Growth response of excised and attached roots to external sucrose concentration

II. Pisum sativum

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In the previous paper (S z we y k o w s k a and Marchlewska, 1964) it has been found that the growth response of roots to sucrose concentration in the medium is different depending on whether the root is detached and grown as an excised organ or whether it grows attached to the shoot. Experiments were made on flax roots and showed that optimal growth of excised roots occurred in lower sucrose concentrations than that of attached roots.

Numerous investigations have been made using excised roots of pea. Bonner and Addicott (1937), Addicott and Devirian (1939) performed studies on nutrient and growth factor requirements of excised pea roots and found in respect to sugar nutrition that roots grew optimally in 40% sucrose. Working on isolated embryos of lupin, a plant belonging to the same family as pea, Hoffmannowa (1963) found that maximum growth of attached roots of cotyledonless seedlings occurred in 80% sucrose, i.e. in a concentration relatively high. Fries (1935) compared growth of excised and attached roots of pea growing in medium containing sucrose at 20% conc. and found that excised roots grew better than attached roots, especially after a culture period lasting above 5 days. The present work was undertaken to investigate the effects of various sucrose concentrations on growth of excised and attached roots of pea and on the sugar accumulation in the organs as a possible cause of growth differences.

MATERIAL AND METHODS

Seeds of $Pisum\ sativum\ "Victoria"\ were\ used in the experiments.$ The seeds were sterilized in 96% ethanol for 1 minute, then in 1% HgCl₂ solution for 10 minutes. After thorough washing the seeds were put into a sterilized Petri dish with water and soaked for 24 hours. From the swollen seeds cotyledonless embryos were isolated and transferred to culture flasks with medium. To obtain the root culture, the swollen seeds were transferred to sterile dishes with wet filter paper. After two

days of germination, 10 mm of root tips were cut from the seedlings and transferred also to culture flasks with medium. A parallel experimental set consisted of 20—30 seedlings resp. excised roots.

The composition of nutrient medium, the culture procedure and the methods of determining protein nitrogen and soluble sugars have been described in our previous paper (S z w e y k o w s k a and Marchlewska 1964).

RESULTS

The growth of pea roots was estimated on the basis of three criteria: length of the main root, fresh weight and protein nitrogen. In a 7 day culture, the excised roots reached a maximum length and fresh weight in 3% sucrose. The mean fresh weight of a root amounted in optimal conditions to 29 mg, the mean length to 47 mm. For attached roots the growth curve had a different maximum, occuring at 8% sucrose. The mean fresh weight of a root amounted here to 60 mg and the mean length to 85 mm. Attached shoots grew best in 6% sucrose (Figs. 1—2).

Nitrogen analyses were made of excised and attached roots growing in 0, 3, 6, 9 and 12 per cent sucrose. The curves obtained were similar to those of the length and of the fresh weight of roots. The positions

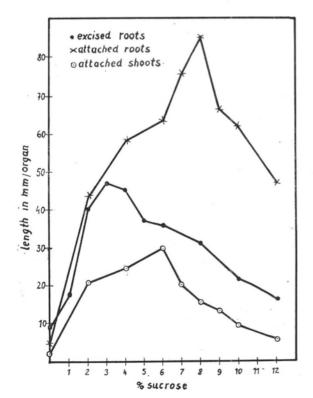
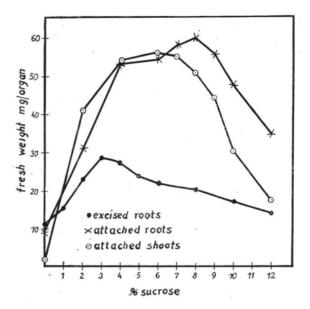


Fig. 1. Effect of external sucrose concentration on length of main roots and shoots in pea seedlings.

Fig. 2. Effect of external sucrose concentration on fresh weight of pea seedling organs.



of the maxima were different in excised and in attached roots and fell in the first case on $3^{0}/_{0}$ and in the second case on $9^{0}/_{0}$ sucrose (Fig. 3).

In this way differences in responses to sugar concentration in the medium were found in pea roots, depending on whether they grew as isolated organs or whether they formed a whole with the shoot system. In pea these differences are much greater then in previously investigated flax roots which perhaps is linked with the generally weak development of root system and the large demand for water in flax, and is why even

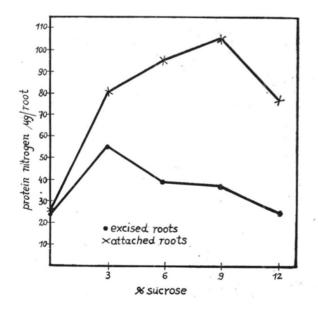


Fig. 3. Effect of external sucrose concentration on protein content in pea seedling roots.

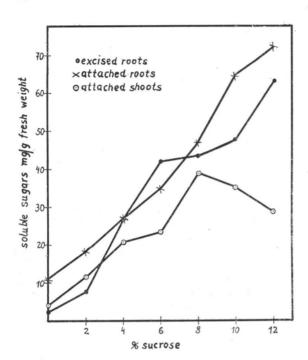


Fig. 4. Effect of external sucrose concentration on soluble sugar accumulation in pea seedling organs.

attached roots do not tolerate the osmotic values of a medium with relatively very high sugar content.

Sugar accumulation in the organs of pea is also proportional to the sucrose concentration in the medium. In pea, however, there are no marked differences in the soluble sugar accumulation between excised and attached roots. The distribution of absorbed sugar in whole seedling is also different from that found in flax. The soluble sugar concentration in attached roots is higher than in shoots (Fig. 4). So in pea seedlings there is no active depleting transport of sugars absorbed from the medium, from the root to the shoot. It is probable that the sugar absorbed is transported passively into the tissues and therefore its concentration does not differ in excised and attached roots and is somewhat lower in attached shoots. This explanation seems possible in view of the data of Hoffmannowa (1963) who in investigating sugar nutrition of isolated lupin and cabbage embryos found distinct differences in growth response between them, and whereas the response of cabbage seedlings to sugar in the medium was very strong, that of lupin seedlings, especially shoots, was much weaker. The authoress suggests that one of the factors of these different responses may be adaptation to various metabolic pathways in lupin and cabbage connected with their mobilizing various storage foods in first stages of development. The same may apply to pea and flax which in respects of the kind of storage foods in seeds represent the same two types as lupin and cabbage.

DISCUSSION

Great differences in the response to external sucrose concentration have been shown between excised and attached roots of pea. These growth differences are not accompanied by differences in soluble sugar accumulation in the organs such as occur in excised and attached roots of flax (Szweykowska and Marchlewska 1964). It has thus been concluded that the pea shoot does not abstract from the root any important quantities of sugar absorbed by the root from the medium, and the sugar content in the root is even somewhat higher than in the shoot. So it is believed that the shoot affects the growth responses of root in another way. First of all, different synthetic abilities of root and shoot metabolisms seem to have to be taken into consideration. It is well known that the pea root is not auxoautotrophic in respect to the thiamine and the nicotinic acid. In the higher sugar concentrations in the medium, still another factors may play a role. Butcher and Street (1960) found that kinetin inhibits growth of excised tomato roots in suboptimal sucrose concentrations in the medium and promotes it in supraoptimal ones. So it is possible that the shoot supplies the root with some factor which enables root meristem to divide actively in high sucrose concentrations resulting even in a maximum growth in conditions which are growth inhibiting for a detached root. The growth responses of roots are thus various and complicated: they are different in excised and in attached roots, they may also differ (in respect to e.g. kinetin) depending on such a relatively simple factor as sugar concentration in the medium.

The results obtained in the present study and the postulated dependence of root on shoot seem to be contrary to the findings and conclusions of Fries (1953). Using a medium with a constant $2^0/_0$ sucrose concentration, Fries found that the growth of attached roots was weaker than that of excised roots. According to this author, roots synthesize substances which are indispensable for shoot development and which the shoot takes up from the root, arginine, glycine and adenine representing the limiting factors in pea seedling root growth. As a result of shoot depleting root of these substances, attached roots develop weaker than excised ones growing without shoots as isolated organs.

In our experiments, on the contrary, attached roots always grew better than excised ones. This inconsistency demands a more detailed study and will be considered in a separate paper.

SUMMARY

The growth responses to various sucrose concentrations in the medium of excised and attached roots of pea were investigated. Fresh weight, length and protein nitrogen were estimated. Three per cent sucrose has been found to be

optimal for the growth of excised roots and eight per cent for that of attached roots.

Sugar accumulation in excised roots and in attached seedling organs (roots and shoots) was also investigated. No distinct difference was found in soluble sugars in excised and attached roots. The possibilities were discussed of the root being supplied by the shoot with some factor, enabling a vigorous growth of the attached root system in at high sugar concentration in the medium.

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Reakcje wzrostowe izolowanych i nie izolowanych korzeni na wzrastające stężenie cukru w pożywce

II. Pisum sativum

Streszczenie

Zbadano reakcję wzrostową izolowanych i nie izolowanych korzeni grochu na stężenie sacharozy w pożywce. Oznaczano długość korzeni, świeżą masę oraz zawartość azotu białkowego. Stwierdzono, że optymalnym stężeniem sacharozy dla korzeni izolowanych było 3%, dla nie izolowanych 8%.

Zbadano także akumulację cukrów rozpuszczalnych w korzeniach izolowanych oraz nie izolowanych organach siewek (korzeniach i pędach). Nie stwierdzono wyraźnych różnic w zawartości cukrów w korzeniach izolowanych i nie izolowanych.

W pracy dyskutowana jest możliwość, iż pędy zaopatrują korzeń w pewne czynniki, które umożliwiają nie izolowanym korzeniom bujny wzrost na stosun-kowo wysokich stężeniach sacharozy w pożywce.