



# A solution for sampling position errors in maize and soybean root mass and length estimates

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## ARTICLE INFO

### Keywords:

Weighted average root mass  
Root length  
Sampling position  
Maize  
Soybean

## ABSTRACT

Root mass and length attributes are difficult to obtain in the field and currently there is uniformity among literature studies in estimating the effect of sampling position error. With the objectives of 1) quantifying the sampling position error in calculating weighted average root values per unit area and 2) developing an algorithm to minimize root position sampling error so that existing data in the literature can be used in future studies, we collected and analyzed root mass and length data across four sampling positions (0, 12, 24 and 36 cm distance from the plant row; row-to-row spacing 76 cm) from two maize and two soybean fields in central Iowa, USA. In-row sampling position (i.e., 0 cm from the plant row) over-estimated root mass and length by 66% and 46% for maize and soybean, while cores taken in the middle of plant rows (i.e., 36 cm from the plant row) under-estimated root mass and length by 34% and 23% for maize and soybean. As sampling distance from the plant row increased from 0 to 36 cm, maize root mass declined four times faster than soybean, while root length declined at almost the same rate between crops. Sampling 10 cm from the plant row provided the closest estimate to the weighted average value in both crops. We developed a new algorithm that predicts weighted average root attributes values with a  $R^2$  of 0.93 for mass and a  $R^2$  of 0.70 for length. The algorithm requires two user inputs (the measured root attribute value and the distance from the plant row). The new algorithm was tested across diverse environments, cultivars, and management practices and proven accurate for subsequent use ( $R^2 = 0.70$  and  $R^2 = 0.87$  for mass and length). This study provides guidance to strategically sample roots in future row crop research and an algorithm to eliminate sampling position bias in existing data.

## 1. Introduction

Root mass and length data are rare in the literature but are imperative to understand soil-plant-atmosphere interactions and crop adaptation to changing environments (Hirte et al., 2018). Among the few published data, there are substantial inconsistencies in the measurement protocols and assumptions used to calculate root attributes at the unit area level (e.g., weighted averages of mass roots per area) that are relevant for soil and crop modeling as well as other agronomic assessments. This problem exists because of the laborious nature of root measurements in the field that limits the number of root samples across space and time (Oikeh et al., 1999; Paez-Garcia et al., 2015). Furthermore, individual soil cores are extrapolated to unit areas by assuming root uniformity across sampling positions in row crops (Maeght et al., 2013; Nichols et al., 2016; Dietzel et al., 2017). Such an assumption may be valid when the objective is to compare different treatments,

such as crop cultivars, but not valid when the objective is to quantify carbon budgets, root/shoot ratios, calibrate simulation crop models, or compare root estimates across different studies (Amos and Walters, 2006; Fan et al., 2016).

A review of literature for 76 cm spaced maize and soybean crops (a widely used row-spacing in maize and soybean systems in the U.S. Corn Belt) revealed a substantial variation in root sampling position (Table 1). About 70% of the studies used 1–2 sampling positions and 30% of the studies used 3–4 sampling positions. The position of the root sampling substantially influences unit area extrapolations of root attributes, the weighted average. For example, Anderson (1988) showed a two-fold difference in maize root mass estimation from 0 cm (within-row) to 36 cm (between-row).

The inherent bias introduced by sampling position could be minimized if relevant information existed to guide sampling position in future studies and algorithms were created to correct for sampling

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**Table 1**

Literature studies reporting root mass and/or length data and their sampling position and depth in corn and soybean crops with ~76 cm row spacing.

Crop Species	Row spacing (cm)	Sampling distance from plant row (cm)	Sampling depth (cm)	Reference
Maize	76	20	100	Jarchow et al. (2015)
Maize	75	10, 20	70	Aina and Fapohunda (1986)
Maize	71	0, 18, 35.5	75	Mengel and Barber (1974)
Maize	76	0, 19, 38	120	Anderson (1987)
Maize	76	0, 38	30	Kaspar et al. (1991)
Maize	76	0, 38	140	Dwyer et al. (1995)
Maize	77	0, 18	60	Durieux et al. (1994)
Maize	75	0, 37.5	90	Eghball and Maranville (1993)
Maize	75	12	90	Oikeh et al. (1999)
Maize	75	0	50	Buyanovsky and Wagner (1986)
Maize	75	0, 18.5, 37.5	180	Mayaki et al. (1976)
Maize	75	0, 9.4, 18.8, 37.5	100	Qin et al. (2006)
Maize	75	15	75	Hirte et al. (2018)
Maize	76	0, 18, 36	110	Russell et al. (2009)
Maize	75	37.5	30, 150	Wiesler and Horst (1994)
Maize	75	5, 15, 37.5	50	Chassot et al. (2001)
Maize	76	20	30	Dietzel et al. (2015)
Maize	76	20	100	Dietzel et al. (2017)
Maize	76	20	100	Nichols et al. (2016)
Soybean	76	0	120	Benjamin and Nielsen (2006)
Soybean	76	20	100	Jarchow et al. (2015)
Soybean	75	5, 15, 25	60	Li et al. (2017)
Soybean	75	0	50	Fernández et al. (2009)
Soybean	76	10	120	Devries et al. (1989)
Range	71–77	0–38	30–180	
Average ± SE	72 ± 13	17 ± 14	86 ± 43	

position bias in past studies. Our review indicates that there are few studies (Gajri et al., 1994; Qin et al., 2006; Li et al., 2017) that provide guidance on ideal sampling position to obtain representative root mass estimates per unit area, however, as Table 1 shows, this guidance is rarely followed. Moreover, we did not find any algorithms in the literature to correct for sampling position bias in existing data. That limits our ability to leverage existing data to inform future studies.

To address the gap we conducted a field study where we collected and analyzed root data taken from different positions in maize and soybean row crops. Our objectives were:

- 1) Quantify the error introduced by sampling position in unit area estimations of roots traits to guide future sampling efforts
- 2) Develop an algorithm to correct for sampling position bias so existing data in the literature can be re-used in future studies

We selected maize and soybean crops for this work for three reasons. First, these crops together contribute more than \$50 billion per year to the US economy (Hatfield et al., 2014), and occupy about 73 million hectares of the US cropland (USDA NASS, 2017). Second, both crops are commonly grown at 76 cm rows apart, thus the sampling position effect is unavoidable. Third, they are morphologically different crops in their root system structure and architecture; maize has seminal roots and soybean has a taproot system from which primary and secondary roots grow and they are distributed along the length of the main root (Lersten and Carlson, 2004; Qi et al., 2012).

## 2. Materials and methods

### 2.1. Site description

Four field experiments (two maize and two soybean) were conducted at two sites in Central Iowa, USA during the 2017 growing season. The Kelley site (42°01'16"N, 93° 46'32.5"W) has a silty clay loam soil and subsurface drainage system at 1.1 m depth to remove excess moisture. The Kelley site has been under no-till management since 2009. The Boone site (41°55'13.9"N, 93°45'00.1"W) has a loam soil without subsurface drainage. The Boone site is managed with

conventional tillage. Both sites have approximately the same weather as the distance is about 10 km apart. Over the growing season the average temperature was 20 °C with a total of 510 mm of precipitation.

### 2.2. Experimental site description

At each site, experimental plots were established in a maize-soybean rotation with three replications (n = 2 crops x 2 sites x 3 replications = 12 plots). Each plot was 1664 m<sup>2</sup> and 778 m<sup>2</sup> for Kelley and Boone sites, respectively. The row spacing was 76 cm for both crops, while plant density was about 8.4 pl m<sup>-2</sup> in maize and 30 pl m<sup>-2</sup> in soybean plots. The same cultivars were used in both sites, a maize hybrid of 111-day relative maturity, Pioneer P1197AMXT, and a 3.2 maturity group soybean, Pioneer P32T16R. Planting dates were April 24th for maize and May 8th for soybean at the Boone site; at Kelley maize was planted on May 15th and soybean on May 30th. The maize plots received nitrogen fertilizer of 168 kg N ha<sup>-1</sup> according to university guidelines for a maize-soybean rotation cropping system (Sawyer et al., 2006). No nitrogen fertilizer was applied to soybeans. Other nutrients and pH were at optimum levels according to university recommendations (Mallarino et al., 2013).

### 2.3. Root sampling

We collected root samples about two weeks after physiological maturity (October 3rd) to a depth of 60 cm. This sampling depth was chosen because earlier measurements indicated that the variation in root attributes between sampling positions mostly occurs in the top 60 cm (see Fig. S2). A hydraulic probe was used to sample soil cores with 6.20 cm diameter (Giddings Machine Company, Windsor CO, USA). Four samples were taken at 0, 12, 24, and 36 cm distance from the plant row. Soil cores were divided into depth increments of 0–15, 15–30, and 30–60 cm. Samples were stored in a cold room at 4 °C.

### 2.4. Root cleaning

Root tissues were separated from soil particles using a root washing system (Smucker et al., 1982; Hirte et al., 2018; Hydropneumatic

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