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The Seed Progeny of *Solanaceae* Chimeras

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Souhrn

Je popsáno získání dvou typů chimér mezi rajčetem a lilkem černým (*Solanum koelreuterianum* WINKLER a *S. gaertnerianum* WINKLER).

V mikromorfologických a anatomických znacích u *S. gaertnerianum* nebylo zjištěno žádné průkazné specifické ovlivnění jednoho pletiva druhým.

Semenné potomstvo chiméry *S. gaertnerianum* se ve všech studovaných znacích shodovalo s kontrolním lilkem.

Sledovány byly znaky: Velikost průduchů, tvar průduchů, tvar trichomů, pokožka plodů, průřez řapíkem, průřez listovou čepelí a počet semen.

Summary

A description is given of the production of two types of chimera between tomato and nightshade (*Solanum koelreuterianum* WINKLER and *S. gaertnerianum* WINKLER).

In the case of *S. gaertnerianum* the micromorphological and anatomical characters showed no signs of the significant specific reciprocal effect of one tissue on the other.

The seed progeny of the *S. gaertnerianum* chimera resembled the control nightshade plants in all the characters investigated.

The following characters were studied: Size of stomata, shape of stomata, shape of trichomes, fruit epidermis, section through the petiole, section through the leaf blade and number of seeds.

Introduction

FROST (1926) divided chimeras according to their origin, which has no connection with the relationship of the components. He recognized autogenic chimeras arising from somatic mutations, and synthetic chimeras in which an artificial measure, specifically grafting, plays a part in the joining of two or more genotypes in the growth apex. The whole question of chimeras is closely connected with the anatomy of the vegetative cone, in which the number of cell layers which are genetically independent of each other determines the formation of monochlamydeous, dichlamydeous etc. chimeras. A considerable volume of work is, therefore, directed to these questions (NOACK 1922, SCHWARZ 1927, LANGE 1927, CLOWES 1957).

WINKLER (1935, 1938) distinguished three layers in the vegetative cone of periclinal chimeras : ectosoma, mesosoma and endosoma, which do not correspond in their delimitation with the embryonic layers of the tunica-corpus. On this basis he distinguishes:

1. ectochimeras (*Solanum koelreuterianum*, *S. gaertnerianum*, *S. tubingense*, *S. proteus*, etc.);
2. mesochimeras, in which a layer of one component is inserted between layers of the second component (e.g. Winkler's *S. mendelianum*, or chimeras described by DERMEN 1947, 1951 in apple and cranberry).

The historical graft chimeras (bizarre, *Laburnocytisus adami*, *Crataegomespili*) aroused interest in their time, but the main attention was paid to them once more following WINKLER and BAUR'S fundamental theoretical work.

Since the practical application of graft chimeras met with no success (SAHLI 1913, 1916, KLEBAHN 1918), research work with them continued to be directed mainly to theoretical questions. Attention was paid to the mutual influence between the components and the effect on the sexual progeny. The question is whether the two kinds of plant tissue which form one morphological unit lose their genetic individuality and their genotypes acquire characters of the new phenotype, or whether their genetic independence remains unchanged. The oldest known graft chimeras were already considered as graft hybrids (e.g. Darwin in L. adami). Winkler himself was convinced at first that his experiments had demonstrated the possibility of forming hybrids in a vegetative manner, i. e. by grafting. There is, however, a fundamental difference between his conception of vegetative hybridization as the fusion of the nuclei of two somatic cells which had been transected during grafting, resulting in the formation of some sort of vegetative zygote (as he assumed with his "burdons"), and the conception of vegetative hybridization as a consequence of mutual genetic influence by assimilates passing from one component to the other (GLUSHCHENKO 1948a, b, LYSENKO 1952, BAZAVLUK 1952, GLAVINICH 1949, 1957, AVAKYAN 1957).

In general it can be said, however, that the findings of the authors do not differ so much as regards facts as in the construction put upon them. This may be due either to different connotations of the terms used (e. g. hybrid, hybridization, heredity), or to failure by the author to exhaust all possible means of arriving at an explanation.

Material and Methods

The tomato *Lycopersicon esculentum* MILL. cv. Immun and *Solanum nigrum* L. with yellowish-green fruits were used in the experiments. Winkler's method was used for the production of chimeras. This depends on the mutual grafting of both components, with subsequent decapitation at the point of fusion and the creation of favourable conditions for callus formation. Cleft grafting was more succesful. Following cutting at the point of fusion, the growth apices from the leaf axils were removed in order to prevent weakening of the growth of adventitious buds growing from the callus formed at the cut surface. The plants require to be kept in a moist atmosphere so that the cut surfaces do not dry up. Cuttings were made from the chimeras and planted out independently. The seeds from the chimeras were germinated on filter paper in Petri dishes and at the cotyledon stage the seedlings were transplanted to flower pots.

Measurement of stomata was carried out by the microrelief impression method (WOLF 1954) with acetone varnish. The stomata were measured on the under surfaces of leaves which had finished their growth.

Results

Two types of chimeras were obtained—*Solanum koelreuterianum* and *S. gaertnerianum*. Winkler's original binomial nomenclature has been used in view of its simplicity, although it cannot be fully justified from the taxonomical point of view.

Solanum koelreuterianum originated in several specimens. It was already known from the work of other authors that this monochlamydeous chimera with nightshade epidermis is notable for its high degree of sterility. Not a single seed was obtained from the present material. The flowers fall either as buds or soon after opening. Only exceptionally was a parthenocarpic fruit formed.

Solanum gaertnerianum developed in 1956 from the grafting of nightshade on tomato. In the majority of its characteristics it tends to resemble nightshade. However, its type of growth is different, the shoots do not stop growth and the plants reach a much greater height (over 1 m).

The flowers are white, mostly five-petalled and in size and form the flower organs resemble nightshade. Other flowers have a multiplied number of petals, in other cases the petals develop irregularly (some are deformed, others normal). There are interesting cases of flowers with petals and stamens very much reduced in size but multiple in number, surrounding an enlarged ovary with a large style, which survives for a long time.

This plant produced no fruits in the year of grafting, in the second year it produced some in the autumn and the greatest number of fruits were formed by clones in spring 1958. The shape of the fruit shows a transition from flattened to oval and finally to very elongated fruits. The elongated forms never contained any seeds.

The shapes of the leaves are very varied. The margins are usually entire, lobed or with an indication of pinnation. The leaf blade is often asymmetric. The leaf bases pass from heart shape to an elongated junction with the petiole. Very long petioles are striking, as is the helmet-like curvature of the leaf blade with a bulge on the upper surface.

The chimera seeds were sown from fruits which ripened in the autumn of 1957. Forty plants were grown from them, all pure *Solanum nigrum* with yellowish-green fruits like the original component of the chimera.

Comparison of characteristics of the chimera, its seed progeny and control plants.

Size of stomata. Stomata were measured in all cases on the same part of the leaf blade, in the centre, but not immediately at the main vein. The measurement showed:

1. *S. nigrum* has larger stomata than *L. esculentum.*
2. The chimera *S. gaertnerianum* does not differ as regards size of stomata from its seed progeny.
3. The seed progeny of chimeras do not differ as regards size of stomata from the control.

Shape of stomata. No significant differences in shape of stomata were found between the different forms examined. Those of tomato (cv. Immun), nightshade, *S. gaertnerianum* and its seed progeny are oval on the lower leaf surfaces, only on the upper surface in tomato some have a single protrusion at one or both ends. This character was not observed in the chimera, but an analogous form was met with in control nightshade plants and the seed progeny. In these cases, however, it is possible by focussing the microscope on the lower part of stoma to observe that the phenomenon is due to a fold of the neighbouring epidermal cell. In tomato there may be an actual thickening of the cell wall at the point of contact and fusion of both guard cells.

Trichomes. In all forms there are basically two kinds, glandular and protective.

Protective trichomes of tomato are usually formed of three cells, of which the apical narrows to the point, while in nightshade they are in most cases club-shaped. The species to which the epidermis of a chimera belongs can be easily recognised by means of these trichomes. Occasionally pointed trichomes of the tomato type were found on some leaves of chimeras, their seed progeny and of control nightshade plants.

Glandular trichomes have a one-celled, rarely two-celled stem on which rests a multicellular head. In tomato this head is made up of one small cell with four round cells on it. In nightshade these short trichomes are varied in form, but trichomes with four-celled heads were not observed on leaves of nightshade or chimeras.

Epidermis of fruits. There are considerable differences between the two parent species. Tomato fruits have an epidermis with very thick-walled cells, nearly square in shape. In nightshade the cells are larger, more rectangular, but with cell walls of about a quarter of the thickness of those in tomato. In the *S. gaertnerianum* chimera (diectochimera) and its seed progeny the cell walls of the fruit epidermis were found to differ in no way from those of the nightshade control.

Section of the petiole. The base of the petiole is similar in all the plants examined. There are either winged petioles (nightshade) or there are indica-

tions of wingedness as in tomato. The vascular bundles are bicollateral in both species, the wood in nightshade falls into three parts, in tomato and chimeras it is divided into many parts, separated by rows of parenchymatous cells. The seed progeny of the chimeras did not differ as regards this characteristic from the control nightshade.

Leaf section. The two parent species differ from each other as regards the proportions of palisade and spongy parenchyma. While palisade cells form nearly one-half of the thickness of the leaf in nightshade, they form only about one-third in tom to leaves. In the chimera *S. gaertnerianum*, where the palisade tissue is formed from the outer component and tomato forms only a smaller part of the leaf blade, the leaf section resembles nightshade in the proportions of its mesophyll. The seed progeny also resemble the control plants.

The effect on the fertility of progeny was followed according to the number of seeds in the berries. In ten plants taken at random from the seed progeny of the chimera, seeds were counted in ten berries from each plant, i.e. in a total of 100 berries. Similarly 100 berries from ten plants were examined in the control. In the seed progeny the average number of seeds in one berry equals $\bar{x} \pm 3 \,.\, s_{\bar{x}} = 30{\cdot}25 \pm 3 \,.\, 1{\cdot}15$. For control nightshade plants with yellowish-green fruits $\bar{x} \pm 3 \,.\, s_{\bar{x}} = 33{\cdot}00 \pm 3 \,.\, 1{\cdot}14$.

$t_{198} = 1{\cdot}62$. For $P = 0.01$ $t_{200} = 2{\cdot}60$ and for $P = 0{\cdot}05$ $t_{200} = 1{\cdot}98$. As the results show, the seed progeny are not influenced in their fertility and completely agree with their controls as regards this character.

Discussion

A great deal has already been written about the so-called vegetative hybridization from various points of view. RYZHKOV (1947) wrote on the subject of including chimeras among vegetative hybrids: "If the term hybrid is used in the exact sense, i.e. if we mean the product of fusion of two cells, then this is in no case a hybrid".

This criterium could be fulfilled only by the so-called burdon, the existence of which has not yet been demonstrated.

If we take into consideration that in more recent genetic definitions a hybrid is identified with a heterozygote, then the condition remains that the hybrid nature of an organism, if it is fertile, must evince itself in some form in later generations.

According to T. D. Lysenko vegetative hybridization would be a relatively easy matter. "It is necessary to force the scion to build up its body completely from nutrients which are supplied by the roots of the stock and from plastic substances produced by the roots of the stock and from plastic substances produced by assimilation by the stock's leaves. In such cases vegetative hybridization will always be completely successful." (Lysenko, p. 239.)

During the formation of the chimera only an insignificant wedge of the second component without assimilatory surface remains after decapitation, from which in conjunction with the first component the chimera shoot de-

velops, which in fact builds up the whole of its body from nutrients and "plastic substances" supplied by the stock. In the experiment described above the supplier was represented by the tomato, and nevertheless neither in the micromorphological characters of the seed progeny nor in the nightshade tissues of the chimera itself was there evidence of any specific influence.

The group of authors already mentioned explain this as being due to developing zygotes in the flowers of chimeras which are able to choose the nutrients of only one genetic norm in agreement with Michurin's teaching on the impossibility of mixing specific characters. (GLUSHCHENKO 1948a, b, BAZAVLUK 1952, AVAKYAN 1957.)

Long before this theory was evolved it was known that in the great majority of cases gametes are built up from the subepidermal cell layers. It was not, therefore, surprising that the progeny of monochlamydeous chimeras are identical with the inner component, the dichlamydeous with the outer. There was thus no demand for a theory which would endow certain cells with such exceptional abilities. Only following the assumption that the transfer of assimilates from one component to the other implies the passing on of hereditary characters, did it become necessary to explain why these characters are never expressed in the sexual progeny. This is therefore a speculative rather than an experimentally founded theory.

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Семенное потомство пасленовых химер

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Резюме

В статье описывается получение химер между томатом и пасленом (*Solanum koelreuterianum* и *S. gaertnerianum*).

По анатомическим и микроморфологическим признакам не удалось у *S. gaertnerianum* обнаружить какого-либо специфического влияния между тканями.

Семенное потомство химеры *S. gaertnerianum* по всем признакам сходно с растениями паслена.

Исследовались следующие признаки: величина устьиц, форма устьиц, форма трихом, эпидермис плодов, разрез через черешок, разрез через листовую пластинку и количество семян.