# Simulating a Macroscopic Rhizosphere for Measuring Potassium Depletion in the Close Vicinity of Rape Roots\*

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## Abstract

Our purpose is to know how deeply K dynamics might be shifted consequently to the K depletion occurring in the rhizosphere of rape. In this scope, a culture device is proposed which increases the root/mineral ratio for exacerbating their interactions. This rhizosphere magnification is obtained through (i) developing a planar root mat assumed as a macroscopic root surface, and (ii) diluting the K-silicate as a coagulated suspension in agar gel. The results show a very deep depletion occurring within a few days of cropping. K concentration near roots rapidly decreased to less than  $10^{-4}$  M. The consequent shift of K dynamics in the mineral phase appears through a concomitant weathering of micas.

# Introduction

In the rhizosphere, nutrient depletion occurs when the demand by the plant, as influenced by root morphology and physiology, exceeds the supply from the soil, as related to nutrient mobility and availability. Because of the small size of depletion zones for poorly mobile elements like K and P, Kuchenbuch and Jungk (1982), after Farr et al. (1969) proposed an original culture device based on the following principle: exacerbating the rhizosphere effects through developing a planar root mat which simulates a macroscopic root surface. After thin slicing the soil in close contact with this "root surface", rhizospheric material is easily obtained in sufficient amounts for its analysis through standard techniques, as a function of the distance from the roots.

But, because of the complexity and heterogeneity of soil material, most of the works on the mechanisms of mineral nutrition were achieved with nutrient solution. In such

<sup>\*</sup>Reviewed

conditions, rhizosphere interactions cannot be encountered. To take them into account, a diffusive and homogeneous medium is proposed as a model of soil for replacing it advantageously in Kuchenbuch's device. This medium is composed of an agar gel including a mineral suspension and/or a nutrient solution (Hinsinger et al., in press). As a consequence of the dilution of the solid phase, rhizosphere is magnified along its third dimension. This scale expand of the distance from root surface authorizes an easier sampling of rhizospheric material.

Applied to potassic nutrition of rape supplied with a potassium salt or a K-bearing phyllosilicate, it allows to measure how deep the K depletion might be in the close vicinity of roots and to what extent K dynamics might be shifted by root absorption activity.

#### Material and Methods

The culture device was adapted from Kuchenbuch and Jungk (1982). It provides a planar root mat through developing a high density of plants on a 30 μm pore diameter polyamide net (Hinsinger et al., in press). After growing 200 seeds of rape (*Brassica napus* cv. Crésor) within the upper part of the culture vessel, for 8 days on deionized water, roots completely covered the net: its surface was then assumed as a "macroscopic root surface". The root mat was then put in close contact with the lower part of the culture vessel for 1, 2, 3 or 4 days of cropping in a growth chamber (25°C 16 h-day period/20°C 8 h-night period; 70% relative humidity; 600 μE m<sup>-2</sup> s<sup>-1</sup> light flux). This lower part contained a cylinder of agar gel of following composition: agar agar 10 kg m<sup>-3</sup>, NH<sub>4</sub>H<sub>2</sub>PO<sub>4</sub> 2 10<sup>-3</sup> M, Ca(NO<sub>3</sub>)<sub>2</sub> 2.5 10<sup>-3</sup> M, Fe-EDTA 10<sup>-4</sup> M, H<sub>3</sub>BO<sub>3</sub> 10<sup>-5</sup> M, MnCl<sub>2</sub> 2 10<sup>-5</sup> M, ZnSO<sub>4</sub> 2 10<sup>-7</sup> M, CuSO<sub>4</sub> 2 10<sup>-7</sup> M, (NH<sub>4</sub>)<sub>6</sub>Mo<sub>7</sub>O<sub>24</sub> 3 10<sup>-8</sup> M. The pots were placed into vessels containing the same nutrient solution for preventing the agar cylinder from dehydration.

In experiments A and B, K and Mg were furnished as soluble salts added into the agar gel, as follows:  $MgSO_4$   $10^{-3}$  M in both cases, and  $K_2SO_4$  5  $10^{-3}$  M in experiment A, and  $K_2SO_4$   $10^{-3}$  M in experiment B. In experiment C, K and Mg were exclusively furnished as constitutive K and Mg of a phyllosilicate included as a  $10 \text{ kg m}^{-3}$  phlogopite suspension in the previous agar gel.

After cropping, the whole plants are easily harvested for analysis. At harvest, the agar gel is rapidly removed and cut in 1 mm thick slices parallel to the "root surface", with a Ranvier type microtome and a sharp blade. For each slice, a physical extraction of an aliquote of the liquid phase constitutive of the agar gel is obtained through following procedure, after Marschner et al. (1987): the slices are freezed at  $-25^{\circ}$ C, then heated at  $60^{\circ}$ C for 30 minutes and finally centrifuged at 19 000 g for 2 minutes. Through this extraction sequence, the volume of the supernatant which is finally obtained is about 50% of the initial volume of each agar slice. Its K concentration is then assayed by flame emission photometry.

#### Results and Discussion

The extraction method was tested through measuring K concentration in a range of agar gels prepared with different solutions of known K concentration. The results, presented in the Table 1, show that the method is accurate, and that its sensibility is the one of flame emission photometer measurement.

Fig. 1a presents the results of experiment A with  $5 \cdot 10^{-3}$  M K after 1, 2, 3 and 4 days of rape cropping. These results show an important depletion occurring under root activity of rape plants. The depletion zone reaches the bottom of the pot (20 mm) after only one day of cropping. Within 1 to 4 days of cropping, K concentration is decreased from 5 to  $0.2 \cdot 10^{-3}$  M in the first millimeters of the rhizosphere and from 5

Table 1. Measured concentrations of K,  $10^{-3}$  M, in initial KCl solutions and in the supernatants extracted from agar gels made with the KCl solutions.

K concentration in solution	0.0	0.1	0.2	0.4	0.6	0.8	1.0	2.0	5.0
K concentration in agar gel	0.0	0.1	0.25	0.5	0.7	0.9	1.0	2.05	5.15

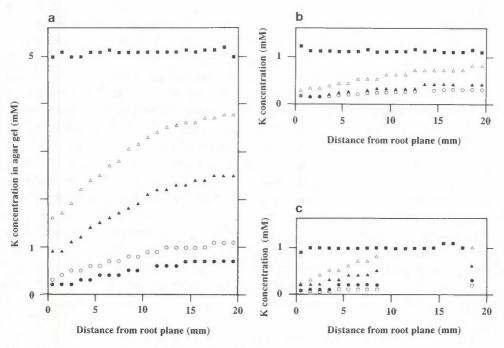


Figure 1. K concentration profiles as a function of cropping time. K and Mg were furnished as soluble salts added into the agar gel in (a) and (b), and as a phyllosilicate in (c). The respective concentrations were:  $MgSO_4 \ 10^{-3} \ M$  and  $K_2SO_4 \ 5 \ 10^{-3} \ M$  in (a),  $MgSO_4 \ 10^{-3} \ M$  and  $K_2SO_4 \ 10^{-3} \ M$  in (b), and phlogopite  $10 \ kg.m^{-3}$  in (c). ( $\blacksquare$ :control pot without plants;  $\triangle$ :1 day;  $\triangle$ :2 days;  $\bigcirc$ :3 days;  $\bigcirc$ :4 days;  $\bigcirc$ :8 days of rape cropping).

to less than  $1 \cdot 10^{-3}$  M on the whole pot depth. That reveals the important sink created by K uptake, and the high value of K diffusion coefficient in agar gel.

The experiment B with  $10^{-3}$  M K (Fig. 1b) presents the same evolution, while the K stock is five time lower. The K concentration near the root plan decreases to  $0.1 \ 10^{-3}$  M after 2 days of cropping, and, then, remains constant.

In experiment C with phlogopite as sole source of K (Fig. 1c), the K profile evolution is slightly different. Exchange reactions between solid and liquid phases maintain a  $10^{-3}$  M K concentration in the agar gel (control pot without plants). As a consequence of these exchange properties of the mineral suspension, depletion zone evolution is retarded: the bottom of the pot is depleted after only two days of cropping. But, as previously, K concentration decreases to  $0.1\ 10^{-3}$  M in the vicinity of roots after 4 days, and to  $0.1\ 10^{-3}$  M on the whole pot depth after 8 days of cropping. Such a severe depletion induces an important shift of K dynamics in the mineral phase and even a concomitant weathering of such phyllosilicate (Hinsinger et al., in press).

For the 3 experiments, the comparison between K total depletion as given by integration of the measured depletion zone and net extraction by the plants show a good agreement (data not shown). The evolution of K extraction as a function of cropping duration confirms that the K supply from agar gel is diffusion controlled.

It is well known that root absorption activity modifies greatly the physico-chemical conditions of the rhizosphere (see Römheld, this congress). In the scope of investigating these particular conditions, an interesting technique is proposed. The technique is based on an experimental exacerbation of rhizosphere effects. The consequent magnification of the rhizosphere is obtained through a 2-dimensional magnification of the roof surface (after Farr et al., 1969) and through a dilution of the solid phase in an agar gel. This dilution expands the scale of the distance from root surface and, then, allows an easy determination of maximal intensity and gradients of rhizosphere effects. If data are not immediately transposable to soil conditions, this experimental device allows to point out the mechanisms involved.

Furthermore, a complete chemical analysis of the agar gel might authorize qualitative and, even quantitative investigations of rhizosphere conditions: organic and inorganic ions, pH, organic compounds. But the technique might be limited by physico-chemical properties of agar itself (buffer capacity, cation exchange capacity). The technique might then be improved by using some more chemically inert media (agarose, for example).

This paper only reports data about K depletion. They show how K dynamics might be as deeply shifted by K depletion effect in the so called rhizosphere that even weathering of insoluble phyllosilicate might be induced. This technique might be interesting for studying other root/microorganisms/mineral interactions because it allows an easy isolation of the different components of the rhizosphere in sufficient

amounts for their investigation through standard techniques of physico-chemical, mineralogical and microbiological analysis.

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