ON MENDELIAN FACTORS IN RADISHES

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INTRODUCTION

This contribution deals with some of the factors of different varieties of summer radishes, as well as the black winter radishes, relating to foliage and to color, shape and size of the roots. No observations have been made on the factors of the flowers and fruits.

I aim to analyze these characters as to their behavior in segregation and recombination and the possible occurrence of new combinations caused by compound characters.

There seems to have been a lack of extensive investigations of this highly recommendable group of varieties. The various colors, shapes, and times of development of the different varieties of radishes offer a very promising and inviting field for researches in genetics.

BARKER and COHEN (1918) worked with the varieties Scarlet Box, Scarlet Globe and White Icicle, of which the coefficients of variability were especially studied. TROUARD-RIOLLE (1920) studied the F_1 generations from reciprocal crosses between *Raphanus Raphanistrum* and *R. sativum* and found that individuals thus produced were in all cases more

vigorous than the parents. FROST (1923) found also that wild and cultivated races of radish are very heterozygous. He noticed especially self-sterility in wild races. HEGI (1921) presents an extensive description of the various forms of the radish in general.

Though the writer was able to isolate various pure lines of radish varieties which are mentioned later on, he wishes to emphasize that there is a great deal of misconception as to the names of the different varieties as sold by most seed growers. For example, the variety Triumph, obtained from several growers, could be easily distinguished in at least three different homozygous types. Though all have white roots with reddish stripes and spots, the roots of one strain were much stouter, the red on the roots darker-tinted, bearing vigorous foliage, whereas the roots of another strain, being of the same age, grown at the same time in the same soil, were smaller, more tender, the stripes lighter red and foliage smaller. A third type showed roots and foliage similar to the first-described, but the stripes were much lighter. Similar observations can be made on almost any "commercial variety"; this, however, is no hindrance in studies of variability and heredity, as soon as pure lines have been established.

METHODS

The biology of the flower has been studied sufficiently by various investigators. On account of their relatively large size they are much visited by bees and other Hymenoptera, as well as some Lepidoptera; consequently, the flowers have to be isolated carefully by paper bags. Emasculation must take place the evening before the flowers will open. There is no difficulty in handling the large flowers. As the pistil and pollen are ripe at the same time, and but few flowers of certain individuals are slightly proterogynous, they can be readily self-pollinated with a small brush, though fertilization does not occur in all ovules and therefore only a small percentage of seeds results, whereas cross-pollination has been always successful in all varieties with which the author worked. In order to be certain of having pure lines, a very large number of self-pollinations should be made and the offspring carefully studied, as not the slightest dependency can be placed upon the reliability of seed growers.

The flowers were usually pollinated in the morning, though a few were pollinated in the afternoon, but no differences could be observed in the number of seeds obtained. Great care has been taken in isolating either single flowers or entire plants.

Radish plants whose roots were studied were replanted at the desired places after they had been deprived of the leaf tops in order to prevent an excessive amount of transpiration which could not be replaced by the water supply of the root system damaged in transplanting.

RESULTS

There are many forms of radishes, which show various characteristics in height, width of foliage, nervature, earliness, and color and shape of the roots. As far as all these characteristics are concerned no new characters occurred with exception of one which was due to hybridization. For example, the shape of the roots in every homozygous strain depended upon one single factor, whether these roots were narrow, long, round or

FIGURE 1.—Comparable sections of epidermis and a portion of the cortex of (1) Long Red, (2) Triumph, (3) Early Red, (4) Early White.

oval. For the study of these characters a large number of experiments was made, though they are not all mentioned in this publication. In all material with which I worked, the first hybrid generation always clearly showed intermediates with respect to size of foliage and shape of roots.

There are, however, several interesting facts concerning the color factors, especially when white and red varieties are crossed.

In red radishes the red pigment, belonging to the anthocyanin group, is to be found in the epidermis and two or three outer layers of the cortex (figure 1, 1). In the Triumph (figure 1, 2) some cells are red, and others lack anthocyanin. Figure 1, 3, is of Early Red whose red epidermis and outer cortex cells are smaller than in Long Red. Figure 1, 4, is of Early White, containing no red pigment in its roots.

The less complicated crosses will be considered first, followed by more complicated ones. In all cases reciprocal crosses were exactly alike in the F_1 , and in the F_2 and F_3 the same segregations and recombinations could be noticed.

Long Red + Icicle

Of both varieties the roots are long, though in Long Red they are somewhat more delicate than in the Icicle. The shape of F_1 individuals resembles more that of the Icicle than of the Long Red. The color is entirely violet to sometimes violet-purplish, from the base to the apex of the root. Violet color of the root has been noticed in all the other crosses when white and red varieties were used.

A large number of F_1 plants, self-pollinated, produced in F_2 the results shown in table 1.

TABLE 1

YEAR	RED HOMOZYGOTE		VIOLET HETEROZYGOTE		WHITE HOMOZYGOTE	
	Number	Percent	Number	Percent	Number	Percent
1920	70	20	189	54	91	26
1921	52	26	104	52	44	22
1922	149	23	305	47	130	20

It is apparent that these color differences behave as in a monohybrid. Though in most crosses with other species or varieties of plants it is difficult or impossible to recognize the heterozygotes, it is easy to distinguish the heterozygotes between red and white radishes on account of their violet color.

Although, as stated previously, there are slight differences in shape of root between Long Red and Icicle, it is not easy to determine a proper ratio in the F_2 as to the shape of these radishes, because fluctuations in both varieties are such that boundaries between the two can not be drawn. This cross should work out as a dihybrid, and no doubt it does, although differences of shape were so difficult to observe that no attempt was made to separate the individuals definitely into homozygous and

heterozygous classes. They were noted as "behaving apparently like a monohybrid."

All the violet heterozygotes segregate in the F_3 and F_4 as they do in the F_2 .

Early White $\times$ *Early Red*

These two varieties with short foliage and flat round roots differ from each other only in color, the former having white roots and the latter red ones with a white point.

All F_1 individuals developed violet roots with white points and short foliage. Their behavior in F_2 was like that of a monohybrid in every respect, as shown in table 2.

TABLE 2

YEAR	NUMBER OF RED ROUND	NUMBER OF HETERO- ZYGOTUS VIOLET ROUND	NUMBER OF WHITE ROUND
1921	73	148	79
1922	101	195	96

Triumph $\times$ *Early White*

Triumph is red-striped and has vigorous foliage, while Early White has white roots and short foliage.

Several reciprocal crosses were made between these two varieties; they all showed in the F_1 flattened roundish roots with strong to faint reddish-purple stripes and spots, on a white background. All individuals developed a strong foliage, though less vigorous than that of the Triumph (figure 2). The first hybrid generation produced, on self-fertilization, offspring with striped *versus* pure-white roots, and strong, medium or short foliage. If R stands for red-striped, r its absence, S for strong foliage and s for short foliage, and the combination Ss produces foliage which is practically always of medium length, there should result in the F_2 , a modified dihybrid ratio, $3RSS : 6RSs : 3Rss : 1rSS : 2rSs : 1rss$; but the following results indicate that the ratio actually present is $1RSS : 2RSs : 1Rss : 1rSS : 2rSs : 1rss$. This 1 : 1 ratio with respect to red striping suggests the action of a gamete lethal.

FIGURE 2.—Triumph, with vigorous foliage $\times$ Early White.

<i>Observed</i>	<i>Expected</i>	
	(3 R : 1 r)	(1R : 1 r)
4	8	5
9	15	10
5	8	5
3	3	5
14	5	10
6	3	5
—	—	—
41	42	40

The fit is very close to expectation on the assumption that red-striped and white occurred in a 1 : 1 ratio.

Neither in F₁ nor F₂ was there any apparent difference between the red of a heterozygote and that of a homozygous Triumph.

Triumph × *Icicle*

The F₁ produces roots with a shape intermediate between the two parents, whereas the striped character is dominant. They behave like a dihybrid. It is not difficult to distinguish heterozygous individuals as far as the form of the roots is concerned (figure 3), but with respect to color only the offspring can show whether its parent was homozygous or not.

In the F₂ the following combinations were obtained:

<i>Observed</i>	<i>Expected</i> (1 R : 1 r)
19 with red-striped, flattened round roots.....	24.5
51 red-striped, intermediate in shape.....	49
24 red-striped, shape of Icicle.....	24.5
13 pure white, flattened round roots.....	24.5
67 pure white, intermediate.....	49
22 with appearance of Icicle.....	24.5
196	196

Long Red × *Early White*

The variety Long Red has long foliage, whereas Early White forms short foliage. The F₁ individuals have medium-sized foliage and broad roundish to conical-shaped roots of a violet color similar to the crosses between other white and red varieties. The second hybrid generation produced long white roots, conical white, round white, long red, conical red, round red, round violet, conical violet or long violet, with either long, medium or short foliage. None of the plants with medium foliage, conical shape or violet color of the roots bred true. The conical shape splits in round, conical and long; the violet color in red, violet and white, and the medium foliage in long, medium and short foliage.

The following combinations were obtained in this cross, with frequencies indicated:

FIGURE 3.—Triumph, with vigorous foliage $\times$ Icicle.

- 5 red, round root, long foliage
- 27 red, conical root, long foliage
- 7 red, long root, long foliage.
- 9 red, round root, medium foliage
- 21 red, conical root, medium foliage
- 6 red, long root, medium foliage
- 8 red, round root, short foliage
- 31 red, conical root, short foliage
- 11 red, long root, short foliage
- 24 violet, round root, long foliage
- 52 violet, conical root, long foliage
- 17 violet, long root, long foliage
- 15 violet, round root, medium foliage
- 43 violet, conical root, medium foliage
- 21 violet, long root, medium foliage
- 20 violet, round root, short foliage
- 57 violet, conical root, short foliage
- 18 violet, long root, short foliage
- 11 white, round root, long foliage
- 23 white, conical root, long foliage
- 9 white, long root, long foliage
- 8 white, round root, medium foliage
- 19 white, conical root, medium foliage
- 12 white, long root, medium foliage
- 5 white, round root, short foliage
- 25 white, conical root, short foliage
- 13 white, long root, short foliage

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Icicle × *French Breakfast*

French Breakfast has short-cylindric red roots and Icicle long-cylindric white ones. The very uniform F_1 has a broad-conical shape and a violet to purple color. When individuals, however, are allowed to be self-pollinated they produce, not only, as was expected, offspring with red, violet or white, short-cylindric, broad-conical or long-cylindric roots, but they also produce individuals of a pink color, which are presented in all shape types. The combinations appeared in the numbers shown in table 3.

TABLE 3

COLOR	FRENCH BREAK-FAST FORM	ICICLE FORM	HETEROZYGOUS BROAD CONICAL	TOTAL
Red with white points	4	2	6	12
Pink with white points	3	1	2	6
Heterozygous violet with white points	38	28	52	118
White	6	3	9	18
Totals	51	34	69	154

These data do not come as close to a trihybrid ratio as might have been expected, owing to the considerable excess of violet; but there can be no doubt that this is the theoretical ratio involved.

Crosses involving Long White

Very interesting were the observations on the commercial variety, Long White. I found in a seed-bed of very young plants two distinct types as to the development of the color pigment, which can be detected only when the plants are very young, before the secondary cortex has developed and before the primary cortex of the root has disappeared. About 90 percent of the seedlings had white primary-cortex cells, but the rest showed red primary-cortex cells. As the roots become fleshy the red color disappears and there is then no difference to be observed. Therefore, these factors can be studied only when the seedlings are small.

I was able to obtain pure strains possessing each of these two alternative characters. Reciprocal crosses were made and all the F_1 plants showed roots with a reddish primary cortex. From self-pollinated plants I expected in the F_2 a ratio of 1 white to 3 red, but this was not always the case; where the red strain was used as mother I found 21 white and 232 red, and when the white strain was used as the mother there were 3 white and 34 red, i.e., ratios which approach 1 : 15. It is here evident that the red color is not due to a single factor, but is the result of two factors which may be called red_1 (R_1) and red_2 (R_2). Many F_2 individuals showed a splitting in the F_3 in 59 white and 188 red, others in 19 white and 51 red, thus closely conforming to a ratio of 1 : 3, whereas others split in 7 white and 118 red and in another case 29 white to 487 red, in which a 1 : 15 ratio can be easily recognized. On the other hand, a certain number bred true as to the red color pigment, but no attempts were made to determine in the latter, whether the formula was $R_1 R_1 R_2 R_2$, $R_1 R_1 r_2 r_2$, $r_1 r_1 R_2 R_2$, $R_1 R_1 R_2 r_2$ or $R_1 r_1 R_2 R_2$.

Evidently two individuals with the color-factor combination $R_1 r_1 R_2 r_2$ have produced the ratios 7 : 118 and 28 : 487, whereas those with $R_1 r_1 r_2 r_2$ or $r_1 r_1 R_2 r_2$ must have split in the ratio 59 : 188 and 19 : 51. There is no doubt that this cross behaved as a dihybrid, due to occurrence of two duplicate red-color factors.

Crosses involving Round Yellow

The Round Yellow radish, which is but very little grown, having round yellow roots and foliage of medium size, was hybridized with certain white varieties, especially Round White, Early White with short foliage,

and Icicle. There were all indications that the yellow pigment was due to only one differential factor. Reciprocal crosses between Yellow Radish and Round White, having the same foliage, gave in the F_2 35 white and 101 yellow radishes, a ratio of 1 : 3.

A cross between Round Yellow and Early White acted as a dihybrid. The F_1 was composed of individuals with yellow roots and a foliage intermediate between the two parent plants.

The F_2 was composed of 172 individuals, of which 140 were yellow and 32 white. Among the yellows I counted 34 with long foliage, 41 with short and 65 with medium-sized foliage, the last being regarded as heterozygous with respect to the foliage. Five of these were self-pollinated, and produced either long-, medium-, or short-leaved plants. Of the 32 white F_2 radishes there were 9 with long foliage, 5 with short and 18 with medium foliage, the latter being considered heterozygous.

Crosses between Round Yellow and Icicle gave rise in the F_1 to long-oval yellow radishes. Not enough offspring were obtained in the F_2 ; consequently sufficient data are not present to show a clear ratio, they were probably of a dihybrid nature. The following plants were noticed: 5 round yellow, 6 round white, 8 long-oval yellow, 5 long-oval white, 1 long yellow and 3 long white.

Crosses involving Black Winter

Another situation was offered when the Black Winter radish was used. The main characteristic of this group is the presence of a cork tissue around the periphery of the roots. In crosses with ordinary radishes this character is always dominant. The main factors in which the Black Winter radish differs from ordinary radishes are: (1) cork tissue; (2) shape of roots in some instances; (3) size of roots in all cases; (4) heavy foliage; (5) late maturity.

There are two distinct varieties of Black Winter radish, a round and a conical one, which when crossed with each other produced intermediates in the F_1 and 38 round, 72 intermediates and 27 conical ones in the F_2 ; consequently, they are of monohybrid nature.

Results obtained between the Black Winter radish and ordinary radishes were never sufficiently extensive to permit the determination of clear ratios, but in some cases there were, no doubt, from 4 to 6 factors to be considered.

A cross between Long Red and Black Round Winter was in the F_1 more or less conical. Through the black or grayish cork layer the red pigment due to Long Red could be easily observed in all individuals, of which 11

were obtained. The foliage was very heavy. From one plant 180 F_2 offspring were secured, of which 120 had the appearance of those of the F_1 , 8 were like Black Round Winter, 12 had the shape of Long Red, but had the color of the F_1 and 23 were large, round and red. These data are clearly inadequate to give a proper idea of the actual ratio.

A cross between Yellow Vienna and Black Round Winter proved to be in the F_1 more or less roundish and black, but the yellow color due to Yellow Vienna, was everywhere visible. But 31 F_2 plants were grown, of which 18 were like the F_1 , 9 had the appearance of Black Round Winter, and 4 resembled the Yellow Vienna. Here also a sufficient number of individuals was not present.

SUMMARY

This paper deals with factors for form and color of the roots, and length of foliage, in crosses between varieties of summer radishes, and between these and the black winter radishes.

In all crosses the form of the roots was determined in each case by a single factor pair and the heterozygote was intermediate between the two homozygous parents. The same thing was true in regard to length of foliage.

When white and red varieties were crossed, the heterozygotes were in every case violet to purple, easily distinguishable from the homozygotes.

A factor, *R*, for red striping gave 1 : 1 ratios following self-fertilization, suggesting the possible presence of a gamete lethal.

In the cross between white-rooted Icicle and red-rooted French Breakfast, there appeared in addition to the expected parental types and purple heterozygotes, a small percentage of pink-rooted plants.

In the variety Long White some seedlings had red primary-cortex cells while others had white. This difference is observable only in the early seedling stage. Some of the red seedlings bred true, others gave ratios of 3 red : 1 white, and still others 15 red : 1 white, indicating the occurrence of duplicate factors.

The yellow color of Round Yellow is due to a single factor difference from white, the yellow being dominant.

Black Winter radishes crossed with the summer radishes showed the corky tissue around the periphery of the roots of the former dominant over its absence, and the black color dominant or nearly completely epistatic over the other colors.

While many of the crosses produced too few offspring for a decisive determination of ratios, the results indicate that the radish varieties are worthy of extensive and intensive genetical analysis.

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