Modality of the Contractile Root Formation in Cultivated Saffron, in Field Condition and in Tissue Culture

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Keywords: contractile roots, Crocus sativus L., tuberous root

Abstract
In cultivated saffron (Crocus sativus L.) various kinds of contractile roots are formed. The most important of contractile roots are produced at the terminal buds. From corm fragments carrying bud protuberance, in MS culture medium containing 2,4-D (0.5 mg/l) and kinetine (9 mg/l), contractile roots are formed and then contracted at the base. Phenolic compounds specially paracomaric acid have positive effect on their formation, growth and contraction.

INTRODUCTION
In the corms of Crocus sativus L. (saffron) a tuber organ is formed, very large and whitish, which at the very beginning was named dropper. Studies have shown that they have root structure and are contracted in the base (Ebrahimzadeh, 1977). Many monocotyledons species have contractile roots (Puetz, 1991, 1992, 1993, 1995, 1999). Pulling and pushing activity of contractile roots enables corms to move into the ground, so corms rest in optimum depth and position of soil (Chio-Sang, 1996). In contractile roots, water, minerals and photo-assimilates are accumulated (Iziro, 1984) but not starch (Ornduff, 1969). Hereby we report our studies on formation of contractile roots in saffron plant.

MATERIALS AND METHODS
Saffron corms were cultivated in sand, and the mixture of sand-clay and clay in 5-cm depth and irrigated with solution containing mannitol, minerals, phytohormons, and phenolic compounds. The modality of contractile root formation has been studied morphologically in the field and in vitro by tissue culture technique. The explants from corms fragments carrying buds, protuberance, and contractile roots were sterilized by Ethanol 70% and 0.25 % MgCl₂. They then were transferred to the MS and MS/2 media (Murashige and Skoog, 1962) containing 2,4-D (0.5 mg/l), BAP (0.5 mg/l) or kinetine (9 mg/l) and 2% coconut milk.

RESULTS
Formation of Contractile Roots in Field
Our studies show that the contractile roots are existed in four different types, which are named in the basis of their formation place. We categorized these types as following:
1. Fibrous Contractile Roots: They are formed before flowering time at the beginning of November and they appear on the fibrous root ring of corm and they will become contracted in few times. In the every corm, about 8-13 fibrous contractile roots were formed (Figure 1 A).
2. Corm Contractile Roots: These contractile roots appear after flowering time in middle of November and are not produced from the fibrous root rings, but rather originate from corm periphery. In the every corm, about 1-2 contractile roots were produced (Figure 1 B).
3. Lateral Bud Contractile Roots: The primordia of these contractile roots originated before flowering, from base of lateral buds and continue to growth after flowering. They
then become contracted later during senescence. In the every corm, about 3-5 lateral bud contractile roots were produced (Figure 1 C).

4. Terminal Bud Contractile Roots: The contractile root primordium appears in December, before flowering in the base of apical bud. Firstly, they are seen as small tuberous root and are thicker than the other types. Their number per corm is mostly 1 but increase to 2-3 rarely (Figure 1 D).

Formation of Contractile Roots from Fragments in the Tissue Culture

From corm fragments carrying bud protuberance, in MS culture medium with 0.5 mg/L 2,4-d and 9 mg/L kinetine, contractile roots are formed which are tuberous at the beginning and then become contracted at the base (Figure 2 A).

From explants carrying bud and contractile root in MS and MS/2 media, containing 2,4-D and BAP (each 0.5 mg/L) and 2% coconut milk callus is formed (Figure 2B, 2C).

Effect of Mannitol, Phytohormones and Phenolic Compounds on the Growth of Contractile Roots

Phenolic compounds, for example paracomaric acid, parahydroxy benzoic acid and galic acid with 99.5 % confidence coefficient, have stimulation effects on the length and thickness growth of contractile roots, contraction intensity and dry weight of buds contractile roots. Mineral and mannitol produce osmotic pressure and clay with 99.5 % confidence coefficient has repressive effects on the buds contractile roots. We can summarize our results on Table1.

DISCUSSION

The contractile roots are produced not only in saffron (C. sativus) but also in C. hausskenechitii and some other monocotyledons, for example Gladiolus and Oxalis (Puetz, 1995). Fibrous contractile roots and corn contractile roots are formed in mother corm before flowering. In the flowering time, they have a great length growth and then they are contracted in the base and mother corm moves under ground (Puetz, 1992, 1993). Water, mineral and photoassimilates are providing factors for them. Terminal and lateral buds contractile roots primordium are formed before flowering but their growth and contraction is after flowering. Therefore they may have important effect on growing and provision of water, mineral and rate of pigment production and material transport into the daughter corms, which change to mother corms in the following year.

The successful formation of contractile roots on explants will able researchers to study them in controlled culture conditions. There is no report about this so far. Phenolic compounds and plant hormones have crucial role in contraction and folding of contractile roots.

Literature Cited


Iziro, Y. 1984. Studies on the role the contractile roots in relation to the thickening growth of daughter corm or bulbs in Gladiolus freesia and Oxalis bowieana. Special Bulletin of the College of Agriculture Utsunomiya University 41: 1-90.


### Tables

#### Table 1. Characteristics of contractile roots in soil.

<table>
<thead>
<tr>
<th>Contractile roots kind</th>
<th>Contraction time</th>
<th>Stimulants factors</th>
<th>Repressive factors</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fibrous and corm contractile roots</td>
<td>Before flowering</td>
<td>Auxin $10^{-5}$ mg/L, Gibberelin $10^{-7}$ mg/L, Mineral and clay</td>
<td>Phenolic compounds</td>
</tr>
<tr>
<td>Buds contractile roots</td>
<td>After Flowering</td>
<td>Phenolic compounds</td>
<td>Mineral, Mannitol, Clay</td>
</tr>
</tbody>
</table>

Fig. 1. A. Fibrous contractile roots in root circle in various stages of contraction. B. Corm contractile root in early stages of contraction. C. Lateral bud contractile roots in various stages of growth. D. Terminal bud contractile root
Fig. 2. A. Formation of contractile roots from corm fragment carrying bud protuberance, in MS culture medium containing 2,4-D (0.5mg/l) and kinetin (9 mg/l). B. From fragment carrying bud and contractile root in MS and MS/2 culture medium containing 2,4-D and BAP (0.5 mg/l from each) and coconut milk. C. Callus is formed.