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The Effect of Transplant in Sweet Maize (*Zea mays* L.) II. Container Root Restriction

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Abstract: The present study was undertaken to (a) evaluate the use of the *shrunken-2* mutants instead of the standard sweet corn through a transplant routine management, (b) determine the mechanism involved in post transplant sweet maize root restriction and (c) show the effect of container cell volume on vegetative growth which let explain previous field results. Sweet maize mutants hybrids Canner (*sugary*) and Butter Sweet (*shrunken-2*) were sown by direct seeded at 1, 2, or 3 L pots or in plastic plug trays (70 cells tray^{-1}) for transplant. The results suggested that acceptance of Butter Sweet *shrunken-2* sweet corn mutant would not be able to limited by germination and field emergence. By the other hand, the decrease in shoot growth observed in transplanted maize seedlings would be related to a previously container size root restriction which would not be an extreme case of mechanical impedance. A mechanism associated to changes in the synthesis or translocation of cytokinin from the root apex has been hypothesized.

Key words: Direct-seeded, root restriction, mechanical impedance, sweet corn, *Zea mays* L.

Introduction

Consumer quality of fresh sweet corn is affected greatly by the sugar and soluble carbohydrate content of the kernels. Standard sweet corn is homozygous for the recessive allele *sugary* (*su*), which results in greatly increased levels of water-soluble polysaccharides and twice the sugar content of field corn. Super Sweet cultivars are homozygous for the recessive allele *shrunken-2* which are commercially combined with *sugary* and increased sweetness

The kernel properties that play an important and interactive role in the poor emergence associated with the *se1* and *sh2* endosperm mutations include: (1) insufficient seedling energy reserves due to reduced starch concentration; (2) the condition of the mother plant and seed maturity at harvest; (3) pericarp cracking during seed maturation and solute leakage during germination; (4) membrane damage associated with high osmotic potential from elevated kernel sugar concentration and rapid influx of water during imbibitions; (5) reduced endosperm starch hydrolysis during germination and seedling growth due to the lowered activity or amounts of α-amylase; (6) reduced endosperm to embryo dry weight ratio and (7) susceptibility of the kernels to infection by fungal seed rot pathogens, particularly *Fusarium moniliforme*.

Field corn and sweet corn have been transplanted experimentally in an attempt to improve stands and hasten maturity (Khehra *et al*., 1990; Miller, 1972; Waters *et al*., 1990; Wyatt and Mullins, 1989). However, transplanting sweet corn remains a questionable practice because it increases production

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costs and often stunts plant development with similar yield than direct seeded on unstressed environment and a decrease in plant yield under latter sowing date (Rattin *et al*., 2006). However, little information is available on whether transplanting sh_2 corn can increase stand establishment and hasten maturity compared with the standard practice of direct-seeding.

Sweet maize does not transplant well because pruned roots do not branch and root replacement is generally poor compared with crops such as cabbage (*Brassica oleracel* L. capitata Group) or tomato (*Lycopersicon esculentum* L.) (Loomis, 1925; Waters *et al*., 1990). Wellbaum *et al*. (2001) showed that transplanted corn had a less extensive root system. The inability of corn roots to regenerate after transplanting resulted in stunted plants.

Materials and Methods

Sweet maize mutants hybrids 'Canner' (*su1)* and 'Butter Sweet' (*sh2*) provided by Semillería Basso (Argentina) were sown by direct seeded (D) at 1, 2, or 3 L pots or in plastic plug trays (70 cells tray^{-1}) (29.66 cm^3 cell^{-1}) (T) using a river waste-base media on 7 November 2004. Seedlings were transplanted fifteen days later.

Plants were grown under greenhouse facilities at Buenos Aires University campus (Argentine) (34°28'S) during 2004 spring season with a weekly soil fertilization of 200 ppm N (1N:0.4P:0.8K:0.4Ca) and were irrigated with a high quality tap water (pH: 6.64 and electrical conductivity of 0.486 dS m^{-1})

Plants were harvested each seven days after transplant stage (fifteen replicates); they were dried at 80°C for 48 h and weighed. Leaf area was determined by drawing around the leaves on paper, cutting out the shapes, weighing them and then calculating the area from the known weight per unit area of the paper. The following regression line of a sample of measured values of A (leaf area), w (leaf width) and *l* (leaf length) to estimate leaf area was used.

$$A\ (\text{cm}^2\ \text{leaf}^{-1}) = 0.2135 + 0.786\ (w.\ l) \qquad R^2 = 0.995$$

Samples for root anatomy were collected for each harvest date and fixed in FAA (formalin-acetic-alcohol), embedded in paraffin, sectioned at 10 or 20 μm on a rotary microtome and stained with safranin-crystal violet-fast green. Root thickness was determined from five roots per treatment with a microtome scale and a grid in one eyepiece of a microscope using a 10× objective.

Data were subjected to a one way ANOVA for a completely randomized design analysis and means were separated by the Tukey test ($p \leq 0.05$).

Results

There were no germination differences for direct seeded or plug tray sown at day 12° for Canner and Butter Sweet maize mutant hybrids; however, direct seeded showed higher vigor for the two hybrids than transplant routine (Table 1). At the transplant stage, there were significant differences in root length behalf of direct seeded plants.

Table 1: Effect of both direct seeded and transplant on germination percentage and root length at the transplant stage (14 days from sowing) for two sweet maize mutant hybrids

	Germination (%)					
	Day 6°		Day 12°		Root length (cm plant^{-1})	
	Direct seeded	Transplant	Direct seeded	Transplant	Direct seeded	Transplant
Canner	89.42a	76.04b	93.53a	91.50a	40.87a	25.03b
Butter sweet	84.53a	77.08b	89.72a	90.28a	37.47a	27.71b

Letter(s) indicate statistically significant differences ($p \leq 0.05$) for hybrids between direct seeded and transplant

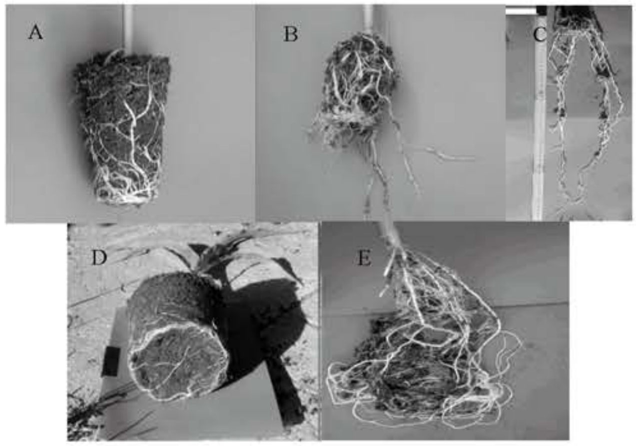

Fig. 1: Root system from sweet corn seedling grown at different container sizes. A: transplant stage, B: V_4 stage, C: V_6: stage, D: V_8 stage (container base) and E: V_8 stage

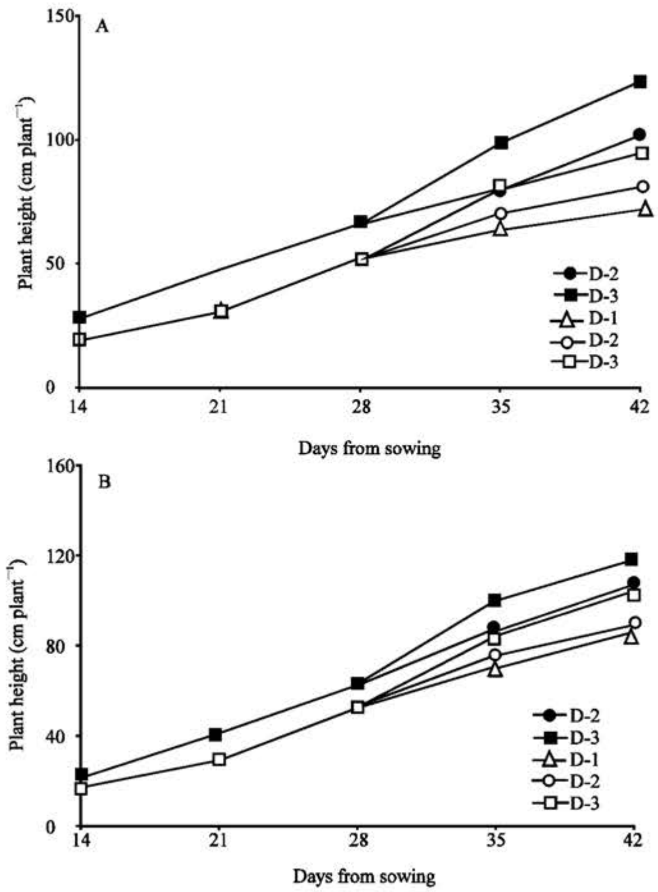

Fig. 2: The effect of container volume (1, 2 and 3 L pot^{-1}) on plant height of Canner (A) and Butter Sweet (B) maize mutant hybrids under direct seeded (D) or transplant (T)

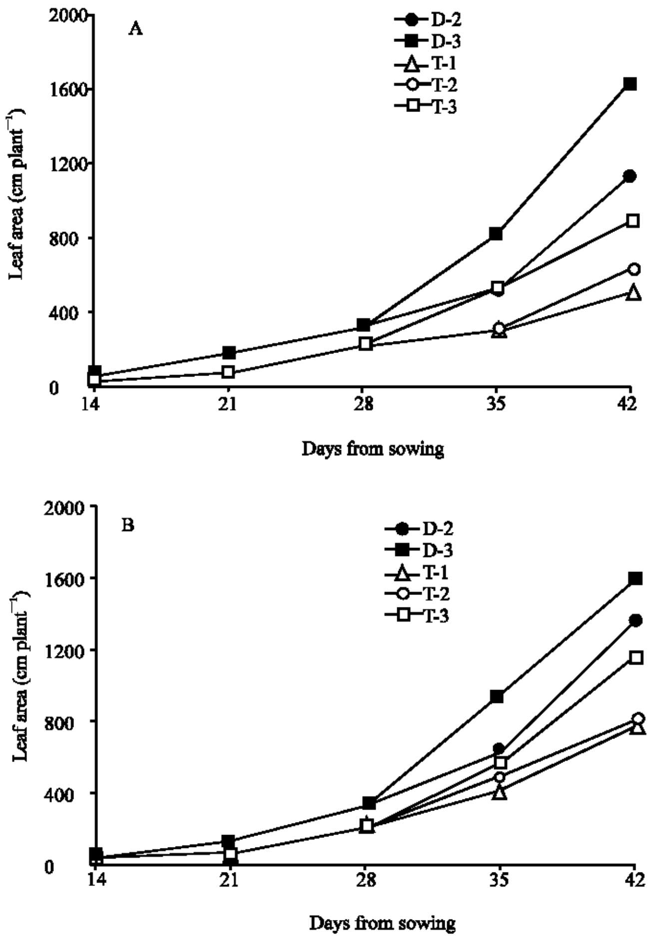

Fig. 3: The effect of container volume (1, 2 and 3 L pot^{-1}) on total leaf area (cm^2 plant^{-1}) of Canner (A) and Butter Sweet (B) maize mutant hybrids under direct seeded (D) or transplant (T)

At the transplant stage, sweet corn seedlings showed a well developed root system with white roots and without damage but the plug cell base determined horizontal root growth around it (Fig. 1A). The root restriction effects remain for the following weeks (Fig. 1B) although a new root systems was developed through the lateral roots that arise adventitiously at the base of the first internode of the stem, just above the scutellar node (Fig. 1B and C). However, the same picture was repeated when root systems from transplanted seedlings, which growing in higher container size, reached to the pot base (Fig. 1D and E).

Plants from direct seeded were higher than from transplant in each pot size during the following four weeks after transplant stage (Fig. 2); by the other hand, the larger pot sizes the higher plant height. The same result for the two maize hybrids tested was found.

Leaf area (cm^2 plant^{-1}) increased since V_3 to V_9 stage; this is, since days 14 to 42 from sowing (Fig. 3). Direct seeded plants developed higher total leaf area than transplanted plants. There were significant differences related to container size and establishment routines, especially for the Canner maize mutant hybrid.

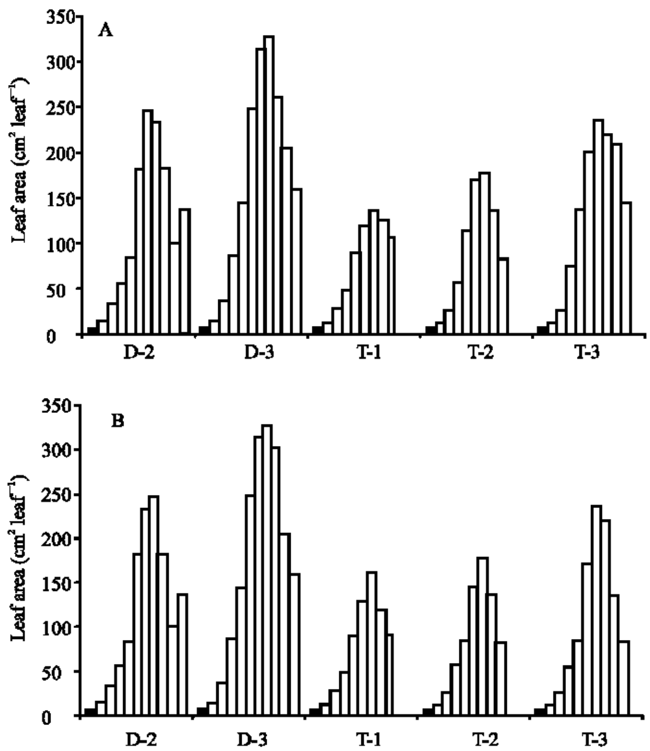

Fig. 4: The effect of container volume (1, 2 and 3 L pot^{-1}) on single leaf area (cm^2 leaf^{-1}) from the first ten leaves of Canner (A) and Butter Sweet (B) maize mutants plants initiated from direct seeded (D) or transplant (T)

Table 2: The establishment routine effects on root thickness for two maize mutant hybrids

	Root thickness (mm root^{-1})					
	Direct			Transplant		
	Day 14°	Day 21°	Day 28°	Day 14°	Day 21°	Day 28°
Canner	0.885a	1.069a	1.000a	0.995a	0.976a	1.020a
Butter Sweet	0.759a	0.878a	1.049a	0.851a	0.970a	0.986a

Letter(s) indicate statistically significant differences ($p \leq 0.05$) between direct seeded and transplant routines for each harvest date

Single leaf area (cm^2 leaf^{-1}) at the V_9 stage, plotted in Fig. 4 showed lower sizes from transplant seedlings starting from leaf 5° for the same pot sizes; differences related to pot size were observed too. Differences were higher for Canner than for Butter Sweet hybrid.

Total dry weight (Fig. 5) showed a similar pattern than for total leaf area; this are, higher photosynthate accumulation for the higher pot sizes and direct seeded. The differences were higher for Canner hybrid too. These results were associated to accumulative differences in root systems and shoots dry weight (data not shown).

A higher single leaf dry weight (Fig. 6) from direct seeded than transplant routines was found. Significant differences between container sizes were associated too, although differences were higher for Canner than for Butter Sweet hybrids.

Table 2 showed that there were no significant root thickness differences between transplant and direct seeded seedlings during the three weeks following the transplant stage for Canner and Butter Sweet mutant hybrids.

Discussion

Although introduction and acceptance of the mutant gene *shrunken*-2 (*sh*2) in place of the standard *sugary* (*su*) gene in sweet corn has been limited due to inferior seed quality, germination and field emergence (Morse, 1999; Hoyt *et al.*, 1994; Parera and Cantliffe, 1994), present results are not in agreement with previous reports because direct seeded showed both high germination percentages and vigor for the standard *sugary* (Canner) such as the *shrunken-2* (Butter Sweet) mutant hybrids (Table 1).

The success of a transplanting operation depends on the characteristics of the plant being grown and the climatic conditions to which it will be exposed after transplanting.

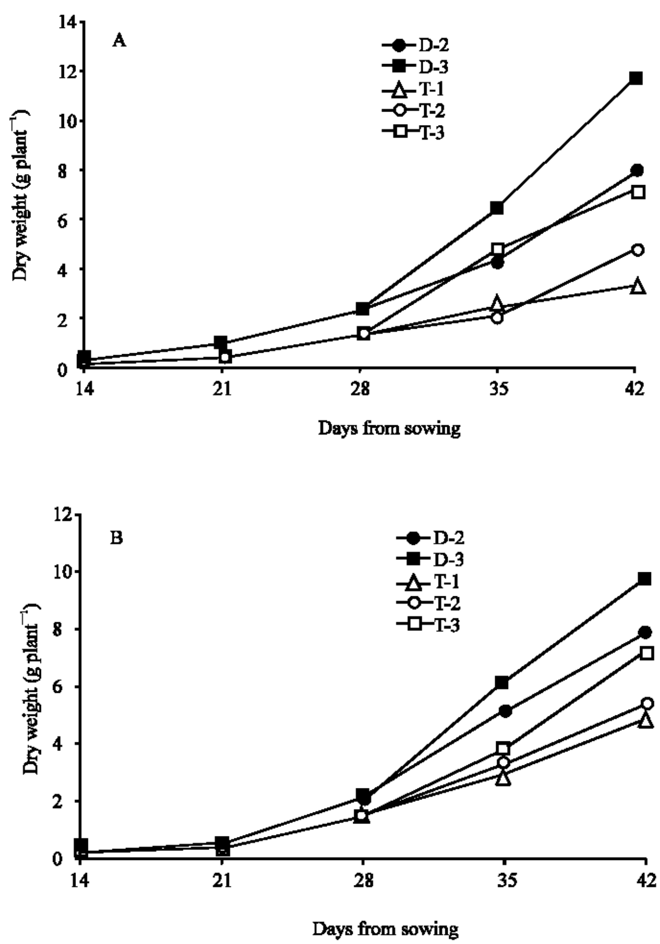

Fig. 5: The effect of container volume (1, 2 and 3 L pot^{-1}) on total dry weight (g plant^{-1}) of Canner (A) and Butter Sweet (B) maize mutant hybrids under direct seeded (D) or transplant (T)

A limited plug tray volume from sowing give a vertical root restriction even under a wide water, nutrient and oxygen supply (Fig. 1A); the same effect would be seen when higher containers was used (Fig. 1B-E). The post-transplant root restriction effects have been previously documented for other species (Di Benedetto and Klasman, 2004, 2007).

The root system of a corn seedling has seminal roots that consist of the radicle or primary root and a variable number of lateral roots that arise adventitiously at the base of the first internode of the stem, just above the scutellar node. The seminal root initials are present in the embryo and are the most important for early growth and establishment. Welbaum *et al.* (2001) showed that seminal roots were broken during transplanting as they were pulled from transplant trays or transplanted, because the polystyrene trays were not optimal for corn transplant production. However, Rattin *et al.* (2006) suggest that it was possible to obtain similar or highest yield from sweet maize mutant plants using a transplant method than direct-seeded under optimal environments. The disagreement with previous reports on the effects of the transplant routine would be associated with the use of plug plastic tray instead of the commonly polystyrene trays which would let avoid the damage to root system when seedlings were transplanted.

The decrease in main root length (Table 1) is a typical morphological change for mechanically impeded roots, although the fact that post-transplant growth may substantially be reduced due to the limited volume of the cells in which the plants were cultivated during the plant nursery period has been recently reported (Kage *et al.*, 2000; Di Benedetto and Klasman, 2004, 2007). Suberization and formation of cutin on the endodermal layer inhibited root formation and reduced water uptake by the

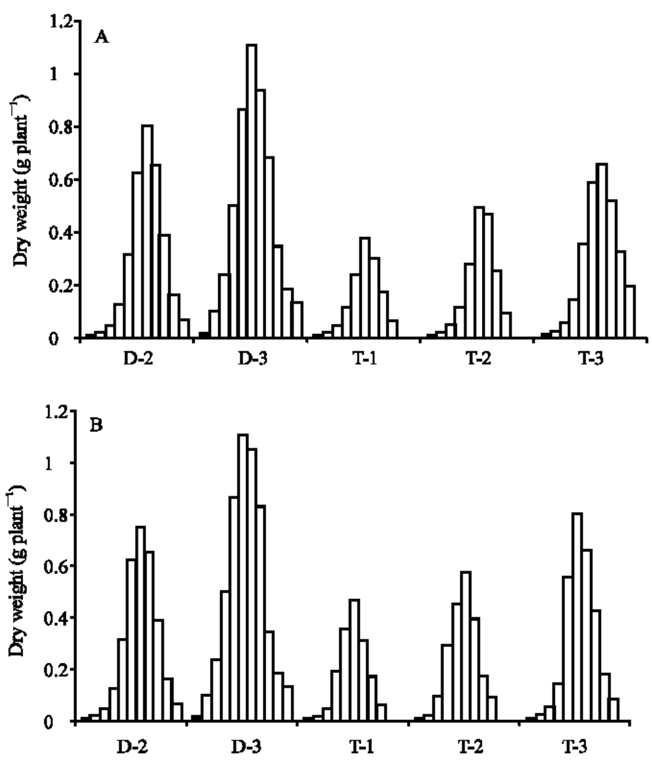

Fig. 6: The effect of container volume (1, 2 and 3 L pot^{-1}) on single dry weight (g leaf^{-1}) from the first ten leaves of Canner (A) and Butter Sweet (B) maize mutants plants initiated from direct seeded (D) or transplant (T)

roots remaining after transplanting. Species that were difficult to transplant also had a greater amount of suberization. However, the container root restriction showed in the present study for the sweet maize mutants tested (Canner and Butter Sweet) would not be the extreme case of mechanical impedance because the commonly root thickness reduction at the transplant stage (Table 2) or root suber accumulation (data not shown) could not be found.

Leaf growth is an important process in crop production systems, characterized by the production rate of new leaves, the total number of leaves produced and the rate and duration of area emergence for each leaf. Pendleton and Egli (1969) proposed one explanation for the maize transplants yielding no more than the early seedlings may lie in their shorter plant height and less leaf surface. Our results showed that transplant routine and small container size reduced total (Fig. 3 and 5) and single (Fig. 4 and 6) leaf area and dry weight, respectively which would be impose a source limitation, but, as was indicated in a previous study (Rattin *et al.*, 2006), it did not necessarily decrease grain yield.

Hormone availability (auxins and citokinins) synthesized in the root apex and reallocated to shoots would be reduced when the vertical root growth was impeded by the container base. Strong indications that cytokinins are root factors which are transported via the xylem to the shoot, where they exert a major regulatory influence on growth, photosynthesis and timing of senescence, date back to early 1960's (Kende, 1964). The indication that root cytokinins are part of the mechanism by which the shoot/root ratio is regulated can be drawn from a number of investigations: In a direct approach, in which the shoot/root ratio was decreased by lowering the nitrogen supply, a positive correlation of this ratio was found with root cytokinins content (Kuiper *et al.*, 1988) and with the amount of cytokinins exported to the shoot (Beck and Wagner, 1994). Similarly, maize roots grown under high mechanical impedance for 5 days contained three times more Indol Acetic Acid (IAA) as compared with roots grown in a loose soil. Such a rise in the IAA content was associated with inhibition of extension growth of the seminal roots (Lachno *et al.*, 1982).

In summary, the present results suggested that acceptance of Butter Sweet *shrunken-2* sweet corn mutant would not be limited by germination and field emergence. By the other hand, the decrease in shoot growth observed in transplanted maize seedlings would be related to a previously container size root restriction which would not be an extreme case of mechanical impedance. The involved mechanisms are obscures but, although a change associated to the synthesis and translocation of cytokinin from the root apex would be hypothesized, a different experimental design is needed and is the matter for the future research.

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