STUDIES ON THE RHIZOSPHERE BACTERIA OF ERICACEOUS PLANTS

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INTRODUCTION

Several reports have been published concerning the nutritional requirements of bacteria in the rhizosphere of cultivated plants and in the surrounding soil. In general the total number of bacteria shows a distinct increase in the rhizosphere. The greatest increase has been observed within the nutritional group requiring amino acids for maximum growth, and within the auxoautotrophic group. It has been concluded that the stimulation of the former group is due to the excretion of amino acids by the growing plants.

Gyllenberg studied the rhizosphere effect of graminaceous plants in virgin soils, and also found a stimulating effect of the roots on amino acid-requiring bacteria. However, when the rhizosphere effect of different tree-seedlings was tested, the woody parts of the roots were found to exert an inhibiting effect on amino acid-requiring organisms in the surface layers of the soil. The contrary was the case in deeper soil horizons.

As compared with the knowledge of the rhizosphere effects in cropped soils, the knowledge of the rhizosphere phenomenon in forest soils is limited. In the present work an attempt has been made to study the latter. The soil samples studied were taken from a Picea abies-Pinus silvestris forest on gneiss rock, where the ground was covered with a mixture of Vaccinium vitisidaea, V. myrtillus, Calluna vulgaris, and mosses (Hylocomium, Dicranum, Polytrichum). The results reported below refer to the rhizosphere effect exerted by the ericaceous plants, the roots of which were highly intermingled in the soil studied.
MATERIAL AND METHODS

Humus extract.

1 kg of humus (pH 3.8 in October 1962) was extracted with 1 litre of tap water at 4° for 24 hours. After centrifugation, the filtrate was made up to 1 litre.

Media

Basal medium: Glucose 0.1% (w/v), (NH₄)₂SO₄ 0.05%, KH₂PO₄ 0.1%, MgSO₄·7H₂O 0.02%, CaCl₂·2H₂O 0.01%, and FeCl₃·6H₂O 0.001%, pH 5.0. In some cases the basal medium was supplemented with 0.2% casamino acids or 0.01% yeast extract (Difco). Humus extract agar: Humus extract supplemented with 0.02% KH₂PO₄, Mycostatin (Squibb Nystatin U.S.P.) 100 units per ml, and 1.5% Difco agar, pH 5.0. Humus extract-glucose agar: 50% humus extract and 50% tap water supplemented with glucose 0.1%, (NH₄)₂SO₄ 0.05%, MgSO₄·7H₂O 0.02%, KH₂PO₄ 0.05%, CaCl₂·2H₂O 0.01%, FeCl₃·6H₂O 0.001%, yeast extract 0.01%, and agar 1.5%. Nutrient broth agar: 0.8% nutrient broth (Difco) supplemented with 1.5% agar. Semisolid medium: Humus extract supplemented with 0.02% KH₂PO₄, 0.01% yeast extract, and 0.5% agar, pH 5.0. Citrate medium: Na₃C₆H₅O₇·5½H₂O 0.3%, NH₄H₂PO₄ 0.1%, MgSO₄·7H₂O 0.02%, KCl 0.02%, yeast extract 0.01%, pH 5.0.

Stock cultures were kept on the semisolid medium.

Further methodical data

The cultural characteristics of the strains were examined on nutrient broth agar, humus extract—glucose agar, and on semisolid medium.

The morphology of the cells was studied partly by using phase-contrast microscope, partly by negative staining of bacteria (Dorner's method). The gram-stain was performed according to Hucker's modification (8 p. 16), and flagella staining according to Rhodes.

Liquefaction of gelatin was studied in stab cultures using humus extract supplemented with 0.02% KH₂PO₄, 0.01% yeast extract, and 12% Difco gelatin.

Hydrolysis of starch was examined in nutrient broth supplemented with 0.2% soluble starch, and on corresponding agar plates.

Reduction of nitrates was studied in humus extract—glucose medium without (NH₄)₂SO₄ and agar but supplemented with 0.1% KNO₃ and Durham tubes. The occurrence of nitrites was tested by means of Griess' reagent every other day up to fourteen days.

Production of indole was tested in humus extract supplemented with 1.0% casitone (Difco) and 0.2% KH₂PO₄, using the technique of Ehrlich-Böhme.

Occurrence of catalase was examined by flooding plate cultures of the organisms with a 10% solution of H₂O₂.

Fermentation of carbohydrates was tested using the following substances, representative of humus: Arabinose, xylose, ribose, glucose,