the colored flowers have also colored stems. This means that mutation occurred in the embryonic stage. As in the case of yellow-inconstant-1, mutation occurs rather frequently in the embryonic stage, rather rarely in post-embryonic somatogenesis and frequently again in the late stage of cell-generation. The fine flecks that occurred on the flecked flowers are believed to be due to mutations having taken place at a late stage of cell-generation of the corollas. The frequency curve of the vegetative mutation of gene flecked in the respective stages of plant ontogeny seems nearly to correspond to that in the yellow-inconstant-1. A significant difference however is observed in the frequency of the small spots occurring on leaves (in the yellow-inconstant) or on flowers (in the flecked), the spots in the flecked being more frequent than in the yellow-inconstant-1.

## General Considerations on Mutability.

To the comments I have made elsewhere (IMAI, in press c) on the mutability of certain labile genes, I wish to add here some remarks, especially on the genetic and non-genetic factors that affect it.

MUTATION TO ALLELOMORPHS WITH DIFFERENT RATES OF MUTABILITY. In another paper (IMAI, in press c), I discussed the recurrent mutation of mutable genes to allelomorphs with different

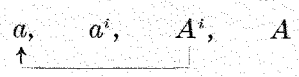
rates of mutability. As the results of such mutations, an allelomorphic group includes two or more inconstant genes, as in the cases of *Zea*, *Capsella*, *Malva*, *Polystichum*, and *Celosia*, as well as *Pharbitis*, for instance. DEMEREC (1929b) concluded that recurring mutations possibly occur in reversible ways in the unstable miniature complex, which is composed of genes miniature-alpha, miniature-beta, and miniature-gamma, all these constituting a group of multiple allelomorphs with normal. Although miniature-alpha and miniature-gamma also mutate to normal allelomorphs, which are highly constant, miniature-beta scarcely change in the same direction. In the duskish allelomorphs of *Pharbitis*, reversible changes take place between some grouping genes with different mutation frequencies.

INCONSTANT NORMALS. The stability of normal allelomorph, which is generally high, is in some cases low. In *Raphanus* (SÔ, IMAI and TERASAWA 1919) and *Chrysanthemum* (MIYAKE and IMAI, in press), recurring mutations occur from dominant red to recessive colorless. In other cases, the inconstant normals, which are mutated from recessive mutable genes, change to the original forms, the mutations recurring reversibly; namely, as exhibited in TERASAWA's *Celosia* (1922), EMERSON's *Zea* (1917), and IKENO's *Plantago* (1923). In chlorophyll variegation of CORRENS' *Capsella* (1919), inconstant normal (green) seems also to come in the same category. KONDÔ's *Oryza* (1927) and IKENO's *Erigeron* (1934) are examples of similar cases of recurring reversible mutations. IKENO, in studying inheritance in the apetalous mutant of *E. annuus*, a diploid parthenogenetic species, found a high rate of reversible mutation between *apetala* and *typica*. KONDÔ's investigation is based on his semi-sterile mutant rice, the character of which is highly labile, changing to the fertile condition. The mutated fertiles in turn frequently change to the semi-sterile state. Sometimes fertile rice gave all or almost all semi-steriles and *vice versa*. Some fertile ears of mosaic plants produced none or almost no fertiles in the progeny. These facts seem, at least in part, to be due to the periclinal nature of the ears or of the plants. In 1930, I obtained an individual showing mosaic with normal and contracted ears in a pedigree culture of a normal rice plant. Shoots of short stature bore contracted ears and had small glumes. Fertility was normal. The progeny of the contracted ears however showed only normal ears, the contracted character having disap-

peared through sexual propagation. Contracted ears are consequently regarded as periclinal, with the normal inner component covered by contracted epidermis, which results in a contracted phenotype of shoots and ears. A similar state of things is expected also in semi-sterile rice.

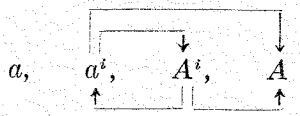
The inconstant normals of the various cases might be collected and expressed by a general formula for recurring mutations in the allelomorphic series somewhat as follows:

In *Raphanus* and *Chrysanthemum*,




In *Zea*, *Celosia*, *Plantago*, *Oryza*, and *Frigeron*,


In *Capsella*, possibly




In a hypothetical series of allelomorphs, $a$ would represents a constant recessive, $a^i$ an inconstant recessive, $A^i$ an inconstant normal, and $A$ a constant normal, the last being the prototype and the others its derivatives. Roughly speaking, the group of allelomorphs, including the two inconstant types, may theoretically be expressed by the series $a$—$a^i$—$A^i$—$A$, though frequently some of them are either absent or undetected. In some cases, the inconstant $a^i$ consists of many grouping genes (cf. IMAI, in press c). In *Antirrhinum*, BAUR (1926b) found an inconstant line, giving a few (about 2 percent) dominant *crispa* mutants.

AUTO- AND EXO-MUTATIONS. Since the mutable genes generally transform to their allelomorphs automatically, they are called auto-mutable. The labile plastids are either auto-mutable or exo-mutable. In the white-striped *Hordeum* (Sô 1921), variegation is indirectly due to a recessive gene carried by the stock, the gene stimulating the green plastids and altering them to white. As the mutant plastids are transmitted with their individuality to the offspring by the egg-cells, exhibiting maternal inheritance, the occurrence of white plastids is due to recurring exo-mutations of plastids,

and the gene carried by the variegated barley, although this is fully constant, acts as a stimulant in this case. In contrast to this, some variegated strains of *Oryza* exhibit auto-mutations of plastids, the inheritance being regardless of genic complex (cf. IMAI 1928). Thus recurring plastid mutations occur both automatically and transitively. Returning to the genic mutation, DEMEREC (1926, 1928a) pointed out that reddish-alpha mutates to wild-type only in the reduction divisions of heterozygous females in *Drosophila virilis*. No reversional mutations occurred in the somatic cells, in homozygous females and in males. Reddish-alpha therefore seems to be an exo-mutable gene, mutations occurring through stimulation of its allelomorphs, one of which is contained in a heterozygous condition in the females. The mutation from reddish-alpha to the wild-type occurs with equal frequency when heterozygous by yellow or by reddish-1. These allelomorphs therefore have the same potentialities for mutation. So far as I am aware, this is the only case in which recurring genic mutations do not arise automatically.

STIMULATING AND SUPPRESSING ALLELOMORPHS. The rate of mutability is either changed or unchanged according to whether the condition is homozygous or whether it is heterozygous. DEMEREC (1930, 1932b) showed statistically that the mutation rate of the rose-alpha gene of *Delphinium* is nearly the same in both cases, and that similar behavior was found in the case of the miniature-3 gamma of *Drosophila virilis*. In these cases, mutation frequency in the homozygotes was double that in the heterozygotes, in which latter the mutable genes were contained in a single dose.

The *globifera* form of *Antirrhinum* studied by BAUR (1924) frequently reverts to normal, as the result of which all the *globifera* sporophytes are more or less mosaic. As far as he has examined, the normal branches thus produced were homozygous for normal. The hybrids obtained by crossing *globifera* with normal were often homozygous. These facts seem to me to imply that the mutability of gene *globifera* is stimulated by its dominant normal allelomorph, thus bringing the heterozygous cells eventually to a homozygous condition, so that the stimulation of the normal allelomorph in the heterozygotes frequently causes them to be homozygous. In *Antirrhinum*, however, *globifera* is an auto-mutable gene and its mutability is stimulated by the presence of normal allelomorph in its heterozygous condition. A similar stimulation may possibly explain

CORRENS' *Capsella* (1919), LILIENFELD's *Malva* (1929), and KIHARA's *Celosia* (1932), in which the heterozygosity stimulates the mutability of the labile genes. Prior to these studies, EMERSON (1922) pointed out the very high rate of mutability in the heterozygous variegated ears of maize as compared with the homozygotes.

In these cases, or at least in some of them, the allelomorphs (normal or others) stimulate the mutability of the labile genes in the heterozygous state. Experimenting with *Antirrhinum*, HERTWIG (1926) made it clear that the mutation frequency in vegetatively reverting plants that were originally heterozygous for *globifera* varies according to differences in the allelomorphs of the 'Def-Serie', namely,

$$\frac{globifera}{globifera} = 100\%; \qquad \frac{nicotianoides}{globifera} = 64\%; \qquad \frac{chlorantha}{globifera} = 17\%$$

In the 'Def-Serie', only *globifera* is mutable, the others being constant. The percentages in the second and third formulas should be doubled because of the mutable *globifera* gene persent in a single dose in these cases. The corrected formulae are

$$\frac{globifera}{globifera} = 100\%; \qquad \frac{nicotianoides}{globifera} = 100\%; \qquad \frac{chlorantha}{globifera} = 34\%$$

The somatic mutability of *globifera* is therefore reduced by the suppression of allelomorph *chlorantha*, when this is contained heterozygously.

MODIFYING GENES. DEMEREC (1928b, 1929a) found in *Drosophila virilis* four genes, three dominant autosomal and one recessive, all of which modify the rate of mutation of mutable miniature. One called *M* works on miniature-alpha, and the others, *S–1*, *s–2*, and *S–3*, on miniature-gamma. Miniature-gamma, with the coexistence of either homozygous or heterozygous *S–1* or *S–3* or homozygous *s–2*, gives nearly all mosaic.

ONTOGENICAL INFLUENCES. The mutation rates of genes and plastids are greatly affected by the different stages of ontogeny. As pointed out in the preceding sections, in the yellow-inconstant-1 and the flecked of *Pharbitis*, mutation is frequently high during embryonic development, low during post-embryonic somatogenesis, and high (or very high) in the late stage of cell-generation in the formation of leaves and flowers. The yellow-inconstant-2 exhibits a very high rate of mutability during embryonic development, a little

high at the post-embryonic somatogenisis, and low in the late stage of cell-generation. In his sterile rice, TERAO (1917) found that reversion takes place at any stage of plant development, although with different frequencies for different ontogenic stages. Fertile spikelets due to reversions on their own part frequently occur, but the early somatic mutations, which include larger fertile parts, are of rare occurrence. Experimenting with mutable large-grained rice, the same author (TERAO 1922) also showed that mutation occurs at every stage of ontogeny, its frequency however being somewhat different at different stages. In his investigation of the variegated flowers of *Delphinium*, DEMEREC (1931) wrote: "Lavender-alpha gene has a high rate of mutability early and late in ontogeny, and has a low rate of mutability or is constant in the intermediate stages of ontogeny." The statistical rate of mutability however is calculated on the basis of its proportion to the observed total. ANDERSON and EYSTER (1928) attempted a survey of the variegated ears of *Zea* by classifying according to the mutation areas on the kernels of the ears. They arrived at the conclusion that the mutation in the ontogeny of the ears increases in the later cell-generations. Measuring the mutated cells of the variegated flowers of *Delphinium*, DEMEREC (1931) showed that the rose-alpha gene mutates at approximately the same rate during the last twelve cell-generations in the development of sepals. These data lead to the conclusion that the rate of recurring mutations generally differs for different stages of somatogenesis. Taking a short period of ontogeny into consideration, the mutation rate at times shows different frequencies, though frequently it may be nearly constant. The variation in the mutation rate at different stages of somatogenesis may differ widely in different species, and at times even in the same species. ANDERSSON (1930), in studying the variegated *Polystichum*, found definite stages of mutation in its gametophytic ontogeny. In *Drosophila virilis*, DEMEREC (1928a) showed that the ages of the females influence the mutability of the reddish-alpha gene. According to the investigations of ANDERSON and EYSTER (1928), in the variegated maize the position of the cell layers also affects the mutation rate. They found that mutations in the dark-crown are about 3.8 times as frequent as in the near-self mutations affecting equal areas, the mutation in the former occurring in tissues of ecto-histogenic origin and in the latter in those of endo-histogenic

origin (maize is probably a di-histogenic plant). In some variegated plants, a certain periclinal type of bud-variation is also more likely to occur than the others[1]. For instance, CORRENS' *Mirabilis*, TERASAWA'S *Celosia*, and KANNA's balsam more frequently bear bud-variations with mutated ecto-histogens than with mutated meso-histogens.

SOMATIC AND GAMETIC MUTATIONS. Mosaics caused by mutable genes are evidently due to somatic mutations. As stated before, the determination of individual mutants, at least some of them, is dificult whether they are due to gametic mutation or to somatic mutation. In *Plantago* (IKENO 1923), mutation however seems to be limited to the gametogenesis. ENOMOTO (1929) found non-glutinous grains on glutinous ears of *Oryza*, but no mosaic ears. With the iodine reaction he observed a few non-glutinous pollen-grains in the glutinous anthers. In this case also mutation seems to be restricted to the gametogenesis. Some ten years ago, Mr. K. KONDÔ of the Niigata Agricultural Experiment Station, sent me microphotographs of two endosperms mosaic for glutinous and non-glutinous. These mosaic endosperms indicate that mutation rarely occurs in triploid tissues: it hardly takes place in diploid tissues. According to DEMEREC (1926, 1928a), in reddish-alpha of *Drosophila virilis*, mutation is restricted to individual mutants, no mosaics being observed. He wrote: "The mutability of reddish-$\alpha$ was found to be limited to a very short period of development, which has been located at the maturation divisions of heterozygous females."

The above instances are regarded as cases in which the mutation is limited or almost limited to maturation divisions, or the individual mutants are due to gametic mutations. Strictly speaking, however, even in these cases we are still not in the position to deny the occurrence of mutation at late somatogenesis or prior to gametogenesis.

Except these cases, recurring mutations generally occur in somatogenesis, probably also in gametogenesis. However, we cannot definitely say whether or not mutations generally occur in gametogenesis. The seminal or individual mutants may partly be of mosaic origin. If mutation occurs in the early embryonic stage and the

1) In two earlier papers (IMAI 1927a; IMAI and KANNA 1927) we erroneously classified the variegated forms into two or three types, unaware of the foregoing facts.

mutated tissue is followed by a prevalent growth that dominates over the original tissues, it will result in such an individual mutant. Some mutants may receive the mutant gene at the late somatic cell-generation stage, prior to gametogenesis. The seminal mutants are generally a mixture of these, probably including some gametic mutations. In the present paper, therefore, in discussing the mutable genes of *Pharbitis*, I have treated them as seminal mutants and refrained from calculating their gametic mutability.

In *Adiantum* (ANDERSSON 1923), recurring mutation occurs in nearly all stages of the plant cycle, including sporogenous stage. According to her observations, in the four (out of nine) examples in which the outcome of all 64 spores from a sporangium was recorded, odd numbers of the two, green (segregates) and pale (segregates and mutants) types were produced—a result that seems to show that mutation also occurs at the second maturation division. Should a mutation or odd mutations take place at the 6th division of sporogenous tissue, it may result in such an odd proportion. The fact that albinotic ferns appear in the green areas of the prothallia in *Adiantum* indicates the possible occurrence of gametophytic or gametic mutation at a different stage.

In the miniature-gamma of *Drosophila virilis* (DEMEREC 1928b), however, mutation occurs only in the somatic cells, producing mosaics.

DIFFERENCE IN MALES AND FEMALES. In the mutable miniature of *Drosophila virilis*, DEMEREC (1932b) found that the rate of mutation is nearly the same in both sexes, or in other words, it is twice as high in females as it is in males, then the mutation rate of the latter must be doubled in their comparison, because although the females are diploid for the X-chromosome the males are haploid.

ENVIRONMENTAL INFLUENCES. EYSTER (1926) found that different climates (chiefly temperature) affect mutability in the variegated maize. DEMEREC (1932a) however, in dealing with the mutable miniature-gamma of *Drosophila virilis*, obtained a negative result in the temperature response.

As pointed out elsewhere (IMAI 1928), since plastid mutations in both variegated barley and rice hardly occur during such times as the egg-cell stage nor during early embryonic development, we cannot generally obtain any white shoots in these cases. This limits to a certain extent the deviation in frequency of the occurrence of

albinotic seedlings. The frequency however is much influenced by differences in sowing times (IMAI, in press b), in which difference, temperature may be the major factor.

As the gene is probably a complicated molecular unit, the lability of the gene means an unstable physico-chemical condition ready to change into another stable or unstable complex molecule. For this reason the mutability of these genes may be greatly affected by certain factors. They may be affected by changes in the chemical or physico-chemical cell components, such changes being induced by cellular genetic and non-genetic factors or ontogenic and environmental influences. Since the different genes may not respond alike to the same factors, the devious ways in which the mutable genes respond to the factors that affect them are so complicated that it seems difficult to collect data under a definite scheme.

## Types of Bud-Variations and their Origin.

In the yellow-inconstant-1 of *Pharbitis* are recorded two forms of periclinal chimeras, the one having a yellow 'skin' and a green 'core' and the other a green 'skin' and a yellow 'core' (IMAI 1930a), the former having the mutated endo-histogen and the latter the mutated meso-histogen, regardless of the nature of the ecto-histogen. In the present paper, I have called attention to two similar types of yellowy periclinals.

Studying the flecked *Pharbitis*, I (IMAI 1931) identified two chimerical types, one of which is flecked with a self-colored epidermis and the other flecked with a self-colored sub-epidermal tissue. U (1930) also distinguished two forms of periclinals in the pine-inconstant, while TABUCHI (unpublished data) identified the corresponding two delicate periclinals, both writers dealing with the morphological traits of this plant. In these cases, the two periclinals have different ecto- and meso-histogens, regardless of the nature of the endo-histogen. The third type of simple flecked periclinals, with mutated endo-histogen, however, is identified in the present paper. This is the first case in which the three simple periclinal bud-variations have been discriminated in a given character.

In experiments with *Celosia*, KANNA (1929), KOJIMA (1930), and

KIHARA (1932) identified two types of periclinals, the one having mutated epidermis and the other mutated sub-epidermal tissue. According to KANNA's observations on *Impatiens* (1926) and on *Mirabilis* (1933), and to DE HAAN's investigation (1931) on *Pisum*, the two types can also be discriminated by the flower color and other characters. As will be discussed later, the ontogenic origin of the plant bodies of most dicotyledonous species are tri-histogenic. If this is also the case with these plants, we should expect three types of simple periclinals as bud-variations. Recently KANNA (1934) published full accounts on the genetics of *Impatiens*, describing a subdued magenta flower with variegation that appeared as bud-sport. This may be identified as a periclinal with the mutated, self-colored endo-histogen.

EMERSON (1917) showed from his breeding experiments two periclinal types of vegetatively mutated kernels in *Zea*, the dark-crown having a mutated epidermis and the near-self a mutated sub-epidermal tissue. Maize is probably a di-histogenic plant. If so, we should then be able to obtain only these two periclinal forms from this plant.

The mutable nature of gene flecked is very useful also in investigations of bud-variation. In the flecked, the following periclinals are known:

| Character | Ecto-histogen | Meso-histogen | Endo-histogen |
|---|---|---|---|
| Type I; stem green and flower self-colored (Pl. XIII, fig. 3) | Self-col. | Flecked | Flecked |
| Type II; stem colored and flower fringed (Pl. XIII, fig. 4) | Flecked | Self-col. | Flecked |
| Type III; stem green and flower flecked with colored rays (Pl. XIII, fig. 5) | Flecked | Flecked | Self-col. |

In Type I, the flower color is as nearly intense as the normal self-colored mutants with colored stems. Type II blooms as a fringed flower, the corolla of which is self-colored in a dilute shade and with small colorless areas on its marginal parts, the whole surface being flecked with a normally intense color. In Type III, the characteristics are similar to the normal flecked, excepting the dilutely colored rays. Both Types I and III usually give flecked in their offspring, whereas in Type II the mutated character is inherited, the self-colored flowers predominating. Since these three forms are periclinals of heterogeneous tissues, their existence in sexual propa-

gation is only temporary. Anthocyanin color is present in the cell-sap of the tissues constituting the corolla, though the pigment is lighter in the inner tissue, whereas only the sub-epidermal cells, that is the outer tissue of cortex, are colored in the stem. Type I is flecked with self-colored ecto-histogen. It therefore bears green stems because their epidermis is always colorless. It blooms self-colored flowers because their epidermis is heavily colored, and it produces only flecked seeds. Type II is flecked with mutant meso-histogen. As anthocyanin pigment is present in the cells of the outer cortex the periclinal has colored stems. It blooms fringed flowers and gives self-colored as dominant segregates. The flowers are dilute in color, that is, the sub-epidermal tissue contains light anthocyanin pigment, the presence of which is due to the gene for self-colored, while the epidermis is colorless. The marginal white parts, or tissues, resulting in the fringing of the flower, are believed to to be formed by multiplication of the colorless ecto-histogenic cells. In maize, the dark-crown kernels have red crowns, while the near-self kernels have corresponding colorless parts, which are regarded as having an epidermal origin (EMERSON 1917). In *Celosia* (KANNA 1929, KOJIMA 1930, and KIHARA 1932), the so-called intermediate flowers, as KANNA calls them, have perianth with whitish marginal bands on both longitudinal sides. In their ontogenic origin, these marginal parts are supposed to differ from the central parts of the perianth. These examples, together with others which will be given later, may be indications that the origination of various parts of plant organs assumes at times different forms, namely, that the ecto-histogen develops into epidermis, extending certain marginal parts of the corolla in *Pharbitis*, the crown of the kernels in *Zea*, and marginal parts of the perianth in *Celosia*. In the cultivated varieties of *Rhododendron indicum*, we have a number of variegated flowers of different types. The variegated azalea frequently puts forth bud-variations of at least two types of flower coloration, the one a self-colored flower without any conspicuous 'red spot' on the upper lobes and the other a dilute flower with intense stripes and 'red spots'. In the second case, the dilute flower has white markings on their peripheral parts (Fig. 4). I have no genetical data for these bud-variations, but it seems very probable that the first type of bud-variation has self-colored ecto-histogen and the second type self-colored meso-histogen. The substance of the white periclinal mark-

ings in the dilute flower is also considered to be derived from the ecto-histogen. The red pattern spots appear usually on flowers of self-colored varieties. The self-colored flowers that appeared as bud-sports, however, generally have no such spots because the flowers do not accompany self-colored meso-histogens by which the 'red spots' are manifested. Since the azalea is propagated vegetatively, the new bud-variation has generally one mutated histogen. The large red part with 'red spots' of Fig. 4 however appears to have two mutated histogens. The origin of the red part is believed to be either due to mutation that occurred in ecto-histogen or a somatic rearrangement of the tissues during flower development. The flecking on fringed flowers of the azalea as well as *Pharbitis*, even on the fringed white parts, is due to mutation of gene flecked to its dominant allelomorph in a late stage of epidermal cell-generation. The color of the flecks is normal in intensity, same as that of the self-colored mutants. In the ordinary flecked flower, the flecks have nearly two grades of intensity, one normal and the other dilute (strictly speaking, three grades by the addition of flecks of Type III, which are faint in color). The intense flecks are due to mutations in the ecto-histogen and the dilute to those in the meso-histogen. Since the meso-histogen carries the mutant gene self-colored, the flecks on the dilutely colored background of the fringed flower are confined to the former. In the variegated flower of other general plants, the stripes, or variegation, occur at least in the two corresponding intensities owing to the mutated cells being located differently in the flower tissues. If the heterozygotic expression is intermediate in color, the number of grades in the intensity of the stripes will be doubled. This may be the case for instance in *Mirabilis*, *Portulaca*, and *Pharbitis purpurea*.

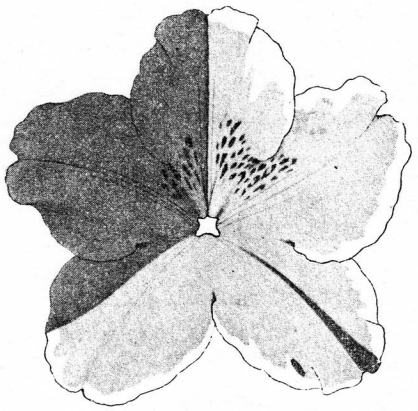


Fig. 4. Mosaic flower of the azalea. The black area represents the part in red, the dilute the pink, and the white the white or whitish.

Type III of *Pharbitis* is regarded as flecked with self-colored

endo-histogen. The stems are consequently green, the flower being flecked with faint-colored rays, while the progeny consists of flecked, including sometimes mutants. The faint-colored rays are due to the pigment in the innermost component of the corolla. Therefore the different parts of the corolla of this plant seem to be formed by multiples of different numbers of histogens, as shown in Fig. 5. If this suggestion is accepted, the larger part of the rays is only a part of what is formed in the corolla by the complete tri-histogens: such tissues may extend over the flower tubes. Being the endo-histogen self-colored, and the others colorless except the flecked parts, the rays alone should be colored in Type III. The rays are therefore faint in color, paler than the dilute color of the fringed flower, because the innermost tissue of the rays are lightest in color. The color of the rays however does not deve'op uniformly, so that the tissues that are developed from the three histogens do not seem to extend over the entire area in the innermost component of the rays.

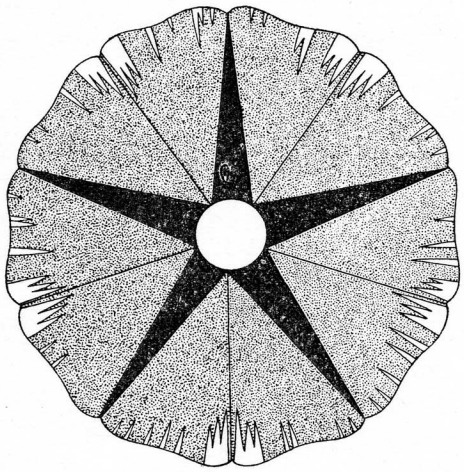


Fig. 5. The substance of the white parts are developed from ecto-histogen only, that of the dotted parts from ecto- and meso-histogens, and that of the black parts principally from a combination of the three histogens.

These three periclinals contain only a single, but different mutant histogen that produced by mono-heterogeneous bud-variation (compare Figs. 6B, 6C and 6D with Fig. 6A, pure flecked).

As stated before, bud-variations with colored stems and self-colored flowers sometimes occurred on flecked plants. This form of bud-variation should have the stem at least with self-colored meso-histogen and the flower with at least self-colored epidermis. The form therefore will have at least self-colored ecto- and meso-histogens, regardless of the nature of the endo-histogen. Whether the endo-histogen be formed from flecked tissue (Fig. 6E) or self-colored tissue (Fig. 6H), the stems at any rate should be colored and the

flowers self-colored. They probably cannot be distinguished from

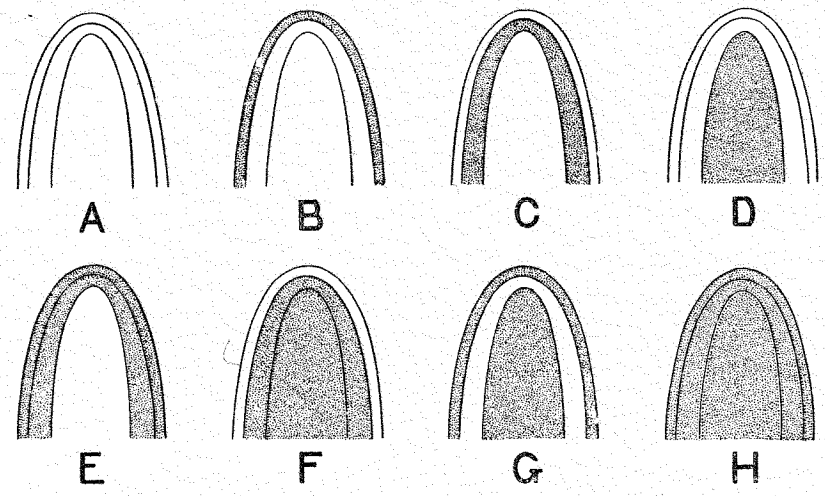



Fig. 6. A diagrammatic representation of homo- and heterogeneous growing points. The white parts are the original unchanged histogens and the dotted parts the mutated histogens.

each other by breeding tests nor from their phenotypes. Root-cutting will enable their distinction, but this method of propagation is unsatisfactory in *Pharbitis*. In the former periclinal form, the two mutated histogens are duplicated, whereas the latter form consists of homogeneous tissues or are produced as a homogeneous bud-variation. There will be another duplicated situation of the di-heterogeneous periclinal, in which both meso- and endo-histogens are of mutated tissues (Fig. 6F). This form will have colored stems and fringed flowers, probably similar to the mono-heterogeneous bud-variation with mutated meso-histogen (Fig. 6C). Lastly, we may have the third di-heterogeneous periclinal with mutated ecto- and endo-histogens, arranged alternately with the inserted original meso-histogen (Fig. 6G). The complete possible cases of bud-variations are as follows:

- Heterogeneous
  - Mono-heterogeneous
    1. Ecto-histogenic (Fig. 6B)
    2. Meso-histogenic (Fig. 6C)
    3. Endo-histogenic (Fig. 6D)
  - Di-heterogeneous
    - Duplicated
      4. Ecto-meso-histogenic (Fig. 6E)
      5. Meso-endo-histogenic (Fig. 6F)
    - Alternative
      6. Ecto-endo-histogenic (Fig. 6G)
- Homogeneous (Fig. 6H)

Since in post-embryonic somatogenesis, including the plumules of the embryos, three histogens are differentiated, the most probable cases of bud-variations to occur during this stage is mono-heterogeneous, only one mutated histogen being involved. The production of di-heterogeneous cases, due to combination of the two mutations in the same growing point, may be of rare occurrence, while that of the homogeneous ones may be rarer still. Actually, however, the origination of bud-variations observed in the flecked and other mutable pedigrees of *Pharbitis* can generally be traced to embryonic causes. The mode of tissue development differs in the embryonic ontogeny, the three histogens being as yet undifferentiated. Therefore, at this stage mutation may result in di-heterogeneous and homogeneous growing points of stems as well as in mono-heterogeneous. Further, bud-variations may sometimes change secondarily into other types through somatic rearrangement of the tissues during plant development. In any case, alternative di-heterogeneous bud-variations may have a slight chance of being produced. Should this compound periclinal form occur in flecked stock, it may bear green stems and self-colored flowers, probably similar to the mono-heterogeneous case with self-colored ecto-histogen. This mechanism of bud-variations applies to tri-histogenic plants in general.

Since, as stated elsewhere (IMAI 1931), bud-variations newly appearing on trees and shrubs as well as on perennial herbs that have been propagated vegetatively may in most cases be due to their origination from a mutated cell during somatogenesis, they are expected in the majority of cases to be mono-heterogeneous. Such mono-heterogeneous periclinals may at times give branches that revert to the original form as the result of somatic rearrangement of tissues during plant development. Such secondary bud-variations are not rare in the periclinal morning glory. In fruit trees especially, we sometimes observe cases in which recurrent vegetative variation reverts to the original form. Some authors regard these as reversible mutation. In such cases however we cannot very well arrive at conclusions without taking into consideration the mechanism of bud-variation formulated above. The occurrence of peach-nectarine mosaic fruits was pointed out by DARWIN (1882), their behavior and structure having been studied by IKENO and NOGUCHI (1929). The mosaic in this case may however be attributed to the mutable behavior of the gene responsible for the nectarine, the

recessive gene reverting very frequently to its dominant peach on the original nectarine stocks. Nectarine is therefore manifested by a labile gene in certain limited cases, though it is in general due to a constant recessive gene (BECKER 1923).

## Di- and Tri-histogenic Plants.

The theory of chimeras, first formulated by BAUR (1909) in the course of his studies in connection with the variegated *Pelargonium*, solved the riddle of the so-called graft-hybrids. With the exception of such writers as NOACK (1922) and his followers this theory was accepted by subsequent investigators. NOACK's own theory however does not readily explain the mechanism of periclinal chimeras found in various plants as well as my cases presented in the foregoing pages. Some writers, such as CHITTENDEN (1927), MASSEY (1928), IMAI (1930a), and others, who have fully discussed this question, are opposed to his theory. MASSEY in her anatomical studies on the development of some periclinally variegated plants, gave ontogenical evidence against NOACK's conception. According to her anatomical observations on *Arabis albida variegata*, *Euonymus japonica aurea*, and *E. japonica argentea*, the apex of these plants is composed of at least three histogens. Regarding the origin of the stems, she wrote "the stem apex seems to be composed of three or four distinct layers of cells surrounding an internal tissue", also that "the third layer shows undoubted division by periclinal walls in some places, while in others it is imposible to tell whether the inner tissue has arisen from this layer or independently." Her drawings, however, seem rather to point to differentiation of the three components in the stem apex as well as in the leaf apex. Long before these studies, DOULIOT (1890) showed that the majority of dicotyledons are of tri-histogenic origin and a few others of di-histogenic origin, and that the monocotyledons, with the exception of some tri-histogenic genera, are derived from di-histogens.

On the basis of data cumulated from genetical and anatomical observations, we may draw the conclusion that the plant body of the common dicotyledons arises from three histogens. For instance, in the leaves the ecto-histogen develops into epidermis (frequently also into the marginal edge of the lamina), the endo-histogen into the inner mesophyll, and the meso-histogen into the remaining part,

the outer mesophyll extending to the marginal tissue on both longitudinal sides of the leaf[1]. Therefore in the white-over-green (or yellow-over-green) chimeras, the white 'skins', which are developed from the meso-histogen, extend to the chlorophyllous tissue of the bordering parts on both longitudinal sides of the leaves in its development. In these chimerical leaves, the whole (or nearly the whole) of the white parts, except the epidermis, arise from the meso-histogen. In some variegated plants the leaves show a *medio-albinata* coloration. The leaves of some variegated strains of *Aucuba*, *Euonymus*, *Elaeagnus*, *Hydrangea*, *Spiraea*, and *Pelargonium* have white (or yellow) centres with wholly white mesophyll, the marginal parts being green. Excepting the outer mesophyll of the central white tissues, the white parts of these *medio-albinata* forms correspond to the green parts of the common white-over-green chimerical leaves. As the outer central mesophyll is also white in the *medio-albinata* forms, the arrangement of green and white tissues is not periclinal. The forms give green seedlings and, from root-cuttings, put forth albinotic shoots (BATESON 1921; CHITTENDEN 1925). On account of the disorderly arrangement of the green and white tissues, the *medio-albinata* forms have been a puzzle. In my opinion (IMAI 1928) the *medio-albinata* forms are periclinal chimeras with green 'skins' and white 'cores'. The breeding results have fully borne out my views. The white plastids in these forms seem to secrete a toxic substance which, on penetrating the periclinal cell-walls of the leaves, affect the outer mesophyll and result in *medio-albinata* leaves. In *Aucuba* we have variegated stocks, the leaves of which are sometimes variegated with yellowish spots in various amounts. The yellowish spots seem to be due to yellowish plasmone, which has the same physiological activity (secreting a toxic substance), and may be produced by recurring mutation. When the yellowish spots are situated in the cells just at the growing points, bud-variations will result; namely, the *medio-albinata* and *albo-marginata* forms, and rarely also as pure yellowish branches. The first form (*medio-albinata*) is the same as that above stated, the second form (*albo-marginata*) bears periclinal leaves with yellowish 'skins'

1) I wish to take this opportunity of correcting certain misprints in my earlier paper (IMAI 1931, p. 216, ls. 35—37). The faulty sentence is now corrected to:- "This suggests that the mesophyll of the longitudinal margins of the leaves is formed generally by the propagation of the sub-epidermal cells, and . . ."

and green 'cores'. In this case the inner green parts are apparently unaffected by the outer toxic yellowish tissues. Therefore the toxic substance scarcely diffuses into the inner tissues and into the lateral cells.

The variegated forms of *Polygonum orientale* carries a recessive gene which, stimulating the green plastids, alters them into white; or in other words, the variegation is due to the recurring exo-mutations of plastids (IMAI, in press a). The white branchlets, which sometimes appear on the variegated stocks, bear white leaves, the stipules of which however are variegated. I have never observed the so-called white-over-green or green-over-white forms, and therefore conclude that the plant body of *Polygonum* arises from two, ecto- and endo-histogens, instead of three. Judging from the variegated stipules of the white branchlets, the substance of the stipules (at least most of it) is believed to be developed from the ecto-histogen alone, which is variegated by recurring exo-mutations of plastids. The common white branchlets are therefore mono-histogenic bud-variations with the white endo-histogen, which constitutes all the mesophyll of the leaves.

According to the classic studies of DOULIOT (1890), all the tissues within the epidermis in certain dicotyledons, including *Polygonum amphibium*, are developed from a single histogen. MASSEY's drawings (1928) lead me to believe that *Veronica gentianoides variegata* is di-histogenic, the ecto-histogen constituting the epidermis, and the endo-histogen all the tissues underlying the epidermis. Some years ago CORRENS (1920) used this variegated form (*V. gentianoides albocincta*) in his genetic experiments. The variegated leaves of *Veronica* have white parts on both their longitudinal margins. He crossed the variegated with the green to ascertain the genetic nature of the *albocincta* form, but obtained only green seedlings in both reciprocal matings. My opinion is that these results can be explained by the fact that since *Veronica* has a di-histogenic composition, the general tissues within the epidermis are developed from the inner component, and that the marginal parts on both longitudinal sides of the leaves are developed from the dermatogen. The ecto-histogen constitutes the epidermis and also the marginal mesophyll of the leaves, and the endo-histogen develops into the other larger parts of the mesophyll. In such an origination of tissues, the *albo-marginata* form of *Veronica* is a

periclinal chimera with white ecto-histogen and green endo-histogen. Recently, NOHARA (1933) published his work on the genetics of *Sesamum indicum*, in which he relates having observed two mosaic specimens with green and *albo-marginata* leaves. From fruits borne on variegated portions, he obtained only green seedlings. The *albo-marginata* form of *Sesamum* may consequently be analogous to that of *Veronica* in the constitution of its tissues. If so, then *Sesamum* is also a di-histogenic dicotyledon.

In both *Veronica* and *Sesamum*, the chimerical, white-over-green leaves have white margins on their longitudinal sides, whereas its converse, the green-over-white leaves of *Polygonum*, have no corresponding green margin. In some cases, therefore, in *Polygonum* at least, all the mesophyll of the leaves is developed from the tissues underlying the epidermis. In a case of this kind we have neither an *albo-marginata* nor a *medio-albinata* form.

In monocotyledons, DOULIOT pointed out that in the majority of cases the plant body arises from two histogens. From an anatomical observation of *Chlorophytum comosum variegatum*, MASSEY (1928) came to the same conclusion, regarding it as di-histogenic. According to COLLINS (1922), the *albo-marginata* and *medio-albinata* types of *Chlorophytum* gave almost one type of seedlings in the progeny obtained by self-pollinations; that is, *albo-marginata* gave green seedlings and *medio-marginata* produced albinotic seedlings. In the light of MASSEY's anatomical view, COLLINS' breeding experiments force us to the conclusion that the two variegated forms of *Chlorophytum* are evidently periclinals, *albo-marginata* being a chimera composed of white ecto-histogen and green endo-histogen, and *medio-albinata* a chimera formed by reversal arrangement of two distinct tissues. Some genetic experiments have been made with the variegated forms of *Hosta japonica* by IKENO (1917), TERASAWA (1923), and YASUI (1929). The *medio-albinata* and *albo-marginata* forms of *Hosta* are di-histogenic periclinals and they gave the same breeding results as in *Chlorophytum*.

Except in *Polygonum*, in di-histogenic plants including both dicotyledons and monocotyledons, the periclinal forms have *albo-marginata* and *medio-albinata* leaves. In di-histogenic plants, the bud-variations may have the following components:

{Heterogeneous {1. Ecto-histogenic
{Homogeneous {2. Endo-histogenic

All the periclinals are mono-heterogeneous, they being much simpler than those in tri-histogenic plants.

---

In concluding this paper I wish to express my hearty thanks to Professor K. MIYAKE for his kind encouragement during the experiments, and also to the Foundation for the Promotion of Scientific and Industrial Research of Japan for the grant from which the expenses incurred in connection with the present investigation were partly defrayed.

## Summary.

1. Twenty (possibly more) mutable genes of *Pharbitis Nil*, situated in seventeen loci, are mentioned with short descriptions of their characteristics.

2. Further observations on the mutable contracted are described, with remarks on chimerical specimens.

3. Data amplifying the mutable nature of gene purple are presented.

4. All the mutable genes so far found in this plant exhibit both vegetative and seminal mutations. Recurring mutations occur from recessive to dominant; the genes come to prototypic normal, excepting in the willow, which generally mutates to maple, a recessive allelomorph to the normal.

5. Two yellow-inconstant stocks showed considerably different proportions of variants in their progeny. The yellow-inconstant-1 gave 5.2 percent green, 9.3 percent mosaic, and 85.5 percent yellow-inconstant, whereas the yellow-inconstant-2 gave 23.3 percent, 46.3 percent, and 30.4 percent respectively. These differences are due to their different mutabilities.

6. The mutation rate differs with different ontogenical stages of the two yellow-inconstant stocks; namely, in yellow-inconstant-1 the mutability is high in embryonic development, low in post-embryonic somatogenesis, and high again in the late stage of cell-generations of the leaves. In yellow-inconstant-2, however, it is very high, a little high, and low in the respective stages, varying differently in the mutation rate during the plant cycle.

7. Gene yellowy is mutable, reverting to its dominant normal.

Somatic mutations produce green, periclinal, and reversal leaves as in the yellow-inconstant. The yellowy give rise to 14.8 percent green individual mutants in the offspring, besides many mosaics.

8. The flecked pedigrees contain 3.5 percent self-colored, 4.4 percent mosaic, and 92.1 percent flecked, the former two being seminal and vegetative variants. Three periclinal forms with different arrangements of the mutated tissues were identified in connection with the bud-variations exhibited in the flecked stock. Many compound chimeras were obtained. The progeny tests of these chimerical forms throw some light on the mechanism of bud-variations.

9. Mutability and the factors affecting it have been generally considered, especially the following problems: mutation to allelomorphs with different rates of mutability, inconstant normals, auto- and exo-mutations, stimulating and suppressing allelomorphs, modifying genes, ontogenical influences, somatic and gametic mutations, difference in males and females, and environmental influences.

10. By developing and somewhat extending BAUR's theory of the chimera, the types of bud-variations have been discussed rather fully with special reference to the mechanism of the periclinals. In flecked *Pharbitis*, three mono-heterogeneous periclinals were identified.

11. The number of histogens in dicotyledonous and monocotyledonous plants is discussed in connection with variegated leaves, especially *albo-marginata* and *medio-albinata* forms.

## LITERATURE CITED.

ANDERSON, E. G. and EYSTER, W. H. (1928): Pericarp studies in maize. III. Genetics 13: 111–120.

ANDERSSON, I. (1923): The genetics of variegation in a fern. Jour. Genetics 13: 1–11.

——— (1930): Variegation in three species of ferns (*Polystichum angulare, Lastraea atrata* and *Scolopendrium vulgare*). Zeitschr. f. ind. Abst.- u. Vererbungsl. 56: 115–201.

BATESON, W. (1921): Root-cuttings and Chimaeras. II. Jour. Genetics 9: 91–97.

BAUR, E. (1909): Das Wesen und die Erblichkeitsverhältnisse der "Varietates albomarginatae hort." von *Pelargonium zonale*. Zeitschr. f. ind. Abst.- u. Vererbungsl. 1: 330–351.

——— (1924): Untersuchungen über das Wesen, die Entstehung und die

Vererbung von Rassenunterschieden bei *Antirrhinum majus*. Bibliotheca Genetica 4 : 1–170.

BAUR, E. (1926a): Untersuchungen über Faktormutationen. I. Zeitschr. f. ind. Abst.- u. Vererbungsl. 41 : 47–53.

——— (1926b): Untersuchungen über Faktormutationen. II–III. Zeitschr. f. ind. Abst.- u. Vererbungsl. 41 : 251–258.

BECKER, J. (1923): Ein Beitrag zur Züchtung der Pfirsiche. Gartenwelt 27 : 274–275, 285–286. Rev. in Bot. Abst. 11 : 1169.

BLAKESLEE, A. F. (1920): A dwarf mutation in *Portulaca* showing vegetative reversions. Genetics 5 : 419–433.

CHITTENDEN, R. J. (1925): Studies in variegation. II. Jour. Genetics 16 : 43–61.

——— (1927): Vegetative segregation. Bibliographia Genetica 3 : 355–442.

COLLINS, E. J. (1922): Variegation and its inheritance in *Chlorophytum elatum* and *Chlorophytum comosum*. Jour. Genetics 12 : 1–17.

CORRENS, C. (1910): Der Übergang aus dem homozygotischen in einen heterozygotischen Zustand im selben Individuum bei buntblättrigen und gestreiftblühenden *Mirabilis*-Sippen. Ber. deutsch. Bot. Gesellsch. 28 : 418–434.

——— (1919): Vererbungsversuche mit buntblättrigen Sippen. I. Sitzungsber. Preuss. Akad. Wiss. 34 : 585–610.

——— (1920): Vererbungsversuche mit buntblättrigen Sippen. III–V. Sitzungsber. Preuss. Akad. Wiss. 6 : 212–240.

DARWIN, C. (1882): The variation of animals and plants under domestication. London.

DEMEREC, M. (1926): Reddish—a frequently "mutating" character in *Drosophila virilis*. Proc. Nat. Acad. Sci. 12 : 11–16.

——— (1928a): Mutable characters of *Drosophila virilis*. I. Genetics 13 : 358–388.

——— (1928b): The behavior of mutable genes. Zeitschr. f. ind. Abst.- u. Vererbungsl. Suppl. 1 : 183–193.

——— (1929a): Genetic factors stimulating mutability of the miniature-gamma wing character of *Drosophila virilis*. Proc. Nat. Acad. Sci. 15 : 834–838.

——— (1929b): Changes in the rate of mutability of the mutable miniature gene of *Drosophila virilis*. Proc. Nat. Acad. Sci. 15 : 870–876.

——— (1930): Washington, Carnegie Institution Year Book, No. 29 : 41.

——— (1931): Behaviour of two mutable genes of *Delphinium Ajacis*. Jour. Genetics 24 : 179–193.

——— (1932a): Effect of temperature on the rate of change of the unstable miniature-3 gamma gene of *Drosophila virilis*. Proc. Nat. Acad, Sci. 18 : 430–434.

——— (1932b): Rate of instability of miniature-3 gamma gene of *Drosophila virilis* in the males and in the homozygous and in the heterozygous females. Proc. Nat. Acad. Sci. 18 : 656–658.

DOULIOT, M. H. (1890): Recherches sur la croissance terminale de la tige des Phanérogames. Ann. Sci. Nat., Bot. 11 : 283–350.

EMERSON, R. A. (1917): Genetical studies of variegated pericarp in maize. Genetics 2 : 1–35.

EMERSON, R. A. (1922): The nature of bud variations as indicated by their mode of inheritance. Amer. Nat. 56: 64-79.

ENOMOTO, N. (1929): Mutation of the endosperm character in rice plant. Jap. Jour. Genetics 5: 49-72. In Japanese.

EYSTER, W. H. (1926): The effect of environment on variegation patterns in maize pericarp. Genetics 11: 372-386.

HAAN, H. DE (1931): Contribution to the genetics of *Pisum*. Genetica 12: 321-440.

HERTWIG, P. (1926): Ein neuer Fall von multiplem Allelomorphismus bei *Antirrhinum*. Zeitschr. f. ind. Abst.- u. Vererbungsl. 41: 42-47.

IKENO, S. (1917): Inheritance of variegation. Rigakukwai 14: 566-571. In Japanese.

——— (1923): Erblichkeitsversuche an einigen Sippen von *Plantago major*. Jap. Jour. Bot. 1: 153-212.

——— (1934): A mutant of *Erigeron annuus*. Botany and Zoology, Tokyo 2: 25-32. In Japanese.

IKENO, S. and NOGUCHI, Y. (1929): Ein Beispiel der Pfirsichnektarinenchimäre in Japan. Jour. Coll. Agri., Tokyo Imp. Univ. 10: 305-312.

IMAI, Y. (1925a): Genetic studies in morning glories. XV. Bot. Magazine, Tokyo 39: 43-52. In Japanese.

——— (1925b): Genetic behaviour of the willow leaf in the Japanese morning glory. Jour. Genetics 16: 77-98.

——— (1927a): The vegetative and seminal variations observed in the Japanese morning glory, with special reference to its evolution under cultivation. Jour. Coll. Agri., Tokyo Imp. Univ. 9: 223-274.

——— (1927b): A genetic study of green-variegated yellow leaves in the Japanese morning glory. Jour. Genetics 17: 329-348.

——— (1928): A consideration of variegation. Genetics 13: 544-562.

——— (1930a): Studies on yellow-inconstant, a mutating character of *Pharbitis Nil*. Jour. Genetics 22: 191-200.

——— (1930b): Description of the genes found in *Pharbitis Nil*. Genetica 12: 297-318.

——— (1931): Analysis of flower colour in *Pharbitis Nil*. Jour. Genetics 24: 203-224.

——— (1933): Linkage studies in *Pharbitis Nil*. III. Zeitschr. f. ind. Abst.- u. Vererbungsl. 66: 219-235.

——— (In press a): An apparently simple inheritance of variegation in *Polygonum orientale*. Jour. Genetics.

——— (In press b): Variation in the rate of recurring plastid mutations in *Hordeum vulgare* caused by differences in the sowing times. Genetics.

——— (In press c): Recurrent reversible mutations in the duskish allelomorphs of *Pharbitis Nil*. Zeitschr. f. ind. Abst.- u. Vererbungsl.

IMAI, Y. and KANNA, B. (1927): On the variability of a white-eared form in *Amarantus paniculatus*. Genetics 12: 242-252.

IMAI, Y. and TABUCHI, K. (1933): A provisional map of the yellow chromosome of *Pharbitis Nil*. Zeitschr. f. ind. Abst.- u. Vererbungsl. 66: 236-244.

KANNA, B. (1926): On the inheritance of balsam. Bot. Magazine, Tokyo 40: 599-619. In Japanese.

KANNA, B. (1929): On a mutable strain of *Celosia cristata* L. Bot. Magazine, Tokyo 43:407-413. In Japanese.
——— (1933): On the striped flowers of *Mirabilis Jalapa*, L. Jap. Jour. Genetics 8:165-178. In Japanese.
——— (1934): Genetic studies in *Impatiens Balsamina*. Jour. Coll. Agri., Tokyo Imp. Univ. 12:421-477.
KIHARA, H. (1932): Genetische Studien an gestreiften Sippen von *Celosia cristata* L. Agriculture and Horticulture, Tokyo 7:1003-1026. In Japanese.
KOJIMA, H. (1930): The inheritance of flower-color in a race of *Celosia cristata* L., blooming in mosaic color. Bot. Magazine, Tokyo 44:328-351. In Japanese.
KONDÔ, M. (1927): Über die Ergebnisse der Pedigree-Zucht der semisterilen Reispflanzen. Ber. ÔHARA Inst. f. landw. Forsch. 3:275-289.
KONDÔ, M. and KASAHARA, Y. (1933): Über eine hellgelbe junge Reispflanze, die aus semisteriler Pedigree abstammte und bald abstarb. Nogakukenkyu 21:75-94. In Japanese.
KUCKUCK, H. and SCHICK, R. (1930): Die Erbfaktoren bei *Antirrhinum majus* und ihre Beziechung. Zeitschr. f. ind. Abst.- u. Vererbungsl. 56:51-83.
LILIENFELD, F. A. (1929): Vererbungsversuche mit schlitzblättrigen Sippen von *Malva parviflora*. I. Bibliotheca Genetica 13:1-214.
MASSEY, K. (1928): The development of the leaves in certain periclinally variegated plants. Jour. Genetics 19:357-372.
MIYAKE, K. and IMAI, Y. (In press): A periclinal stock with variegated flowers in *Chrysanthemum sinense*. Zeitschr. f. ind. Abst.- u. Vererbungsl.
MIYAZAWA, B. (1929): On the inheritance of the "Matusima"-variegation in the Japanese convolvulus. Jap. Jour. Genetics 4:167-184. In Japanese.
——— (1932): On the Matusima-variegation in Japanese morning glory. Bull. Miyazaki Coll. Agri. a. Forestry 4:111-125. In Japanese.
NOACK, K. L. (1922): Entwicklungsmechanische Studien an panaschierten *Pelargonium*. Jahr. f. wiss. Bot. 61:431-458.
NOHARA, S. (1933): Genetical studies on *Sesamum indicum*, L. Jour. Coll. Agri., Tokyo Imp. Univ. 12:227-386.
SCHIEMANN, E. (1926): Eine Mutation in der *graminifolia*-Sippe von *Antirrhinum majus*. Zeitschr. f. Abst.- u. Vererbungsl. 41:53.
SÔ, M. (1921): Inheritance of variegation in barley. Jap. Jour. Genetics 1:21-36. In Japanese.
SÔ, M., IMAI, Y. and TERASAWA, Y. (1919): On the abnormal inheritance in *Raphanus sativus*. Bot. Magazine, Tokyo 33:21-30. In Japanese.
TERAO, H. (1917): On reversible transformability of allelomorphs. Amer. Nat. 51:690-698.
——— (1922): On the mutation in the "large-grained" rice, especially regarding the frequency of allelomorphic transformation. Jap. Jour. Genetics 1:127-151. In Japanese.
TERAO, H. and U. N. (1930): Studies on the appearance of mutation in the morning glory. Jap. Jour. Genetics 6:195-198. In Japanese.
TERASAWA, Y. (1922): Vererbungsversuche über eine mosaikfarbige Sippe von *Celosia cristata*, L. Bot. Magazine, Tokyo 36:75-83.
——— (1923): Über die Vererbung von *Funkia ovata*. Jap. Jour. Ge-

netics 2:13-21. In Japanese.

U, N. (1930): On the recurring mutation of pine type of morning glory. Jap. Jour. Genetics 6:199-202. In Japanese.

YASUI, K. (1929): Studies on the maternal inheritance of plastid characters in *Hosta japonica* ASHERS. et GRAEBN. f. *albomarginata* MAK. and its derivatives. Cytologia 1:192-215.

## EXPLANATION OF PLATE XIII.

Fig. 1. Flecked.
Fig. 2. Self-colored.
Fig. 3. Flecked with self-colored ecto-histogen.
Fig. 4. Flecked with self-colored meso-histogen.
Fig. 5. Flecked with self-colored endo-histogen.


1
2
3
4
5