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Residual N fraction in senescent leaves is an indicator of root activity in rice

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Abstract : Studies conducted to find the effect of drainage on growth and yield of rice revealed that leaf senescence is a physiological process of aging. During the process of senescence the stored reserves are translocated to root system. Leaf senescence is also decided by the soil conditions. In well drained soils the senescence is delayed. Roots under ill-drained situation becomes poorly active and could not accept the reserves from senescing leaves. When the root systems are not active then there is higher residues of stored reserves that are left in the senescing leaves. N fraction in roots in that situation show a low status compared to senescing leaves at maturity. In well drained soils the roots are active till maturity, contain higher N fraction in roots and good root activity but lower N fraction in senescing leaves. Grain yield was more with N fractions low in senescing leaves and high in root. *(**Key words :** Leaf senesce, N fraction, Drainage, Root activity, Bio-mass partitioning)*

Rice plant at any given time is composed of leaves of physiologically different ages and this suggests that those leaves are different in their contribution to the growth of the whole plant. The leaves are different not only in age but in position relate to panicles or roots. Physical distance appears to control the direction of assimilate movement. At milky stage, the flag leaf exports assimilates mainly to panicle, whereas the lowest leaf exports a large amount of assimilates to the roots (Yosida, 1981). During physiologically active period the leaf weight continues to increase because of accumulation of proteins and starch and increase in cell wall materials such as hemi-cellulose. The leaf weight decreases at later stages of growth because of proteins, sugars and starch are translocated to upper parts. Senescence, a physiologically inevitable process is an indication of decrease in leaf weight and leaf activity. The relationship between rate of leaf senescence and grain filling is complex. In some cases, faster leaf senescence results from a faster translocation of carbohydrates and proteins from leaves to grains, which in turn may be related to faster grain filling. In other cases faster leaf senescence indicates unfavorable weather or soil conditions. Here I try to identify the relations between leaf senescence; root condition and grain yield through field studies.

Materials and Methods

Series of four field experiments were conducted at Tamil Nadu Agricultural University, Coimbatore (11°N, 77°E), India between 1990 and 1991. The soils of the experimental fields are loamy clay soils (40% clay, 22% silt, 22% fine sand, and 16% coarse sand with low available N and high P and K contents.

Experiment 1, 2, and 3 focused on the interaction between drainage (improved percolation rate), plant density and nitrogen application rates. The drainage treatments were 'non-drained' (Dn)-no special provision for improving the existing percolation rate (viz., 3.0 mm. day^{-1}) and 'drainage' (Dw)-provision of open trench (width and depth 0.60m) to improve the percolation and lateral movement of water (the mean field percolation measured after this was 12-14mm day^{-1}). Water was pumped out from the trenches. Two plant densities, viz., P1 - 50 & 66 hills per m^2 and P2 - 66 & 80 hills m^2 respectively, for medium duration, short duration varieties, were tested. The N leves were 100, 150 and 200 kg N ha^{-1} applied as prilled urea in three splits as 50% at basal, 25% at 15 or 20 days after transplanting (DAT), and the remaining 25% at 30 or 40 DAT as per the cultivar. Other nutrients applied were P_2O_5 and K_2O (50 kg each), and $ZnSO_4$ (25 kg ha^{-1}). Green manure (*Sesbania rostrata)* was applied uniformly and incorporated before transplanting at 12.5 t by fresh weight ha^{-1}, (roughly 75 kg N ha^{-1}).

Twenty five-day-old IR 50 seedlings were transplanted on 13th July 1990 in Expt 1 and 19th June in 1991 in Expt 3. Experiment 2 was transplanted with IR 20, on 3-Nov. 1990. Field remained fallow for 15 days after the harvest of the first crop (IR 50) and was then flooded for 10 days during the field preparation and the second crop (IR 20) was transplanted. A detailed root study (Expt 4) was conducted to collect information on roots under the two drainage treatments as imposed in Expts 1 to 3. The treatments comprised drained and undrained plots in combination with 120 kg N ha^{-1} (60 kg basal at transplanting; 30 kg at 20 DAT and 30 kg at 40 DAT. A factorial split plot design

with 4 replications for Expt 1-3 and a randomized complete block design with 5 replications for Expt 4 were adapted. In all the experiments adequate plant protection measures were taken throughout to avoid any incidence of pests and diseases and irrigation was provided daily to keep a level of 5 cm ponded water in all plots until 7 days before harvest. In this paper the drainage factor alone is presented and discussed for the leaf senescence.

In Expt 1-3, observations on biomass partitioning as green leaves, senescent leaves, stem, roots, and panicles was conducted at 10 days intervals starting from tillering. Five plants from sample rows were removed carefully along with roots and used for N estimation (micro-Kjeldahl method) also. The grain yields recorded were corrected to 14% moisture. Based on the color of the roots, roots were separated into active (white), moderately active (brown), and less active (black), as suggested by Cheng (1983). The proportion of differently colored roots was determined at various growth stages and expressed as percentage of the total number of roots.

In Expt 4 the oxidizing activity of the roots was determined by measuring oxidation of alpha-naphthylamine (a-NA). The a-NA oxidation is related to the rate of respiration (Ota, 1970). One gram of fresh roots was transferred into a 150 ml flask containing 50 ml of 20 ppm a-NA. The flasks were incubated for two hours at room temperature in an end-over-end shaker. After inoculation, the aliquots were filtered and 2 ml of aliquot was mixed with 1 ml $NaNO_3$ (100 ppm) and 1 ml sulphanilic acid and the resulting colour was measured by spectrophotometer.

Results and Discussion

The leaf senescence was found early in the non-drained plots (Dn) compared to drainage provided plots (Dw). An accelerated rate of senescence was seen starting form the period of root biomass disintegration. The start was early and the rate was relatively higher in non-drained fields (Dn). Seasonal variation too amplified the situation, short duration cv IR 50 cultivated during June-Sept. had early senescence than a medium duration cv IR 20 raised between Oct. and Feb. because of better rooting ability (compare root weights in table 1).

The nitrogen (N) content in dead leaves was increasing as the crop age advanced. A highest quality of 25-kg N/ha was estimated with cv IR 20 at harvest stage. Moreover, the N concentration in the senescent leaves was also increasing from 0.3 to 0.7% towards maturity, but it was significantly low under drainage (Table 1). Leaf senescence has been viewed as part of aging and that results in translocation of considerable amount of starch and sugars to other parts. In this case there is enough reason to argue that a near perfect translocation, as observed from residual N fraction (%) in senescent leaves is a process by which the roots are being nourished by a lower leaves. This may occur either when they are under active photosynthesis period or under the process of senescence.

An interesting relation existed between the rate of leaf senescence, quantity of root biomass and root N fraction. Accordingly, higher leaf senescence always accompanied low root N fraction. In these experiments total root weight increased up to heading and was greater under drained field condition. There was 14-23% loss in root weight to its maximum accumulated biomass in drainage provided field compared to 26-29% in un-drained situation.

The observation made on the root color has a distinct feature. Drainage effected 36-42% of the total roots as white in the tillering stage and continued to maintain the proportion fairly at higher level till maturity. In no-drainage the proportion was coming down sharply and there was no more white roots after heading, rather, the black root proportions were increasing (68% at maturity). The oxidized brown roots were roughly two third in drained situation compared to one third in un-drained at maturity. Variety differences were found in these proportions with more brown and white roots in the medium duration cultivar (IR 20), but in both the cultivar the trend observed was similar (Table 2). Drainage increased the grain yield substantially. The effect was clear in the first-season crop (IR 50) than in the second season crop (IR 20). There was positive interaction between drainage and root activity; and negative relation existed between root activity and leaf senescence; residual N content in the senescent leaves and root N at maturity.

Since it is well known that cytokinins, produced mostly in roots, play an important role in grain filling through better remobilization of the stored reserves (Soejima et al., 1992; Ray and Choudhuri, 1981), and leaf longevity by balancing the ABA - a growth regulator advancing leaf senescence (Soejima et al., 1992; Ray and Choudhuri, 1981), I propose here the hypothesis that *the primary effect of drainage on rice yields is via root condition and the associated level of cytokinins production in roots.* Recently it has

been reported that less cytokinins in the xylem due to poorly developed root system (Soejima et al., 1992) lead to free ABA concentration in xylem (Bano et al., 1993), which signals the stomata closure (Davies and Zhang, 1991) and also advances leaf senescence (Ray and Choudhuri, 1981).

It is also hypothesized that *the residual N fraction in the senescent leaves is an indicator of the degree of active roots systems present by that time, as lower the N fraction in senescing leaves higher the root activity and vice-versa.* Leaf senescence has been viewed as the result of N extraction from leaves and translocation to growing panicles (Sinclair, 1990). The present studies do not support this view, under the governing experimental conditions. The well-drained plots, in that case, would have shown more dead leaf mass. Moreover, dead leaf N concentration increased with crop age, reaching values of 0.7%. Highest values were found in non-drained plots. These observations suggest that senescence was not the result of N extraction from the growing parts alone. It is therefore reasonably assumed that the N extraction from the senescent lower leaves in mainly contributing to the nourishment of the roots. The lowest N fraction of a senescing leaf recorded in our experiments at the early stage of the rice crop goes to show the perfect translocation of stored reserves and also the ability of the young roots to accept the remobilization from the leaves. Thus the highest N fraction left un utilized towards maturity of the crop could be related well with aged, less active roots (Table 2). The hypothesis becomes clear when this time-course residual N fraction left in the senescent leaves is compared with the root activity of the 'non-drained' and 'drained' plots.

An increase in the N fraction in the senescing leaves from young age through maturity could be well linked with root activity wherein a root activity equal to oxidize 5.0 mg of alpha naphthylamine in one hour by one gram of fresh roots was seen in the young age and at maturity it came down to 2.1 mg hr^{-1} gm^{-1} in un-drained plots. Whereas it was 4.38 mg hr^{-1} gm^{-1} in drained plots (Table 3). Fairly a good degree of root activity is essential to facilitate the movement of reserves from senescing leaves to other parts. The grain yield in all the experiments was directly related with root biomass and root activity (Table 1).

Leaf senescence is a physiological process of aging. During the senescence the stored reserves are translocated to root system. Leaf senescence is also decided by the soil conditions. In well drained soils the senescence is delayed. Roots under ill-drained situation becomes poorly active and could not accept the reserves from senescing leaves. Higher grain yield was found with N fraction of low in senescing leaves and high in root. Differences in the measured N fractions in senescing leaves and roots at matuity may be considered for judging the 'root activity'.

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Table 1. Effect of drainage on senescent leaves and root biomass in rice.

Treatment/DAT	Expt 1 (IR 50) 60	70	80	88	Expt 2 (IR 20) 41	51	61	71	91	110	Expt 3 (IR 50) 53	63	74	84	91
N fraction of senescent leaves (%)															
Non-Drained	0.35	0.49	0.62	0.65	0.43	0.51	0.53	0.65	0.72	0.78	0.30	0.35	0.47	0.62	0.68
Drained	0.46	0.58	0.59	0.58	0.50	0.55	0.50	0.60	0.67	0.72	0.30	0.34	0.41	0.57	0.61
SEd	0.02	0.03	0.03	0.03	0.03	0.03	0.04	0.02	0.02	0.02	0.01	0.02	0.02	0.02	0.02
CD (P=0.05)	0.05	0.07	NS	0.06	NS	NS	NS	NS	NS	0.05	NS	NS	0.04	0.04	0.05
N fraction of root (%)															
Non-Drained	0.60	0.54	0.45	0.42	1.14	1.15	1.20	1.18	1.01	0.93	1.13	1.08	0.93	0.80	0.64
Drained	1.04	0.99	0.95	0.89	1.15	1.19	1.26	1.32	1.20	1.08	1.14	1.18	1.10	1.05	0.99
SEd	0.03	0.03	0.04	0.03	0.03	0.03	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.04
CD (P=0.05)	0.07	0.07	0.08	0.06	NS	NS	NS	0.09	0.08	0.08	NS	NS	0.08	0.08	0.09
Root biomass (kg/ha)															
Non-Drained	1530	1469	1359	1137	515	1191	2270	2248	1472	1199	1258	1394	1395	1331	990
Drained	1598	1604	1505	1367	543	1300	2451	2483	2110	1633	1450	1593	1572	1520	1284
SEd	45	48	40	37	15	35	61	60	53	55	42	43	42	40	31
CD (P=0.05)	NS	108	90	84	NS	79	138	135	121	125	95	97	94	90	71

Table 2. Effect of drainage on root activity as root color (%) under lowland rice eco-system

Stages	Tillering			Panicle initiation			Heading			Milk ripening			Grain
Treatment	White	Brown	Block	White	Brown	Block	White	Brow	Block	White	Brown	Block	yield (t/ha)
Expt 1 (IR 50)													
Non-drained	12.3	70.0	17.7	4.6	59.7	35.7	0.0	57.3	42.7	0.0	32.0	68.0	6.77
Drained	36.3	60.3	3.4	31.7	64.0	4.3	28.0	69.3	2.7	25.3	73.0	1.7	7.62
Expt 2 (IR 20)													
Non-drained	26.6	74.4	0.0	18.3	70.3	11.4	9.0	68.3	22.7	0.0	64.4	35.6	6.04
Drained	41.5	58.5	0.0	38.3	61.7	0.0	36.3	63.7	0.0	34.0	66.0	0.0	6.66
Expt 3 (IR 50)													
Non-drained	17.3	75.0	7.7	9.0	63.3	27.7	3.3	50.3	46.4	0.0	42.3	57.7	6.83
Drained	40.0	59.3	0.7	38.5	58.4	31	33.4	63.6	3.0	20.4	69.3	5.3	8.61

Table 3. Alpha naphthylamine oxidation of rice roots (mg/hr/gm of fresh root) Expt 4 (IR 20)

DAT	0	31	52	92	118	Grain t ha-1
Non-drained	5.04	4.99	4.12	3.10	2.10	6.50
Drained	5.04	4.96	5.02	4.83	4.38	7.39
SEd	0.09	0.072	0.074	0.067	0.052	0.21
CD (P=0.05)	NS	NS	0.16	0.15	0.11	0.46