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ORIGINAL ARTICLE

An Anatomical Study of Vascular System of Spikelet Supplying Translocates to Differentially Growing Grains of Wheat

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ABSTRACT

The characteristics of vascular system were investigated within a spikelet of wheat (*Triticum aestivum* L. var. *PBW-343*). The results divulged that the vascular system of florets 1 and 2 was more conducive to precipitation of photo-assimilates than floret 3 or 4. The whole pervading vascular system in floret 1 and 2 possibly pumped more assimilates as compared to floret 3 or other florets. The pattern seemed to be similar in all the segments of spikelets within the same ear. Further it was elucidated that floret number 3 which possessed smaller grains had longer and slender vascular bundles which probably resisted the movement of assimilates.

Key words: Partitioning of assimilates; rachilla; *Triticum aestivum* L.

Introduction

One of the key factors which directly affects the grain yield in wheat is the partitioning of assimilates between grains and vegetative biomass [1, 2, 7, 9]. Frequently, the efficiency of the vascular system for mobilizing assimilates to the growing grains has been pointed out as prime factor which could limit the grain yield [8]. However, little information is available about the ways and means used by the vascular systems for the translocation of assimilates from the peduncle to the grains which are strikingly variable in their capacity in accumulating food. The objectives of this present study were to establish the characteristics of the vascular system in the different parts of rachilla through which food was supplied to individual differentially growing grains of spikelet and correlate the same with their innate natures.

Materials and methods

The investigation was conducted with a common bread wheat (*Triticum aestivum* L. var. *PBW-343*), which was sown in circular earthenware pots (50x30x30 cm) containing 35 kg of soil mixed with farmyard manure (4:1). Eight seeds per pot were sown and after 15 days, seedlings were thinned to two. Hoagland's nutrient solution [3] was supplied to the pots. For anatomical studies of axis of spikelet, the young mother shoots inflorescence from ten replicate plants were detached at the time of anthesis from the main stem (preliminary observations had shown that all vascular tissues in the spikelet were mature by that stage). The ninth and tenth spikelet from the base, occupying a middle position on the spike and producing heavier grains, was excised. Two spikelets were analyzed fully by serial microtome and free hand sectioning, the results agreeing closely with one another. Similar studies were performed for spikelets in proximal and distal segments of spike.

All sample tissues were fixed in FAA and then dehydrated in TBA series [4]. After embedding in paraffin wax (58-60ºC melting point) longitudinal and transverse sections (8-12 micron thick) were cut with a rotary microtome (Spencer 820, American Optical

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Company, USA) and stained with safranin and fast green combination according to Purvis *et al*. [6]. Staining samples mounted in D.P.X. and photographed with Olympus photomicrography microscope and drawing prepared from Corel Draw Software (version 11) through windows XP office desktop.

Results and discussion

The axis of spikelet (rachilla) is a sinuous, notched structure and is composed of a number of short segments. In general, each segment is narrow at the base and broader at the apex, with one side being more or less convex while the other side as flattened or slightly concave. The segments are homologous to internodes (Figure 1 A, B and C).

At each node, the rachilla bore a lemma in whose axil a floret axis ascended. The lemmata were further present in alternate sequence and so were the florets and both developed in an acropetal succession. Internodes 1 and 2 were very short but relatively broad. The glumes (G_1 and G_2) and lemmata (L_1 and L_2) which the internodes sustained appeared to have condensed into one massive unit with florets (F_1 and F_2) in their axils (Figure 1 A, B and C).

A look into Figures 1, 2, 3 and 4 outlines a representation of the vascular system of a rachilla in the middle segment of the ear. They show that at the disc of insertion of floret 1 (F1), 42.7 percent of the bundles merged into one unit and formed a collective channel (Figure 1D and 2) for its possible nourishment. The balance 57.3 percent of vascular bundles remained independent for a while to organize in a circle to lead to second internode. On reaching to the disc of insertion of floret 2 (F2), 37.5 percent of the bundles again united and formed a joint supply line to second floret. Somehow, 19.8 percent of them were merged to form the vascular supply to floret 3 (F3). In contrast to internodes 1 and 2, all vascular bundles present in internode 3 formed a dead end continuous cylinder which was absorbed in the disc of insertion of floret 3. However, this cylinder also emanated accessory sub-vascular bundles equal to the number of bundles that it absorbed. These bundles which did not arise directly from the axil of the rachilla but were derived from the vascular cylinder formed at the disc of insertion of the distal florets were called as sub-vascular bundles (Figure 4E and F). This shows that floret 3 (F3) would be supplied by double the number of vascular bundles than what entered (half principle and half sub-vascular bundles) (Figures 2, 3D, E, F and 4A, B, C and D). However, the sub-vascular bundles which formed at the disc of insertion of successive distal florets became gradually smaller and were indistinguishable (Figure 1 B) e.g. the disc of insertion of floret four (F4) showed that four sub-vascular bundles were available to it (Figure 4E and F) and their size were significantly smaller than vascular bundle in basal florets.

The anatomical studies of vascular bundles of rachilla (spikelet) in two other segments of spike (proximal and distal) showed that the pattern of vascularization in different florets was similar to that of central spikelets (Figure 3A, B and C). Since the basal and distal spikelets formed lesser number of grains per spikelet as compared to middle segment, it may be the disparities in dimensions of vascular bundles in these segments which might steer this attribute rather the pattern of vascular bundles which as mentioned above was by and large same amongst them. Nevertheless, the studies hint that while florets 1 and 2 were sustained by the principal vascular bundles, the florets 3 was nourished partly by principal vascular bundles and partly by sub-vascular bundles. On the other hand, floret number 4 and 5 were supported exclusively by sub-vascular bundles, initiated at the disc of insertion of the concerned florets and hence a differential distribution of vascular bundles as compared to first three florets operated in them.

According to the pattern of vascular bundles in different types of grains (bold and small) in spikelets, there were distinct differences in vascular system of individual grains developing within the same spikelet. As mentioned earlier, the first three grains namely F1, F2 and F3 were supported by the principal vascular bundles of the rachilla, though with varying magnitude, while the vascular system of the distal grains on each spikelets (F4 or F5) were sustained by sub-vascular bundles, a sub-division of the principal vascular cylinder after traversing a long distance. Moreover, florets 1 and 2 which later on nurtured bold grains appeared to be on advantageous positions by virtue of their proximity to main vascular system. On the other hand, the vascular system of third floret, which formed small grains subsequently, consisted of a minor segment of main vascular bundle with its branched sub-vascular bundles where their sizes of vascular bundles were significantly smaller than the two bold grains.

It is elucidated that floret 3, possessing small grains had longer and slender vascular bundles which probably resist the movement of assimilate. In other words, the vascular system of floret 1 and 2 was more conducive to precipitation of photo-assimilates than floret 3 or 4. The whole pervading vascular system in floret 1 and 2 possibly pumped more assimilates as compared to floret 3 or other florets. The pattern seemed to be similar in all the spikelets within the same ear.

The unique revelations that each floret in a spikelet had an assigned quota of its supply of vascular bundles, which somehow, happened to be disproportionate in quantity and quality for each

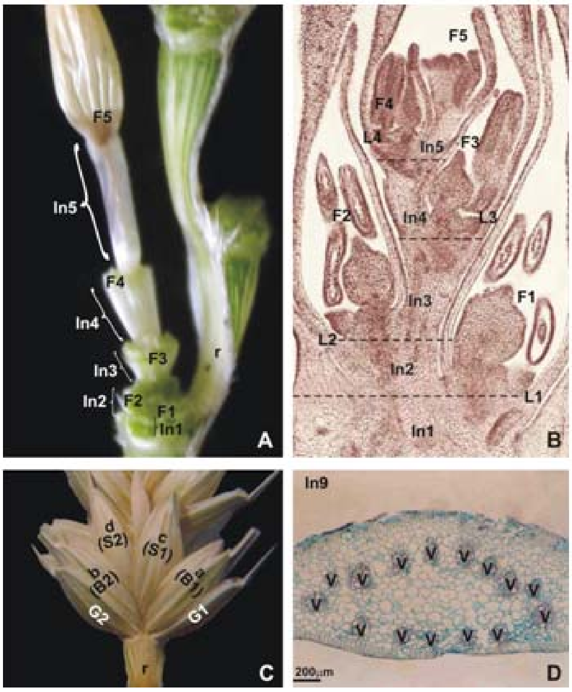

Fig. 1: A- Photomicrograph magnified the rachilla of a spikelet to show the region of the florets (F) attachment (In, internode); B- Longitudinal section of central spikelet (10^{th} spikelet from the base) showing the internodes of the rachilla, and lemma (L) of individual florets (F), X50; C- Photomicrograph showing a segment of the rachis (r) and the region of spikelet attachment. The two sterile glumes (G) and the paleas of the two lower florets (a and b) as bold grains and two upper florets (c and d) as small grains are evident; D- Microtome cross section of the rachis internode number 9 in the middle segment of rachis with 15 central vascular bundles (V).

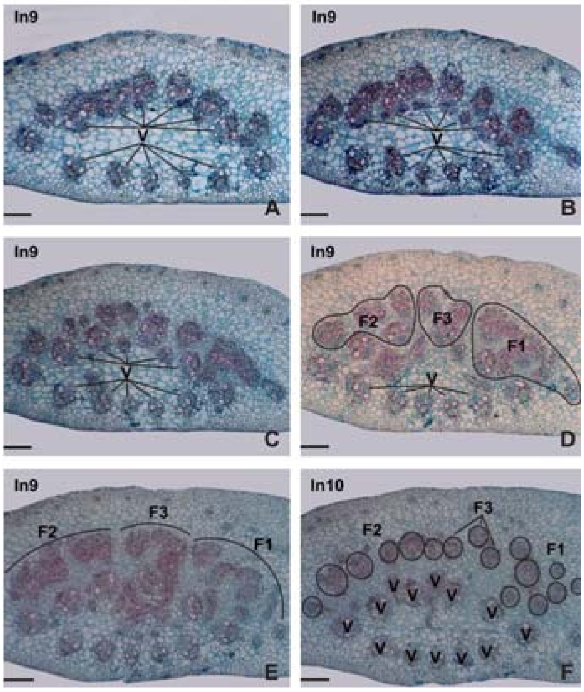

Fig. 2 A-E: Serial microtome transverse sections of vascular bundles (V) in the rachis internode number 9 in the middle segment of a spike (the attachment point of spikelet number 10) showing the stages of branching of vascular bundles to individual florets (F1 to F3) of spikelet. F- Microtome transverse section of internode number 10 showing the number of central vascular bundles (13 central bundles). Scale bar = 200 μm

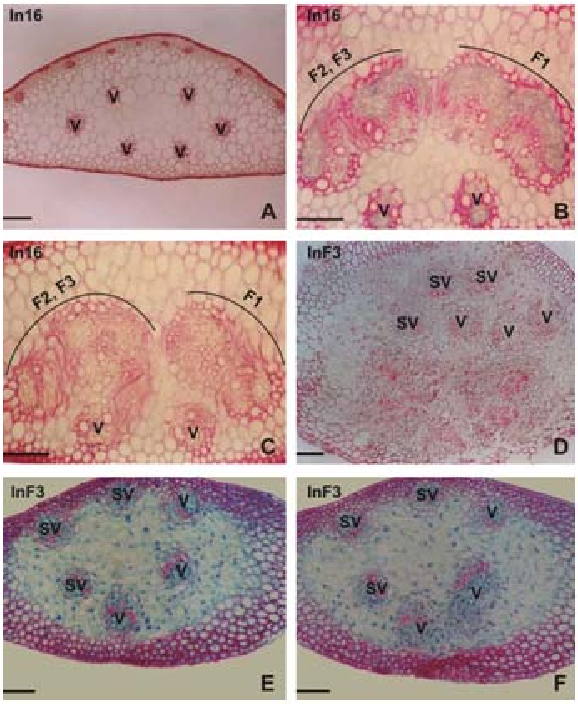

Fig. 3: Transverse sections of rachis and rachilla. A-C: Hand-cut cross sections of vascular bundles in the rachis internode number 16 in distal segment of spike (the attachment point of spikelet number 17) showing the distribution of vascular bundles in individual florets of spikelet (F1 to F3); D-F: Serial microtome sections of central (V) and sub-central bundles (SV) of rachilla in the internode of floret number 3 (InF3) showing distribution of vascular bundles supplying floret number 3 (small grain) on the spikelet. Scale bar = 100μm

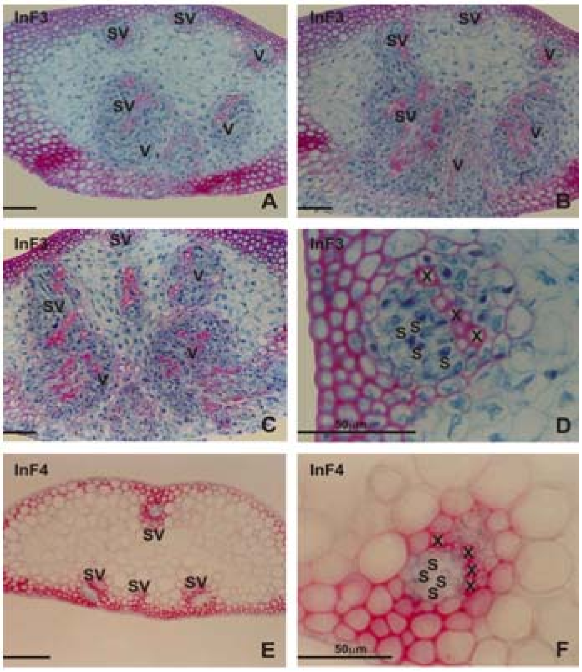

Fig. 4: Serial microtome transverse sections of vascular bundles of rachilla. A-C: The distribution of vascular bundles supplying floret number 3 (small grain) on the spikelet in the middle segment of spike; D: Transverse section of sub-vascular bundle (SV) of floret number 3 with high magnificent (X, xylem vessels; S, sieve tubes); E: hand cut cross sections of sub-vascular bundles (SV) of rachilla in the internode of floret number 4 (InF4) showing distribution of vascular bundles supplying floret number 4 (small grain) on the spikelet in the middle segment of spike; F: Transverse section of sub-vascular bundle of floret number 4 with high magnification. Scale bar = 100 μm

spikelet e.g., the supply of vascular system to florets one and two was preferentially better while the same got truncated to floret three or even still worse to floret number four, wherever they were present. This means that the latter florets (number 3 or 4), inherently became the less privileged and deprived by virtue of their positions in a spikelet and ultimately were the homes of small grains. The report of Lopez-Garrdio *et al*. [5] advocating the hypothesis that the genetic improvement in grain yield in wheat has not simultaneously kept pace with the evolution of an efficient vascular system for the mobilization of assimilates to grains is most pertinent in the present context.

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