

Comparison of the Effects of Water Stress on the Root Systems of Two Cultivars of Upland Rice (*Oryza sativa L.*)

J. F. BOIS

Orstom: CEN Cadarache, DB/SRA, BP No. 1, 13115 St Paul-Lez-Durance, France

and PH. COUCHAT

Cea: CEN Cadarache, DB/SRA, BP No. 1, 13115 St Paul-Lez-Durance, France

Accepted: 11 March 1982

ABSTRACT

Neutron radiography was used to study root development in upland rice (*Oryza sativa L.*) during and after a short period of drought. The limit of resolution (approx. 0·1 mm) of the method allows for the study of the adventitious root system in rice during the tillering stage. However the resolution is not sufficient to study development of the seminal system. Enlargement of neutron radiographs shows further details of primary root system.

Thirty-six-day-old seedlings of two rice cultivars were subjected to a 4-day water stress followed by rewatering. Neutron radiography revealed dehydration and inhibition of root growth during water stress. During post-drought rewatering, the two cultivars behaved differently with regards to the secondary root growth recovery.

Key words: *Oryza sativa L.*, rice, water stress, neutron radiography, root growth, drought tolerance.

INTRODUCTION

In West Africa and especially in the Ivory Coast, rainfall is very irregular and the soil used for cultivation of upland rice *Oryza sativa L.* usually has a low water table (Gigou, 1973). Harvesting is affected when a drought period, even of short duration, occurs at a crucial time in the life cycle. This is particularly true of upland rice which is less drought tolerant than most cereals (Moorman and Veldkamp, 1978). Development of the root system helps the plant to limit drought effects, by deeper penetration into the soil during water stress and by rapid recovery of growth when conditions return to normal (Russell, 1977).

The depth of the root system in upland rice has been studied as an adaptation to drought. (Chang, Loresto and Tagumpay, 1972; Reyniers, Kalms and Ridders, 1976; Reyniers *et al.*, 1979; Purkrige and O'Toole, 1980). However few studies have been conducted on growth of the root system during stress and rewatering, because of the difficulty in making observations *in situ*.

The non-destructive neutron radiography technique allows observations to be made of a complete root system in the soil. This technique has been described by Couchat and Moutonnet (1974) and applied to study the germination of *Zea mays L.* (Couchat *et al.*, 1980), and *Glycine max L.* (Willat, Struss and Taylor, 1978; Willat and Struss, 1979). Neutron radiography is similar to the X-ray method. The object to be analysed is placed in a thermal neutron beam of a nuclear reactor. The neutron flux is modified by light elements (mainly hydrogen contained in soil water and roots) and exposes a sensitive

film. In the present study, the technique was used to study the effects of water stress on development of the root system in upland rice. After studying the reliability of this technique at different stages of seedling development, two upland rice cultivars were used to compare the behaviour of the root system after a short drought period followed by rewatering.

MATERIALS AND METHODS

The two upland rice cultivars used are widely cultivated in the Ivory Coast: Irat 13 is a irradiated-mutant having drought-tolerant properties (Reyniers and Jacquot, 1978) and Iguape cateto, from Brazil, is considered to be moderately drought tolerant (Chang, Loresto and Tagumpay, 1974).

The seedlings were grown in sand (granulometry: less than 2 mm; dry density: 1.6 g cm⁻³); irrigation water supplied the nutrients. The sand trays were made of 1 mm aluminium sheets. They were 40 cm high 24 cm wide and 2 cm thick and had one removable side. The experiments were carried out in a growth chamber under the following conditions: 12 h day at 28 °C, 70 per cent r.h. and 400 μE m⁻² s⁻¹ illumination, the night time temperature was 22 °C with 85 per cent relative humidity.

Before neutron radiography, the sand was dried by a flow of ambient air through the sand trays to a residual moisture content of about 2 per cent by weight. After the radiography, the sand was watered to saturation.

The Mirene reactor was used to obtain the neutron radiographs (Houelle, Mercier and Revol, 1975) following the technique described by Couchat *et al.* (1980). The quality of the prints allowed for suitable enlargement of the photographs.

At the end of the experiment, the root system of each seedling was entirely recovered and the spatial arrangement was maintained with the needleboard technique (Bonzon and Picard, 1969). Then, a root system photograph was taken. Root elongation and new growth measurement were carried out using tracings applied to the successive neutron radiographs. Primary or adventitious roots from the tillering layer, were distinguished from secondary roots that are first order branching (Picard and Jacquot, 1976).

In the study of root systems at different stages of development, neutron radiographs were taken on three Irat 13 rice seedlings, at 18, 28 and 48 days, respectively. They were grown under unlimited water supply. The vegetative development was as follows: on the 18-day seedling, there was one tiller with three leaves; on the 28-day seedling, one tiller with five leaves; and on the 48-day seedling, three tillers with 7, 4 and 3 leaves.

To examine the effect of water stress two seedlings (cvs Irat 13 and Iguape) were subjected to a 4-day drought by stopping the irrigation on day 36 post-sowing. Neutron radiographs were taken on day 36 (before stress), on day 40 (at the end of the stress period), on days 44 and 48 (4 and 8 days after rewatering). A control seedling was watered continually. Due to the low-water retention capacity of the sand a high degree of stress was obtained within 3 days as shown by leaf rolling and wilting. At the end of the experiment, the vegetative development was as follows: control seedling (Irat 13), two tillers with five and six leaves; Irat 13 stressed seedling, two tillers with four and five leaves; Iguape cateto stressed seedling, two tillers with three and five leaves.

RESULTS

Study of three development stages of the root system of Irat 13 seedlings under unlimited water supply

The neutron radiograph from an 18-day seedling shows the seminal root system (Fig. 1A) but not in any detail. The first days of development of upland rice roots cannot be studied by neutron radiography because the roots are too thin. However, this was possible with maize which has thicker roots (Couchat *et al.*, 1980).

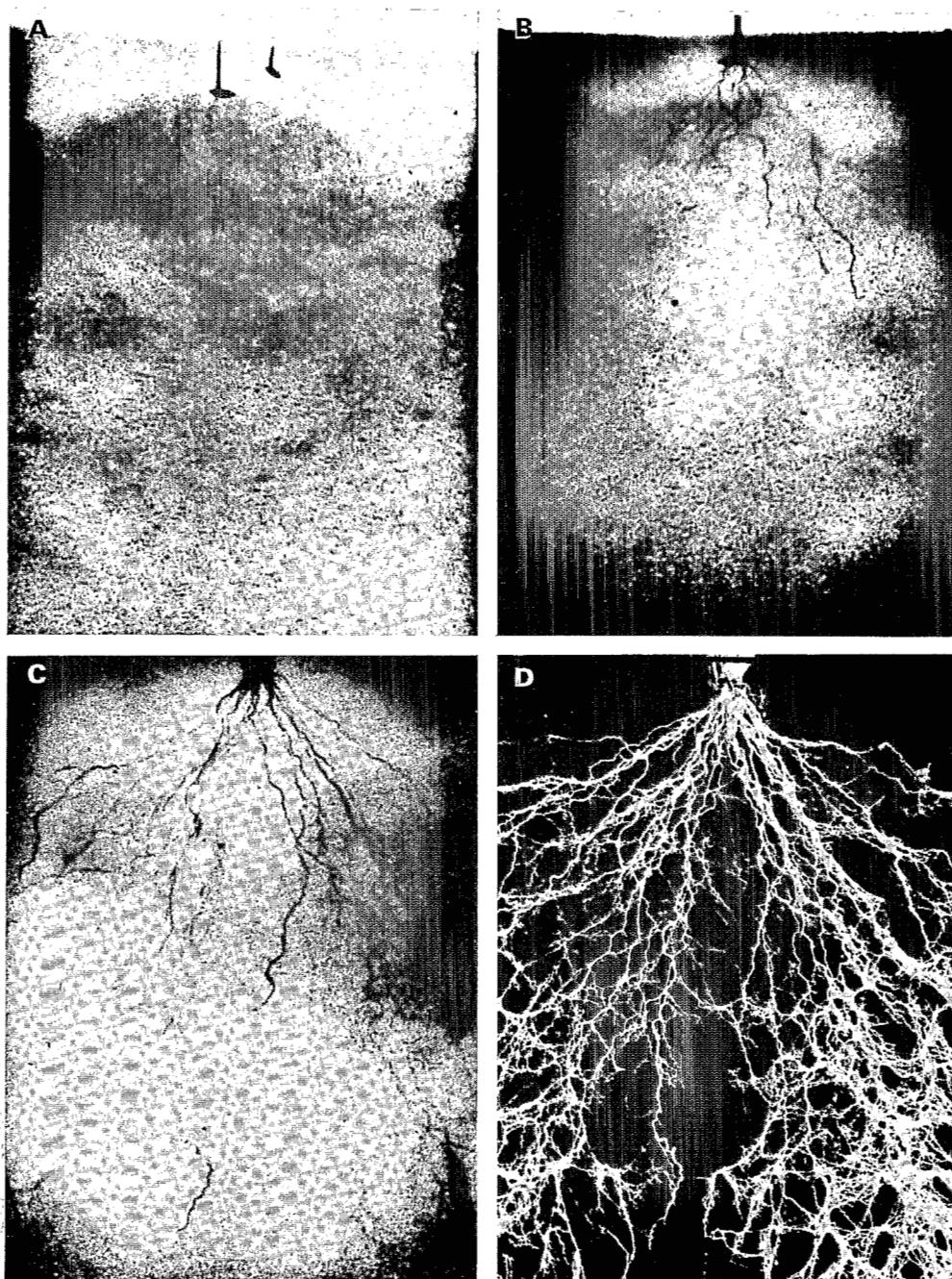


FIG. 1. Root growth of two seedlings 28 and 48 days old. A, Neutron radiograph of the 18-day-old seedling. B, Neutron radiograph of the 28-day-old seedling. C, Neutron radiograph of the 48-day-old seedling. D, Photograph of the same seedling after root system recovery.

On the 28-day-old seedling, about ten primary roots of several centimetres long were observed (Fig. 1B). Figure 1C shows a well developed root system with roots longer than 25 cm, on the 48-day-old seedling. Comparison of neutron radiograph (Fig. 1C) with the corresponding photograph (Fig. 1D) reveals that 90 per cent of the primary roots are recorded by neutron radiography. The number of primary roots counted at 3 cm from the tillering level, was as follows: 25 on the neutron radiograph, 27 on the photograph and 28 on the rice seedling. Primary roots of 0.5 to 1 mm in diameter can be observed except when there is print superposition over the print. Thinner sections and secondary roots (less than 0.1 mm in diameter) are less visible. Note that a 0.33 mm resolution has been reported by Willat *et al.* (1978). The upper portion of the print is easier to study because it is an area rich in young and thick roots and long roots with few rootlets. On the other hand, the lower portion has several intertwined and ramified roots, making study more difficult.

A closer look at the neutron radiograph in Fig. 1C reveals a dark line in the centre of several thick roots. Enlargement of the neutron radiograph (Fig. 2) shows that this dark area corresponds to the central cylinder (stele) which is richer in water content. El

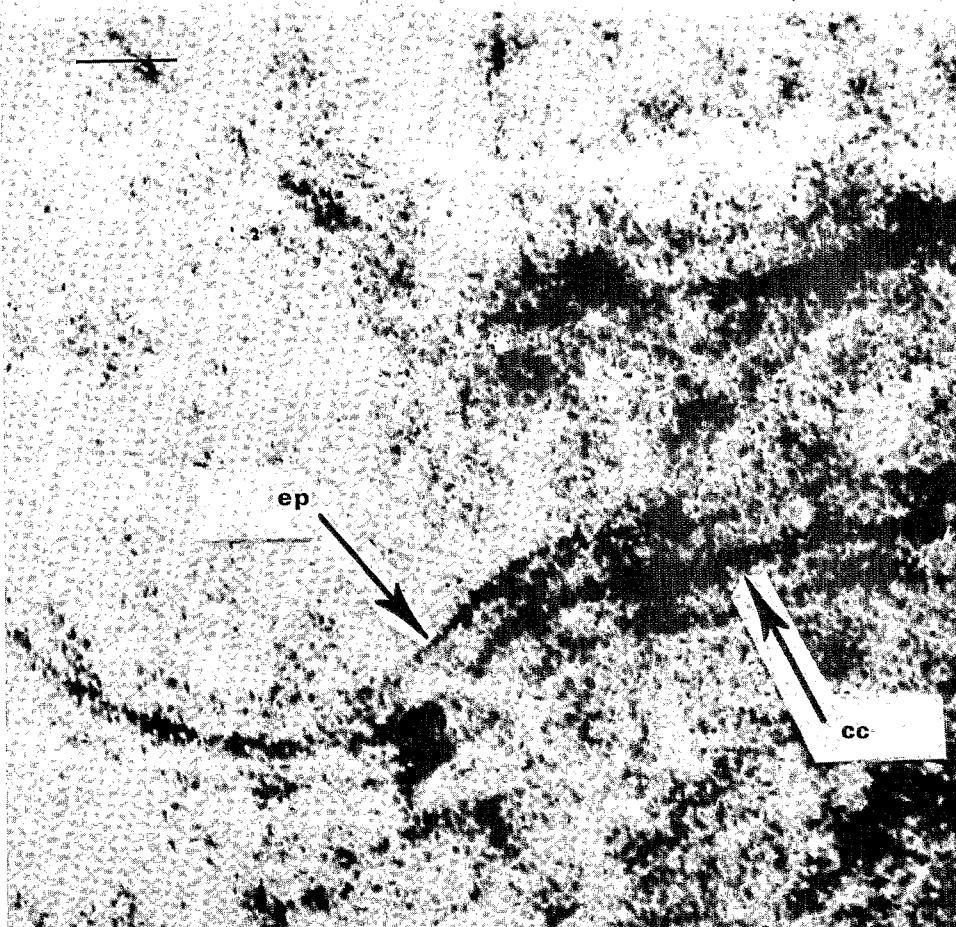


FIG. 2. Primary root structure detailed by neutron radiography. Enlargement. ep, Epidermis; cc, central cylinder. Bar = 1 mm.

Aishy (1979) estimated the stele to be 25 per cent of the root diameter, that is about 0.15 mm here, which is close to the limit of resolution of the method.

Effects of a short drought period on upland rice root development

Qualitative results. Comparison of neutron radiographs taken on days 36 and 40 post-sowing revealed, for the control seedling, the growth of several primary roots (Fig. 3A, B) whereas for the seedlings stressed from days 36 to 40, growth was almost nil (Fig. 3D, E, G, H). On day 40, some primary roots appear lighter than on the day 36 photographs; their apparent diameter is smaller which is also shown by the neutron radiograph (Fig. 4A, B). This phenomenon is a drought effect and the roots lose part of their water content. Huck, Kepper and Taylor (1970) on *Gossypium hirsutum L.* and Cruziat (1974) on *Helianthus annuus L.* and *Phaseolus vulgaris L.* reported decreases in root diameter of 30 to 50 per cent during drought. Root suberification and necrosis can also explain the lighter area, which is sometimes noticed on non-stressed seedling.

After a normal irrigation period for all seedlings, on day 44, the stressed seedlings showed some growth recovery and production of numerous more visible thicker secondary roots (Fig. 3F, I). Secondary roots formed on roots having apices that were not growing (Fig. 3H, I), (Fig. 4C, D) as well as on elongating roots. Primary root production was almost nil contrary to the observation of Picard (1973) on *Panicum*

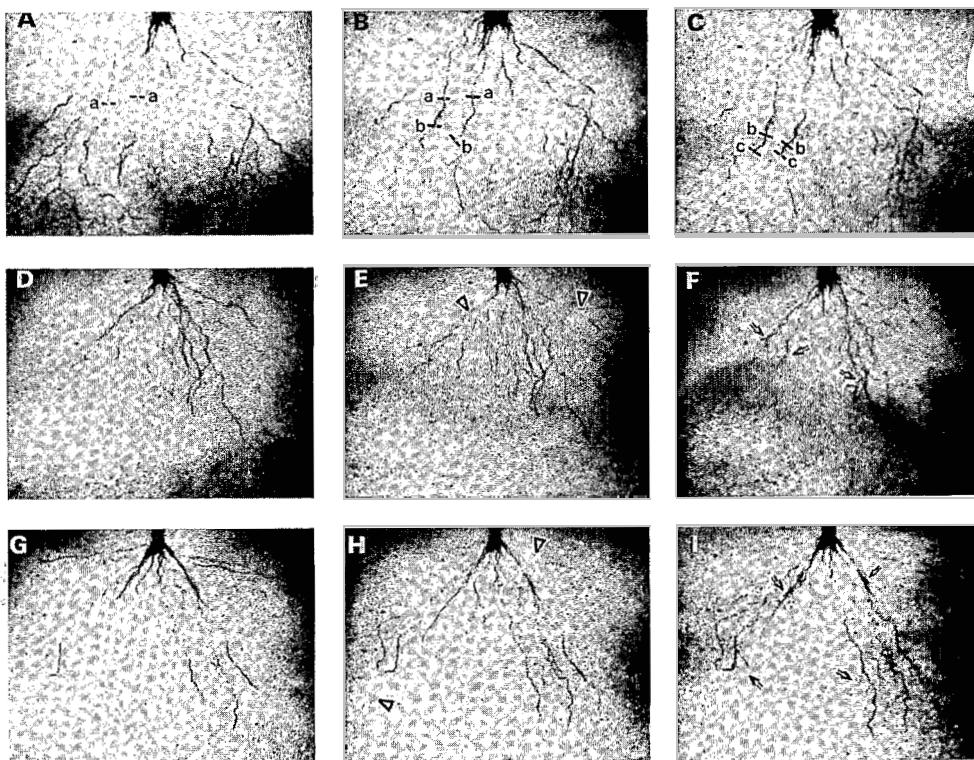


FIG. 3. Effect of a short drought period (between days 36 to 40 after seedling) on the root system of two upland rice varieties. A, D, G, day 36; B, E, H, day 40; C, F, I, day 44. A-C, cv. Irat 13 – normal irrigation; D-F, cv. Irat 13 – water stress; G-I, cv. Iguape Cateto – water stress. Arrow heads indicate area of root diameter decrease; arrows indicate new secondary root starting area; a--, b--, root elongation area.

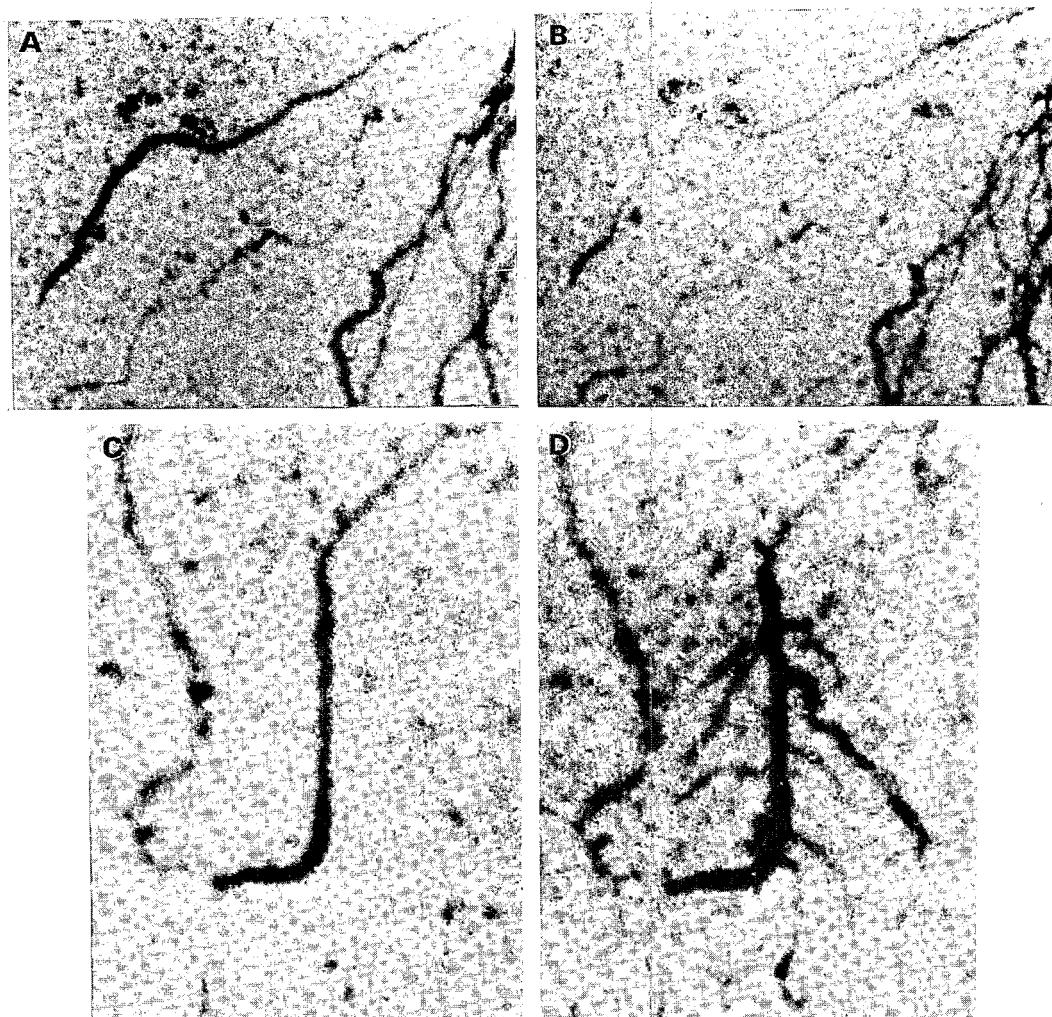


FIG. 4. A (day 36), B (day 40), Decrease of root diameter in cv. Irat 13 under water stress. Enlargement. C (day 40), D (day 44), New secondary roots after rewatering, cv. Iguape Cateto. Enlargement.

maximum Jacq. for which there was a stimulation of production when the soil was rewatered. By day 48, new secondary roots produced were longer and thinner. Primary root elongation was also observed except for one or two roots having a damaged apical meristem.

Quantitative results. The upper 12 cm of the neutron radiographs were used for measurements and counting. The number of primary roots visible on neutron radiographs represented about 75 per cent of the true root number counted at the end of the experiment on Irat 13 seedlings and 60 per cent of Iguape Cateto seedlings having thinner roots.

Table 1 shows elongation of the total root system and the mean elongation rate during the three 4 day periods. The number of observations was too low to distinguish between primary and secondary roots.

TABLE 1. Growth of the root system of three upland rice seedlings

	Period from day 36 to 40		Period from day 40 to 44		Period from day 44 to 48	
	Total elongation in 4 days (mm)	Average elongation rate (mm day ⁻¹ per root)	Total elongation in 4 days (mm)	Average elongation rate (mm day ⁻¹ per root)	Total elongation in 4 days (mm)	Average elongation rate (mm day ⁻¹ per root)
Irat 13 Control (constant water supply)	110 (12)*	2.3	90 (12)	1.9	140 (15)	2.3
Irat 13 Water stress (from days 36 to 40)	0	0	160 (18)	2.2	165 (14)	2.9
Iguape Cateto Water stress (from days 36 to 40)	8 (1)	—	435 (44)	2.5	250 (16)	3.8

* Numbers in parentheses are numbers of roots observed.

Differences in elongation rate between the three periods were not significant for the control seedling (rate: 2 mm day⁻¹). However, root extension of the other two seedlings almost stopped during the stress period. Rewatering stimulated elongation of primary roots and secondary roots (2.35 mm day⁻¹ per root) at a rate comparable to the control seedling. During the last period, the elongation rate increased for the stressed seedlings (2.9 and 3.8 mm day⁻¹ per root).

Comparison of these values can be made with field data which usually consists of overall measurements of rooting depth *vs* time: Chopart and Nicou (1976) measured the average length of the five longest roots of three rice cultivars at the three growth stages. A growth rate of 9.5 mm day⁻¹ between days 1 and 32 can be calculated from their results. Picard and Jacquot (1976) gave starting growth rates (elongation rate of young roots, smaller than 3 cm long) of 4–5 to 10 mm day⁻¹ between days 15 and 51. Truong and Beundard (1978), under aeroponic growth, measured directly the maximum length reached *vs* time: this gave a growth rate of 8.5 mm day⁻¹ between days 31 and 38 for Irat 13 cultivar.

Elongation rates measured from neutron radiography were slightly slower. This is to be expected since root penetration resistance, nil under aeroponic conditions, is higher in sand (density of 1.6 g cm⁻³) than in cultivated soil (density between 1.2 and 1.5 g cm⁻³). Moreover, measurement is obtained from a representative sample of the root system whereas the values quoted above are based on the long roots or very young roots.

Table 2 summarizes changes in the root system of each seedling during the 4-day periods: After 4 days of rewetting numerous secondary roots emerged especially on the Iguape Cateto cultivar. During the last period of unlimited water supply, production and elongation rates of roots of the stressed seedlings became comparable to the rates in control seedling.

DISCUSSION

Neutron radiography at three development stages of upland rice seedlings revealed the difficulty in studying the fine seminal root system by this technique. The method can be improved by neutron radiograph enlargement and in this case, it is well above the technique's resolution limit which is close to 0.1 mm. The seminal root system degenerates

TABLE 2. Breakdown of the changes observed in each seedling root system between the taking of two neutron radiographs

		Period days 36 to 40	Period days 40 to 44	Period days 44 to 48
<i>Irat 13</i>	New primary roots	2	1	0
Control (constant water supply)	New secondary roots	3	4	7
	Elongating primary roots	6	5	2
	Elongating secondary roots	1	2	6
	Total of growing roots	12	12	15
<i>Irat 13</i>	New primary roots	0	1	1
Water stress (days 36 to 40)	New secondary roots	0	16	2
	Elongating primary roots	0	1	3
	Elongating secondary roots	0	0	8
	Total of growing roots	0	18	14
<i>Iguape cateto</i>	New primary roots	0	2	2
Water stress (days 36 to 40)	New secondary roots	0	38	4
	Elongating primary roots	1	4	5
	Elongating secondary roots	0	0	5
	Total of growing roots	1	44	16

(Angladette, 1966) and is quickly replaced by the adventitious root system. The development of this root system during the tillering stage can be very well followed by neutron radiography. The primary root macrostructure can also be observed by this technique.

The water stress applied under the conditions described, caused, in the two rice cultivars, partial dehydration of the root system which is shown by a decrease in diameter. This water loss inhibits growth by loss of turgescence in the elongation zones (Lawlor, 1969) and by increased root penetration resistance in dry sand (Baver, Gardner and Gardner, 1972). After rewatering the substrate, growth recovery is revealed by primary root elongation and by secondary root production especially in the Iguape Cateto cultivar. In several graminaceous plants, recovery occurs first by the rapid development of new primary roots (Russell, 1977). It seems that recovery is achieved first from the existing root system. The observed lack of primary root production could have been due to an inadequate moisture content of the same medium at the tillering level. This could be tested by using soil in place of sand as the growth medium.

The different behaviour of the two cultivars could be interpreted in terms of drought adaptation. The cultivar Iguape Cateto showed a greater growth recovery after stress than Irat 13. This compensates for the delay due to the drought and can be viewed as useful adaptation. But another hypothesis can be formulated: Iguape Cateto could have a more sensitive root system, leading to a greater reaction to water stress. The two contrary interpretations reveal the complexity of the drought tolerant phenomenon resulting from different adaptation mechanisms.

ACKNOWLEDGEMENT

This work was made possible by the help of E. Bonnin (Orstom), since deceased, and by the contribution of M. Houelle of the Criticality and Nuclear Safety Research Department of the CEN at Valduc.

LITERATURE CITED

- ANGLADETTE, A., 1966. *Le Riz*, 930 pp. Maisonneuve et Larose. Paris.
- BAVER, L. D., GARDNER, W. H. and GARDNER, W. R., 1972. *Soil Physics*, 498 pp. 4th edn. John Wiley and Sons, New York.
- BONZON, B. and PICARD, D., 1969. Matériel et méthodes pour l'étude de la croissance et du développement en pleine terre des systèmes racinaires. *Cahiers Orstom, série Biol.* **9**, 3-18.
- CHANG, T. T., LORESTO, G. C. and TAGUMPAY, O., 1972. Agronomic and growth characteristics of upland and lowland rice varieties, pp. 645-66. In *Rice Breeding*. International Rice Research Institute, Los Banos, Philippines.
- — — — — 1974. Screening rice germ plasm for drought resistance. *Sabrao Journal* **6**, 9-16.
- CHOPART, J. L. and NICOU, R., 1976. Influence du labour sur le développement radiculaire de différentes plantes cultivées au Sénégal. Conséquences sur leur alimentation hydrique. *Agronomie Tropicale* **31**, 7-28.
- COUCHAT, PH. and MOUTONNET, P., 1974. Une application originale de la neutronographie: l'étude en place du développement racinaire. In *Isotopes and Radiations Techniques in Soil Physics and Irrigation Studies*, 517 pp. Proceeding of IAEA Symposium, Vienna.
- — — — — HOUELLE, M. and PICARD, D., 1980. In situ study of corn seedling, root and shoot growth by neutron radiography. *Agronomy Journal* **72**, 321-4.
- CRUIZIAT, P., 1974. Détermination des pertes en eau subies par les différents organes d'une plante soumise au déshèchement. *Annals of Agronomy* **25**, 539-54.
- EL-AISHY, S. M., 1979. Effects of water regime on growth, yield and anatomical root structure of rice plants. *Zeitschrift für Acker und Pflanzenbau* **148**, 310-7.
- GIGOU, J., 1973. Etude de la pluviosité en Côte d'Ivoire. Application à la riziculture pluviale. *Agronomie Tropicale* **28**, 858-75.
- HOUELLE, M., MERCIER, C. and REVOL, H., 1975. A mini reactor for neutron radiography Mirene. In *Radiography with Neutrons*, Thomas Telford, London.
- HUCK, M. G., KEPPER, B. and TAYLOR, H. M., 1970. Diurnal variations in root diameter. *Plant Physiology* **45**, 529-30.
- LAWLOR, D. W., 1969. Plant growth in polyethylene glycol solutions in relation to the osmotic potential of the root medium and the leaf water balance. *Journal of Experimental Botany* **20**, 895-911.
- MOORMAN, F. R. and VELDKAMP, W. J., 1978. Land and rice in Africa: constraints and potentials. In *Rice in Africa*, ed. P. Buddenhagen, 356 pp. Academic Press, London.
- PICARD, D., 1973. Incidence d'une période sèche de courte durée sur l'émission des racines primaires de *Panicum maximum*. *Compte Rendu Hebdomadaire des Séances de l'Académie d'Agriculture de France* **59**, 1475-85.
- — — and JACQUOT, M., 1976. Rythmes d'émissions comparés des racines nodales de trois variétés de riz. *Agronomie Tropicale* **2**, 156-69.
- PUCKRIDGE, D. W. and O'TOOLE, J. C., 1980. Dry matter and grain production of rice, using a line source sprinkler in drought studies. *Field Crops Research* **3**, 303-19.
- REYNIERS, F. N., KALMS, J. M. and RIDDER, J., 1976. Différence de comportement d'un riz irrigué en conditions hydriques déficitaires. *Agronomie Tropicale* **26**, 179-87.
- — — and JACQUOT, M., 1978. Démarche pour l'obtention de la résistance variétale à la sécheresse. *Ibid.* **33**, 314-17.
- — — TRUONG, B., BOIS, J. F., BONNIN, E. and THOMIN, G., 1979. Caractérisation de l'enracinement du riz pluvial in situ avec le phosphore 32. In *Isotopes and Radiation in Research on Soil-Plant Relationships*. Proceedings of IAEA Symposium, Colombo, Sri Lanka.
- RUSSELL, R. S., 1977. *Plant Root Systems: Their Function and Interaction with the Soil*. McGraw Hill, London.
- TRUONG, B. and BEUNARD, P., 1978. Etude de la croissance racinaire de six variétés de riz pluvial en culture aéroponique. *Agronomie Tropicale* **33**, 231-6.
- WILLATT, S. T., STRUSS, R. G. and TAYLOR, H. M., 1978. In situ root studies using neutron radiography. *Agronomy Journal* **70**, 581-6.
- — — — — 1979. Germination and early growth of plants studied using neutron radiography. *Annals of Botany* **43**, 415-22.