A GENETIC STUDY OF GREEN-VARIEGATED YELLOW LEAVES IN THE JAPANESE MORNING GLORY.

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(With Five Text-figures.)

CONTENTS.

Introduction.

In looking over the old books on the Japanese morning glory, *Pharbitis Nil*, we often find the illustrations of half green and half yellow[1] leaves growing on one plant, and also green and yellow leaves occurring alternatively on another. These forms of chimaera are known to us as "bicoloured leaf[2]" or "Matsushima leaf[3]" (Fig. 1). Besides these there is recorded and illustrated in our old books what is called "Matsushima-variegated" (Fig. 2), in which irregular bicoloured mosaics

[1] By "yellow," we mean not pure yellow, but yellowish green or *chlorina* type.

[2] In the classical books published in the years during the Bunka and Bunsei eras (1804–1829), this nomenclature was usually adopted, but afterwards this type was generally known as Matsushima leaf.

[3] In Iwasaki-Kanyen's *Sômoku-Sodategusa* (1833, 1–2, p. 31), an old Japanese book on plant breeding, the term "Matsushima leaf" was first applied to the yellow leaf having green variegation of the Japanese morning glory. This term seems, as some authors stated, to have derived from the analogous diversity in features of the bicoloured patterns of the plant to the multiform islets of Matsushima, a famous scenic place in North Japan.

Fig. 1. The so-called Matsushima specimen pictured in *Asagao-Sô* (1817). This and most other pictures of Matsushima forms given in the old books are more or less exaggerated and not quite true to nature.

Fig. 2. The so-called Matsushima-variegated leaf pictured in *Tohi-Syûkyô* (1857).

occur on the leaves. The writer had an incidental opportunity of studying these peculiar bicoloured leaves. The results of experiments showed that the mosaics are due to somatic mutations occurring on the yellow-leafed plants.

A Historical Sketch.

The *Kadan-Asagao-Tsû* (1815), one of the oldest books on the Japanese morning glory, gives a coloured illustration of a specimen called "Chônotomo[1]." This specimen has normal leaves and dilute red flowers with crapy corollas and is evidently one of the so-called Matsushima-leafed individuals. The illustration represents a sectorial chimaera of the green and yellow parts, on most of whose leaves bi-coloration is found, just one-half being green and the other yellow. In *Kengyû-Hinruizukô*, which was compiled about the same time as the above-cited book, we find a coloured sketch of a Matsushima-leafed specimen illustrated as a bicoloured leaf. The leaves of this specimen are all bicoloured in complicated mosaics of green and yellow. It blooms as dilute red. Another Matsushima-leafed specimen (Fig. 1), with maple leaves and blue double flowers, is represented in *Asagao-Sô* (1817). This specimen is a sectorial chimaera, all of the leaves displayed in the coloured picture being half green and half yellow. Two years later, *Kengo-Hin* (1819) was published. In it we find also a sketch of the Matsushima type bearing normal leaves and crapy corollas. A note reads that there are varieties of the Matsushima type, some normal and others narrow, with such corollas as funnel-shaped, cherry-flower-like, crapy, gentian-like, etc. According to this statement, the Matsushima type enjoyed great popularity at this period, and there were cultivated many races which appeared by new combinations of factors on natural crossing. Many more examples of the Matsushima and Matsushima-variegated leaves are recognisable when we study such books as *Asagao-Hanaawase* (1853), *Santo-Itchô* (1854), *Tohi-Syûkyô* (1857), etc., published during the eras of Kaei and Ansei (cf. Fig. 2). Such relatively prevalent cultivation of the Matsushima forms, however, came to an end after the Restoration of the Imperial power in 1868. The Japanese morning glory was much admired and cultivated in and after the era of Meiji (1868–1912) for the peculiar shapes and forms of its flower and foliage, but the existence of bicoloured leaves almost slipped from the memory of our cultivators.

[1] "Chônotomo" may be literally translated as "friend of butterfly."

THE RECOGNITION OF THE TYPE, ITS BEHAVIOUR AND HYBRIDISATION.

The writer produced by selection a pedigree strain bearing yellow cordate leaves and dilute magenta flowers, designated by GY, from among a mixed population obtained from a seedsman. On hybridising this race with NN, a pure pedigree strain characterised by green *Nandina* leaves and dilute purple corollas, an F_2 consisting of green and yellow leaves in the usual ratio, as was expected, was reared by tracing the following generation of the green-leafed hybrids. Contrary to expectation, two individuals making a sectorial chimaera of green and yellow colorations on the foliage were observed among the F_2 sister plants. These unexpected plants bore some entirely green leaves and some yellow ones, and still the leaves occupying their seats on the border of two forms showed longitudinal bicoloration. Unfortunately the writer made no further observations upon the yellow sisters of F_2, beyond noticing these two abnormal plants. Owing to the lack of special observation on the F_2 seedlings, where their occurrence might have been suggested at an earlier stage, the appearance of these two chimaeras came as a surprise. The key to the abnormal behaviour of the yellow leaf in this cross was first given by inspecting the F_3 seedlings, and confirmed by subsequent observations upon the successive stages of the plant growth. The appearance of green-yellow-chimaeras may be accounted for as a form of vegetative mutation. The writer, encouraged by a new fact bearing upon the yellow leaf, carefully examined the progeny of one of the parental pedigree strains. The yellow-leafed strain, GY, had been selfed since 1922, and gave one green exceptional plant in 1923. In 1924 close observation was made of the seedling bed of this strain, and some cotyledons having green variegation were found among the yellow seedlings. The observations, though few in number, are given in Table I.

TABLE I.

Pedigree	Green leaf	Green-variegated yellow leaf	Yellow leaf	Total
A	0	2	8	10
B	0	0	3	3
C	0	1	5	6
Total	0	3	16	19

From the green plant obtained in the preceding year, however, we had a mixed progeny of green and yellow seedlings, with one green-variegated yellow specimen among them. Table II contains the result thus gained.

TABLE II.

Pedigree	Green leaf	Green-variegated yellow leaf	Yellow leaf	Total
D	27	1	6	34
		7		
Expected from a 3 : 1 ratio	25·5	8·5		34

The segregation of green and yellow leaves practically corresponds to the usual simple ratio, indicating the green rogue to be heterozygous for the yellow leaf. Notwithstanding the fact that the plant is thus a heterozygote there is no evidence of hybridisation. The plant, therefore, must be regarded as a mutant, of which we shall give full discussion in a later section.

THE RESULTS OF F_2, F_3 AND F_4.

The true nature of the yellow leaf in this cross being brought, for the first time, to light in 1924, the F_2 records, as indicated in Table III, were straightforward except for the occurrence of the two sectorial chimaeras. Such chimaeras may be identified with the so-called Matsushima leaves, of which we gave a brief account at the beginning of this paper in calling the attention of the reader to the classical literature.

TABLE III.

Pedigree	Green leaf	Matsushima leaf	Yellow leaf	Total
1	45	2 (*A* and *B*)	9	56
2	23	0	7	30
3	33	0	10	43
Total	101	2	26	129
		28		
Expected from a 3 : 1 ratio	96·75	32·25		129

An F_3 was produced from 17 green and 4 yellow F_2 plants, 21 in all. The progenies of green F_2 are classified into two groups, viz. those breeding true to green, and those throwing the recessive forms. The recessives in this case do not all consist of pure yellow leaves, but in addition to these there often appeared green-variegated yellows and rarely green-yellow-chimaeras, the so-called Matsushima leaves. Four yellow-leafed F_2 gave the yellow offspring, which, however, contained a few green rogues, besides some Matsushima-variegated yellows and two Matsushima leaves. The summary of the F_3 data will be found in Table IV. The number of greens which bred true to the type was 9, while 8 threw the yellow forms. In all the observations of the latter, we found 1 sectorial Matsushima and 6 Matsushima-variegated leaves among

TABLE IV.

Character of F_2	Pedigree number	Green leaf	Matsushima leaf	Matsushima-variegated leaf	Yellow leaf	Total
Green leaf	Total of 9 families	113	0	0	0	113
	1	113	1 (*C*)	2	19	135
	4	10	0	0	3	13
	7	32	0	0	7	39
	10	3	0	0	1	4
	14	18	0	2	4	24
	15	20	0	2	4	26
	16	5	0	0	1	6
	19	15	0	0	5	20
	Total	216	1	6	44	267
Yellow leaf	9	1	1 (*E*)	2	6	10
	13	2	1 (*D*)	3	18	24
	18	0	0	2	5	7
	21	0	0	0	2	2
	Total	3	2	7	31	43

51 yellow segregates, which thus form only 19·10 per cent. of the total, while the ratio was 21·71 per cent. in F_2. Both percentages are thus lower than the normal expectation. In the four yellow pedigrees we counted 3 greens in a total of 43, the rest being yellow. From these yellow-leafed F_3, an F_4 generation was grown on, the data obtained being represented in Table V.

There were observed actually a few green-variegated yellow seedlings, but they were included in the yellow class in the table, because the record was taken in the seedling bed. Three green seedlings appeared among 111 F_4 of yellow pedigrees; a proportion which is rather low compared with F_3.

TABLE V.

Pedigree number	Green leaf	Yellow leaf (contains Matsushima forms)	Total
9—1	0	5	5
—2	0	19	19
—3	1	26	27
Total	1	50	51
13—1	1	18	19
—2	0	2	2
—3	0	4	4
—4	0	3	3
—5	1	3	4
—6	0	7	7
—7	0	15	15
—8	0	6	6
Total	2	58	60
Grand total	3	108	111

MATSUSHIMA-LEAFED INDIVIDUALS AND THEIR PROGENY.

In Fig. 3, one of the Matsushima-leafed plants, which appeared in F_2, is diagrammatically shown in a spiral form indicating the phyllotaxy. Divergence of phyllotaxy in the Japanese morning glory is two-fifths. The plant *A*, of the two chimaeras, has a mixture of about three-fifths of green, and *B* only one-fifth of its original yellow, the rest being converted into green tissue. Also the yellow tissue of the latter disappeared from

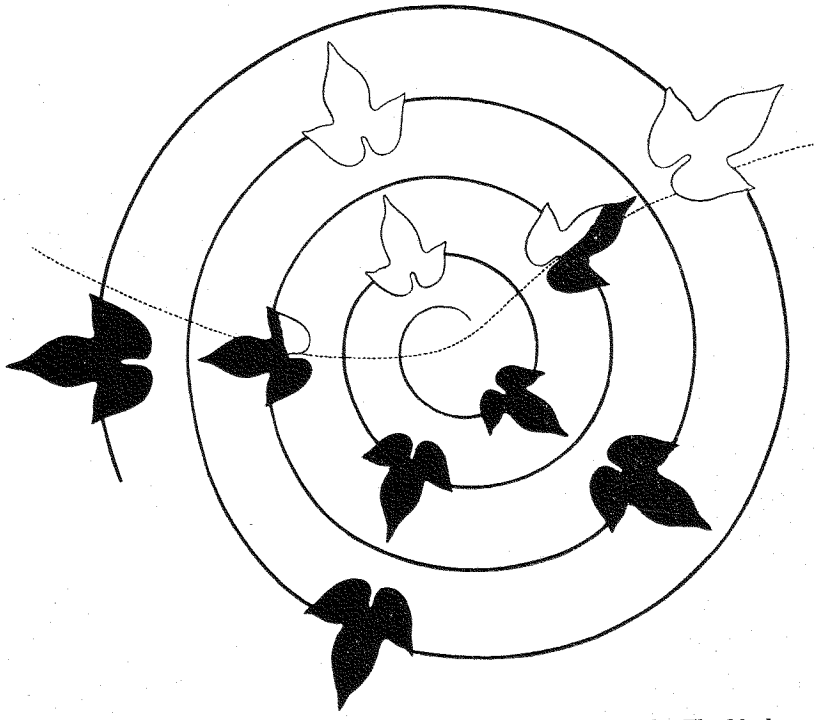

Fig. 3. The spiral diagram of a sectorially bicoloured specimen (plant *A*). The black part of leaves represents green and the white part yellow.

the upper part of the main stem, and afterwards the plant grew to be a green-leafed one. In both cases bicoloration coincided with phyllotaxy, and it very clearly shows the nature of sectorial chimaera. The Matsushima-leafed plants, *A* and *B*, gave few offspring, which, however, seem to be enough to represent the genetic nature of chimaeras, the results obtained being as given in Table VI.

Thus the two chimaeras segregated almost in the same manner; from the green part we obtained a small number of yellows among green-

TABLE VI.

Plant and part, from which seeds were obtained	Green leaf	Matsushima-variegated leaf	Yellow leaf	Total
A. Green part	7	0	3	10
Yellow part	1	2	5	8
B. Green part	9	1	2	12
Yellow part	0	0	3	3

leafed sisters, while the yellow part gave yellow leaves including a few Matsushima-variegated. Besides, one green individual appeared in the progeny of the yellow part of the plant *A*. The difference in the progenies between the green and the yellow parts of the chimaera-plants indicates a genetic differentiation of the much-discussed character, or in plain words the fact is accounted for by the transformation of the yellow leaf factor into a green leaf one in a vegetative tissue. The progenies of the green part of the plants *A* and *B* contained 6 yellows among 22 individuals, in a recessive proportion. From this ratio of segregation we can recognise that the yellow plants owe the genetic nature of the varied green part to one factor change. Consequently the tissues of the green part are heterozygous in constitution. Hence, the green-leafed F_3 plants should be composed of homozygotes and heterozygotes at the usual ratio. The raising of F_4 brought about the expected results as shown in Table VII.

TABLE VII.

Pedigree number	Green leaf	Yellow leaf (contains Matsushima forms)	Total
3	8	0	8
1	10	2	12
2	5	1	6
4	7	2	9
5	15	5	20
Total	37	10	47

As the records were taken by observation on the seedling bed, the green-variegated plants were counted together with the yellows. Out of five families, excepting the one which may be regarded as homozygous, all segregated yellow seedlings approximately in the expected ratio, the actual proportion being 21·28 per cent.

A further statement will be made of three Matsushima leaves, which appeared in F_3. One of them, the plant *C* obtained in pedigree 1 was a sectorial chimaera, while the plant *D*, which appeared in pedigree 13, turned out to be a periclinal chimaera. The former plant contains about

three-fifths of mutating tissues, as is diagrammatically shown in Fig. 4, and on the upper part of the main stem the tissue was entirely replaced by the prevalent green cells. The progenies of the green and yellow parts are recorded in Table VIII.

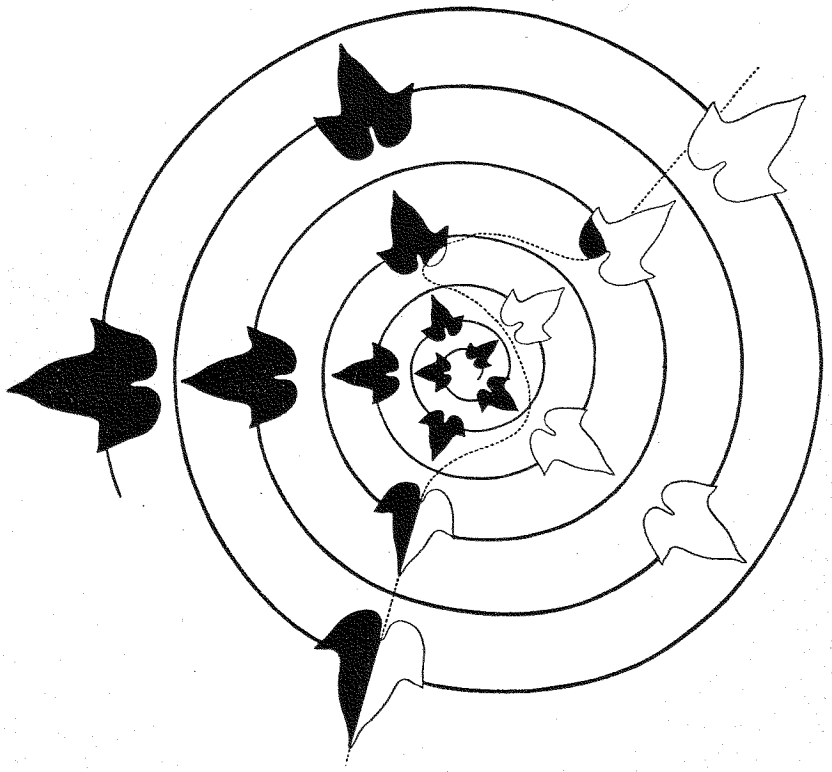

Fig. 4. The spiral diagram of another sectorially bicoloured specimen (plant *C*).

TABLE VIII.

Part from which seeds were obtained	Green leaf	Yellow leaf (contains Matsushima forms)	Total
Green part	10	3	13
Yellow part	0	5	5

As the census was taken by observing the seedlings, the green-variegated yellows were reckoned with the yellows. The result showed, as was expected from previous experience, that the green part of a

sectorial chimaera was produced by a factor mutation of yellow into green, giving heterozygous tissues. The point *D* bore yellow leaves of a similar pattern with a yellowish green patch at the centre. A microscopic examination of the section of the leaves showed a periclinal constitution of green and yellow layers, a yellow skin covering over a green core. Owing to the regular development of the plant tissues, the periclinal condition was maintained throughout the plant growth, as far as the writer's observation went. As the sub-epidermal layer of the plant tissues is composed of yellow cells, the subsequent generation might be expected to breed true or nearly so. The progeny of this periclinal plant was raised, and our actual examination of 21 individuals showed all to be yellow seedlings, except one green. A word may be added on the origin of this rogue. As was already stated, our yellow-leafed plants sometimes threw green individuals, and the green just mentioned is to be considered as having the same origin as the other greens.

Of the figures given in the old books, the one illustrated in *Tohi-Syûkyô* (1857) seems to be a periclinal chimaera of the Matsushima leaf, the yellow leaves, as in the plant *D*, having a greenish centre.

The plant *E* was an extreme type of the Matsushima leaf, and practically it is a green plant. A statement on this plant will be given in the next section.

GREEN MUTANTS.

The four F_2 yellows, as shown in Table IV, produced, in addition to yellows, a few greens. These green rogues may be considered as a result of either gametic mutation or somatic transformation of a factor in the tissues of the mother plants. In addition to these possibilities may be suggested another which will account for the origin of green rogues. Somatic mutation may occur in the embryonic stage or early growth of the seedling, and the mutated tissues, overcoming the prototypic yellow part in development, may produce greens. This is no mere speculative suggestion. Actually the plant *E*, which appeared in pedigree 9 of F_3 (see Table IV), had only a yellow patch on the lobe edge of its cotyledon, the rest of which, including the growing point of the bud, was entirely green and the plant afterwards grew up to be a green specimen. Of course the plant *E* was characterised by somatic mutation in the early stage of its embryonic development, and the prevalent growth of green tissues formed by the multiplication of a mutated cell must have caused the appearance of such a practically green plant. An

attempt was made to raise the F_4 generation with three F_3 specimens, two pure green rogues and one practically green one (plant E) to ascertain their genetic conditions. The results obtained are shown in Table IX.

TABLE IX.

Pedigree number	Green leaf	Yellow leaf (contains Matsushima forms)	Total
9—*a* (*E*)	25	7	32
13—*a*	8	2	10
—*b*	17	4	21
Total	50	13	63
Expected from a 3 : 1 ratio	47·25	15·75	63

The segregation of greens and yellows agreed nearly with a 3 : 1 ratio, the recessive proportion being 20·63 per cent. Whatever the origin of the greens which appeared among the yellows might be, their genetic constitution thus proved to be heterozygous, as far as the experiments were concerned. The average proportion of the green rogues among the Matsushima-lined yellow families is about 3·5 per cent. (the data, with which this calculation is made, are collected from Tables I, IV and V).

Hagiwara (1926) has lately reported a case of yellow leaves habitually throwing some greens. Such green-throwing yellow leaves may be identified with our Matsushima-lined yellows, but Hagiwara did not describe the occurrence of bicoloured leaves, notwithstanding the fact that he reared families from yellows which contained a high proportion of green rogues. As the writer has stated in other papers (Imai, 1924, 1925), the eversporting phenomena of the Japanese morning glory observed in several cases of different characteristics accompany vegetative variation in the same direction, as is also the case in the other plants such as de Vries' striped snapdragon, Emerson's variegated corn, Blakeslee's dwarf *Portulaca* and so on. Hagiwara's case, therefore, seems to be in need of thorough examination from this point of view. He also suggests the possibility of the occurrences of mutation in the green leaf factor to its recessive yellow allelomorph, but the evidence offered by him seems to stand on a rather insufficient basis.

Behaviour of the Factor for the Matsushima-Lined.

From the experimental data so far obtained, we can safely conclude that green on the Matsushima forms is due to the difference of a single

factor from the other prototype in its genetic relationship. And the only difference between the Matsushima and Matsushima-variegated leaves lies in the size of area of the green mutated tissues, and the area is determined by the point of plant growth, at which mutation occurred. When, for instance, it takes place in a cell of the growing point of the stem, a Matsushima leaf may come out, but in other cells, the production results in the Matsushima-variegated leaves. An analogy is seen in the variegated *Mirabilis* (*variegata*), with which Correns (1909) worked. *Variegata* is a green-variegated yellow- (*chlorina*) leafed specimen, variegation occurring always so definitely in every individual that Correns regarded the form to be due to a *variegata* factor. In the Japanese morning glory, however, the characteristic of *variegata*, or Matsushima variegation appears in only a few individuals among apparently pure yellow sisters. To what cause is this difference due? The writer is of the opinion that the *variegata* of *Mirabilis* is due to a factor which gives mutations of yellow into green as it occurs habitually in the Matsushima-lined yellows of the Japanese morning glory, the only difference between these two plants lying in the frequency of mutability. Vegetative mutations occur so much more frequently in *Mirabilis* than in the Japanese morning glory, that in the former all the individuals obtained were variegated. And both *variegatas* give a few green mutants.

The next problem to be solved is the allelomorphic relationship of green, Matsushima yellow and ordinary yellow. In *Mirabilis*, Correns presumed two factors to account for their relationship, besides a ground factor, because the three forms do not constitute a series of multiple allelomorphs. In the Japanese morning glory, the green leaf always behaves as a simple dominant to the yellow, and the Matsushima and yellow forms are transmitted as a recessive to green. But we have no data for determining the further relationship. The study of the genetic behaviour of Matsushima forms suggests some difficulty, because only a few plants assume the characteristic of variegation though they all carry a factor for the type. If the genetic cause of the Matsushima-lined is due to a modification of the yellow leaf factor, we should have a triple series of allelomorphs of green, Matsushima-lined yellow and ordinary yellow. But if it is not a case of multiple allelomorphs we may have to presume another factor governing the mutability, besides the yellow leaf factor. The final determination will be left for a future study.

The Mutability of the Matsushima Factor.

We had only a few green-variegated leaves among the offspring of the Matsushima-lined yellows, most of the rest being apparently pure yellows. Of 59 (3 green mutants omitted from this calculation) offspring of the Matsushima-lined yellows (the data taken from Tables I and IV), we recognised 12 Matsushima forms, *i.e.* about 20 per cent. In short, one-fifth of the yellows carry mutated cells in their bodies, while the rest remain in their prototypic condition. In these calculations, we neglected the green mutants. If these are produced by vegetative mutation in their embryonic stage after fertilisation, and so grow up to be apparently green ones owing to the predominant growth of the mutated tissues, the value may become larger than the figure above given. The calculation is based on so meagre a number that we cannot attach much importance to it, but it is to be recognised that the conditions are different from the case of Correns' *Mirabilis*. In *Mirabilis*, the green-variegated yellows stand at 100 per cent., while they form about 20 per cent. in the Japanese morning glory. We are presuming that the present cross is concerned with the monohybrid segregation of allelomorphs for the green and the Matsushima-lined yellow leaves, though there is no definite evidence for this. If, however, the case is dihybrid and contains some ordinary yellows the frequency as calculated above must increase.

On the Segregating Ratio.

The segregating ratio of the green and the yellow leaves in the offspring of the heterozygous greens or the green part of the Matsushima leaves is generally lower than expected. When we make a total of such data (the data taken from Tables II, III, IV, VII and IX) we have 431 greens and 109 yellows (the latter including the Matsushima forms) out of the total number of 540. The proportion of the yellow leaves is only 20·19 per cent., where we expect 25 per cent. The yellow specimens being generally somewhat weaker than the green, it is no rare matter in ordinary cases to observe some discrepancy in the segregating ratio of the green and the yellow leaves. The above low ratio may be partly accounted for by such a cause, but we must not lose sight of the ever-sporting nature of Matsushima-lined yellows, owing to which the number of the yellow segregates will tend to decrease.

THE ORIGIN OF THE MATSUSHIMA-LINED.

The fact that the occurrence of Matsushima or Matsushima-variegated leaves is found in the books published one hundred and ten years ago[1] shows that the characteristics of those variations were recognised in the beginning of the extensive cultivation of this plant. The yellow leaf, which is now very commonly found in our gardens, however, was as rarely visible at that time as the Matsushima forms. Though the phenotype of the leaf colour of the Matsushima-lined yellow was yellow, it was genetically different from the common yellow. Now we wish to determine whether the old yellow leaves are the true yellows or the Matsushima-lined ones.

The existence of the Matsushima forms gives a chance of obtaining the yellow leaves, because the former throw the latter in a high proportion in their offspring. The authors of the old books unanimously denied any certainty in the breeding behaviour of the Matsushima forms. For instance, the author of *Kadan-Asagao-Tsû* (1815) states that the green-variegated leaves appear irregularly among the offspring of the yellows, and the mono-chromatic leaves are precariously produced from the green-variegated yellows. The author of *Kengo-Hin* (1819) arrived at the same conclusion. He says in effect: "The Matsushima forms are produced occasionally from yellows, but the seeds of the Matsushima leaves do not always give similar offspring, only a few Matsushima forms being obtainable among many sisters." His argument went a step further towards recognition of the fact that the Matsushima characteristics are inheritable. In the identification of the old yellows we must keep such breeding behaviour of the Matsushima forms in mind.

In both *Kadan-Asagao-Tsû* (1815) and *Kengyû-Hinruizukô* (1815), there are some descriptions of the yellow and the Matsushima leaves, such as the following:

	Yellow leaf	Matsushima leaf
Kadan-Asagao-Tsû	1. Normal leaf, funnel-shaped flower with dilute blue corolla	1. Normal leaf, dilute red crapy corolla
Kengyû-Hinruizukô	2. Normal leaf, dilute red crapy corolla	2. The same as above
	3. Normal leaf, funnel-shaped flower with dilute red corolla	
	4. Normal leaf, funnel-shaped flower with dilute blue corolla	

The yellow leaf, which was illustrated in *Kadan-Asagao-Tsû*, is the same as one of the three specimens listed in *Kengyû-Hinruizukô*, and

[1] Since the importation of the seeds of the Japanese morning glory from China, over a thousand years have elapsed, but the period during which the plants were grown extensively with abundant variations extends little over one hundred and ten years.

further, both books give similar Matsushima leaves. The Matsushima and the yellow leaves were rarely found in those days, and these traits seem to have made their first appearance some years before the publications of these books. Two years later, *Asagao-Sô* (1817) and *Teichû-Asagao-Fu* (1817) were printed. The former has a list of over five hundred forms[1], among which we find several yellow and Matsushima leaves as follows:

Yellow leaf	Matsushima leaf
1. Maple leaf, split flower with dilute blue-spotted corolla (specimen not listed, but given in painting)	1. Maple leaf, split flower with spotted corolla
2. Maple leaf, split flower with spotted corolla	2. Maple leaf, irregularly split flower with dilute blue corolla
3. Normal leaf, funnel-shaped flower with intense blue corolla	3. Normal leaf, funnel-shaped flower with spotted corolla
4. Normal-formed, variegated leaf, funnel-shaped flower	4. Normal leaf, funnel-shaped flower

The description, though sometimes inadequate in the original, is accurate enough for the present consideration. In the books of 1815, the Matsushima and the yellow forms have been confined to the normal-shaped leaves, but now in this book we have two maple specimens each in yellow and Matsushima leaves, indicating a progressive step in evolution. And in *Teichû-Asagao-Fu* there is found a yellow-leafed specimen of a narrow willow leaf with narrowly split, dilute red flowers, which was less common in those times. The following table will give a comparison between the yellow and the Matsushima leaves as listed in *Kengo-Hin* (1819):

		Yellow leaf	Matsushima leaf
Leaf form	Normal	Present	Present
	Maple	,,	,,
	Pear	,,	Absent
Flower form	Funnel-shaped	,,	Present
	Crapy corolla	,,	,,
	Cherry-blooming	,,	,,
	Peacock-blooming	,,	Absent
	Gentian-blooming	,,	Present

In these three tables, we find a very close relationship in variation between the yellow and the Matsushima leaves. This circumstance may allow us to conclude that these yellows are not ordinary ones, which are expected invariably to breed true to the type, but that they are Matsushima-lined ones, which may produce some greens and Matsushima forms in their offspring.

The yellows are now almost confined to the ones which breed true

[1] The author of this book tells of the existence of over seven hundred forms at that time.

to the type, and the Matsushima-lined yellows have become very rare in our gardens. The cultivation of the Matsushima forms attained its zenith of prosperity in the period of Kaei and Ansei eras (1848–1859). But their cultivation fell off suddenly in the era of Meiji (1868–1912), giving place to the ordinary yellows. As stated in the foregoing pages the yellow leaf which first appeared was that of the Matsushima-lined. So, we may suggest the following two possibilities with regard to the phylogenetic relationship between the green, the Matsushima-lined yellow and the ordinary yellow:

(1) Green → Matsushima-lined yellow
Green → Ordinary yellow

(2) Green → Matsushima-lined yellow → Ordinary yellow

The third possibility of green → ordinary yellow → Matsushima-lined yellow, must be rejected in view of the evolutionary history mentioned above. But we cannot decide which of the other two possibilities is the true one, because we have no key to solve this problem.

TRICOLOURED LEAVES.

In the Japanese morning glory, variegation usually means a green or yellow leaf with white or pale creamy patches, which condition is transmitted as a simple recessive to the monochromatic condition. From the fact that a variegated leaf is found as one of the popular characteristics in such books as *Asagao-Tsû* (1815) and *Kengyû-Hinruizukô* (1815), we may regard this as rather an old character. According to such literature, however, there was yet no variegated yellow leaf, the variegated leaves being all green. The occurrence of the variegated yellow leaf was first recorded in *Asagao-Sô* (1817). In *Kengo-Hin* (1819) we are given some description of this form. The author of this book when treating of the ornamental importance of the variegated yellows, said that this form is the best of the yellow leaves, which means practically the best among all leaves found in the Japanese morning glory. From this statement, we see that the cultivators of those days set a high value upon this type.

From the fact that variegation occurs in the yellow leaves, it is but natural that we find tricoloured leaves of green, yellow and white, or in other words, variegated Matsushima leaves. In the recent books on the Japanese morning glory, the term "tricoloured leaf" is often mentioned, but few have made actual observations on them. The author of *Asagao-Mizukagami* (1818) first pointed out a tricolour type, but he gave

Fig. 5. A tricoloured leaf pictured in *Tohi-Syûkyô* (1857).

no picture. In a list given in *Asagao-Hanaawase* (1853), we find tricoloured leaves. The oldest figures (Fig. 5) of this type are found in *Santo-Itchô* (1854) and *Tohi-Syûkyô* (1857). The writer has not yet actually observed a tricoloured leaf, and an experiment to obtain such a leaf by crossing is in progress.

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SUMMARY.

1. The so-called Matsushima and Matsushima-variegated leaves of the Japanese morning glory are bicoloured in green and yellow. The study of such peculiar forms shows that they are yellow specimens in which occurs a somatic variation to green.

2. The Matsushima forms are obtainable among the progeny of yellows in the average proportion of about 20 per cent. Such yellow leaves cannot be phenotypically distinguished from the ordinary yellows, which always breed true, but difference lies in their genotypes.

3. The Matsushima-lined yellow leaf behaves as a recessive to green, and segregation takes place practically in a 3 : 1 ratio, where the recessive may contain some Matsushima and Matsushima-variegated leaves.

4. Three sectorial and one periclinal Matsushima leaves were observed in the hybrid progeny. The green part of the Matsushima leaves showed itself to be heterozygous for green and yellow, so the green tissues owe their occurrence to a single factor change. The periclinal plant had yellow sub-epidermal layer and gave almost all yellow offspring.

5. The Matsushima-lined yellows give a few green mutants in their offspring. A breeding experiment shows that they are heterozygotes of green and yellow.

6. By a close study of old literature we are able to conclude that the old yellow leaves are of the Matsushima-lined type, and not ordinary yellow. The common yellow, however, is a variety recognised rather recently.

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