





Figure S1. A ground-based brace root phenotyping robot (BRobot) was used for image capture. BRobot is a modified Superdroid LT2 Tracked ATR Robot Platform with a custom controller. The red arrow highlights the side-mounted FLIR 3.2 MP Color Blackfly camera that captures images.

RGB Image Acquisition



1434_Plant_10A1434_Plant_10B

Root Tagging GUI

main.py

Please use fullscreen

Click to draw circles, place them according to the mode you are in. hit the enter key to change modes. you are done with a plant when the text 'done, click next' appears. Hit 'r' to reset the progress made for the current plant..

Start

Click through workflow

Previous

Mode: draw triangles draw the triangles startnig with the highest point first and connect it to the point right below it (same x value). Then draw the third to the left or right of the bottom of the vertical dots.

12

11

Enter # of brace-roots lowest level goes in lowest input box.



Scale 40.79px

Root width 30.08px

Stalk width 95.26px

Triangles Complete

Next

Pixel Data

process_rootpixel_data.py

Scaled Phenotypes

Number of roots/whorl (not shown)

2 = Single root width

3 = Stalk width

4 = Height of whorl (a)

5 = Stalk-to-root grounding (c)

6 = Root angle (B)

7 = Spread width

8 = Number of whorls in soil

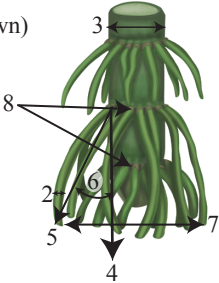


Figure S2. A semi-automated root tagging workflow was developed to optimize image processing. After RGB images are acquired, a python script (main.py) launches the root tagging graphical user interface (GUI). A tag of known size (0.5 inches) was placed on the stem prior to image acquisition for scale. First, the tag is selected for a pixel (px) scale, then each additional screen will prompt the user to click and/or record/identify specific regions in the image. The red dots shown on the image highlight the following phenotypes that have been tagged: 1) the number of pixels within a 0.5-inch region, 2) the stalk width, and 3) the single root width. The purple dots shown on the image highlight the right triangle that is used to identify the following phenotypes: 1) the height of the whorl, 2) the stalk-to-root grounding, and 3) the root angle. The number of roots within each whorl was counted and typed into the white boxes (shown on the left). This records the total number of roots per whorl and the number of whorls in the soil. After recording or identifying phenotypes, the user will hit “enter.” After all phenotypes have been recorded, the user will hit “Next” (red bar on the right of the screen) to begin the next image. After completing all images, a python script (process_rootpixel_data.py) is used to convert pixel data to scaled phenotype data. All data is exported to a .csv file for processing and analysis.

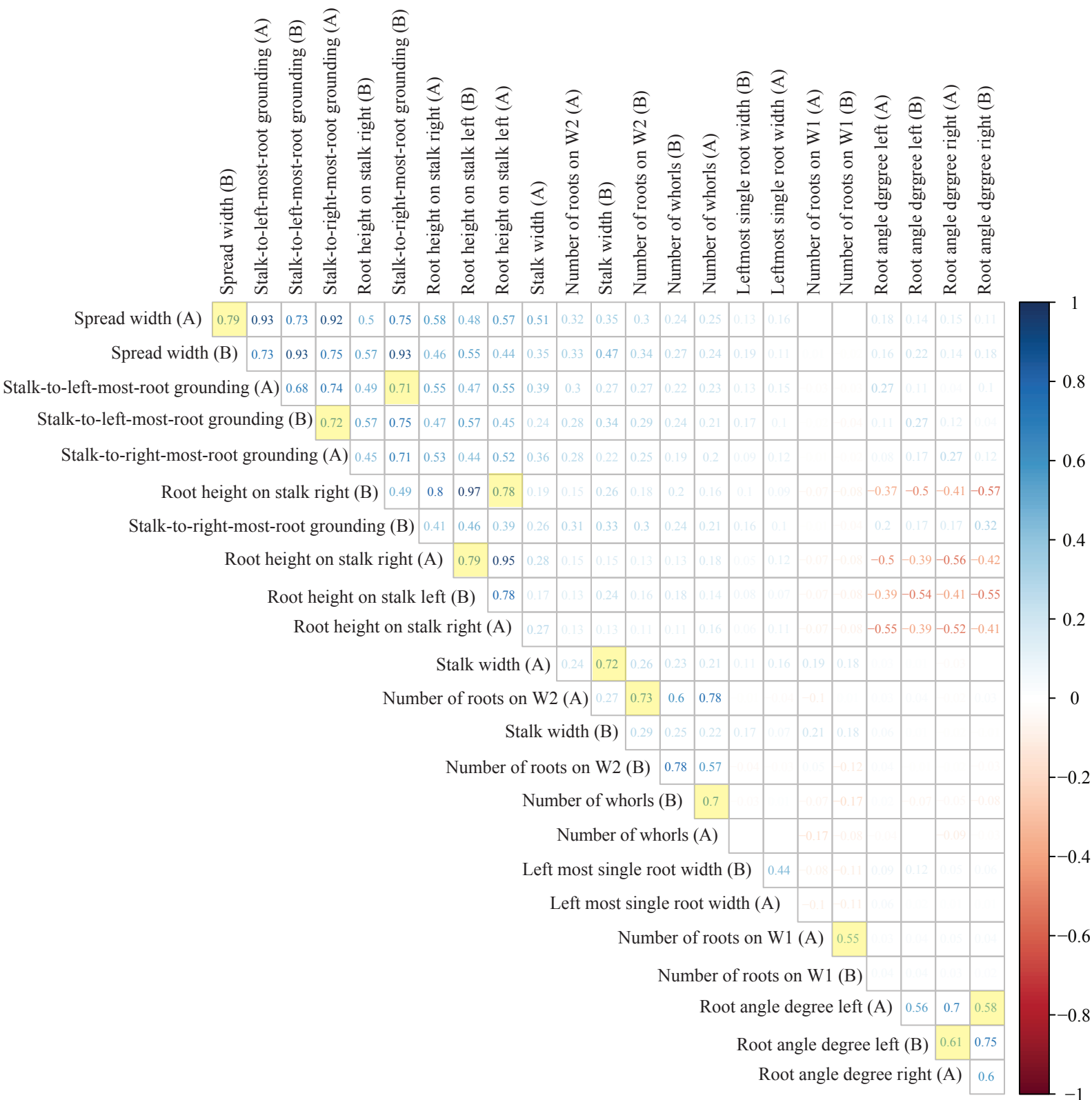


Figure S3. Phenotypes extracted from left and right images were highly correlated. Left and right images (labeled A and B) were acquired for each plant and phenotypes were extracted from images with the root tagging GUI. A Pearson correlation analysis was run to determine the precision of our root tagging GUI. Cells highlighted in yellow indicate the same phenotype from both sides of the plant.

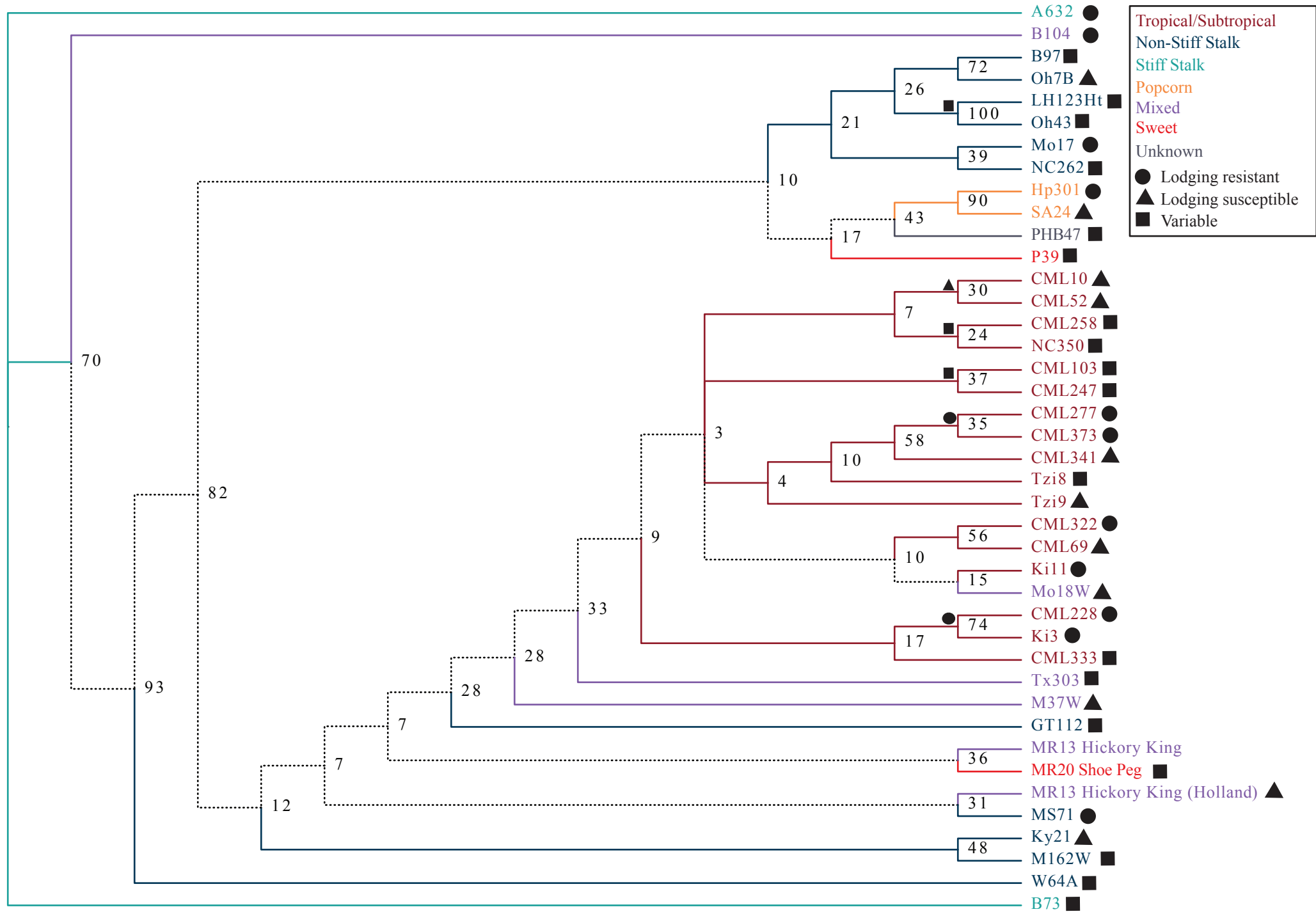


Figure S4. Root lodging is not monophyletic. A species tree was generated for 41 of the 52 maize inbred genotypes included in this study. Genotype names are color coded according to their assigned subpopulation information [Flint-Garcia et al (2005) and Liu et al (2003)]. If all genotypes within a clade are from the same subpopulation, branches are solid and colored with the respective subpopulation. Branches are dotted if the clade includes genotypes from more than one subpopulation. Genotypes were identified as lodging resistant, lodging susceptible, and variable per Figure 2. The assigned lodging classification is illustrated to the right of the genotype with a circle, triangle, or square. Clades where all genotypes within the clade are a part of a single lodging classification are highlighted with the corresponding shape at the node.

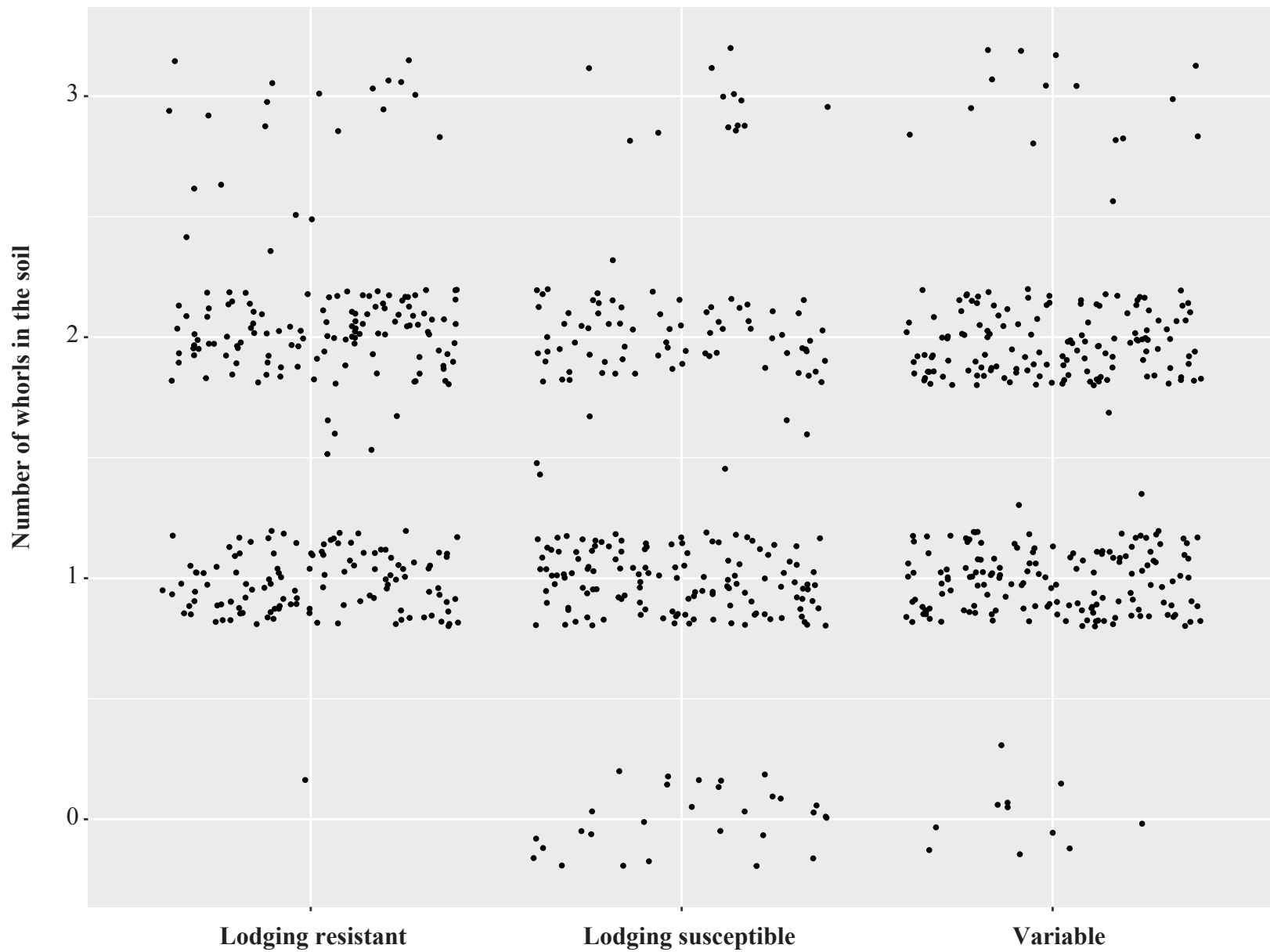


Figure S5. Root lodging resistant genotypes had more brace root whorls in the soil. Images were acquired with BRobot one day before Tropical Storm Isaias. From the RGB images, the number of brace root whorls that entered the soil was extracted. Genotypes were classified as lodging resistant, lodging susceptible, and variable according Figure 2. Genotypes that were root lodging resistant had more brace root whorls in the soil compared to those that were root lodging susceptible ($p < 0.05$).

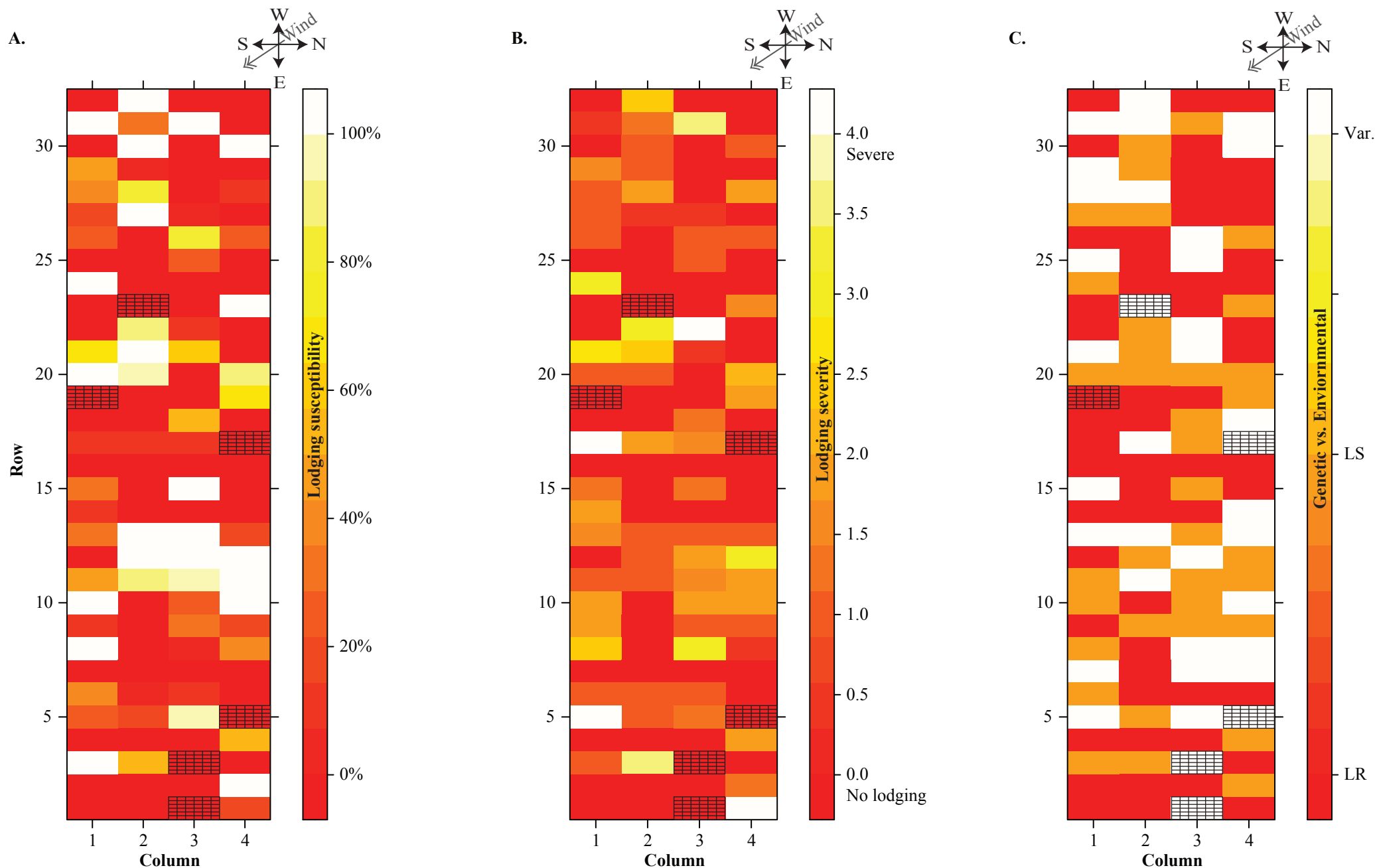
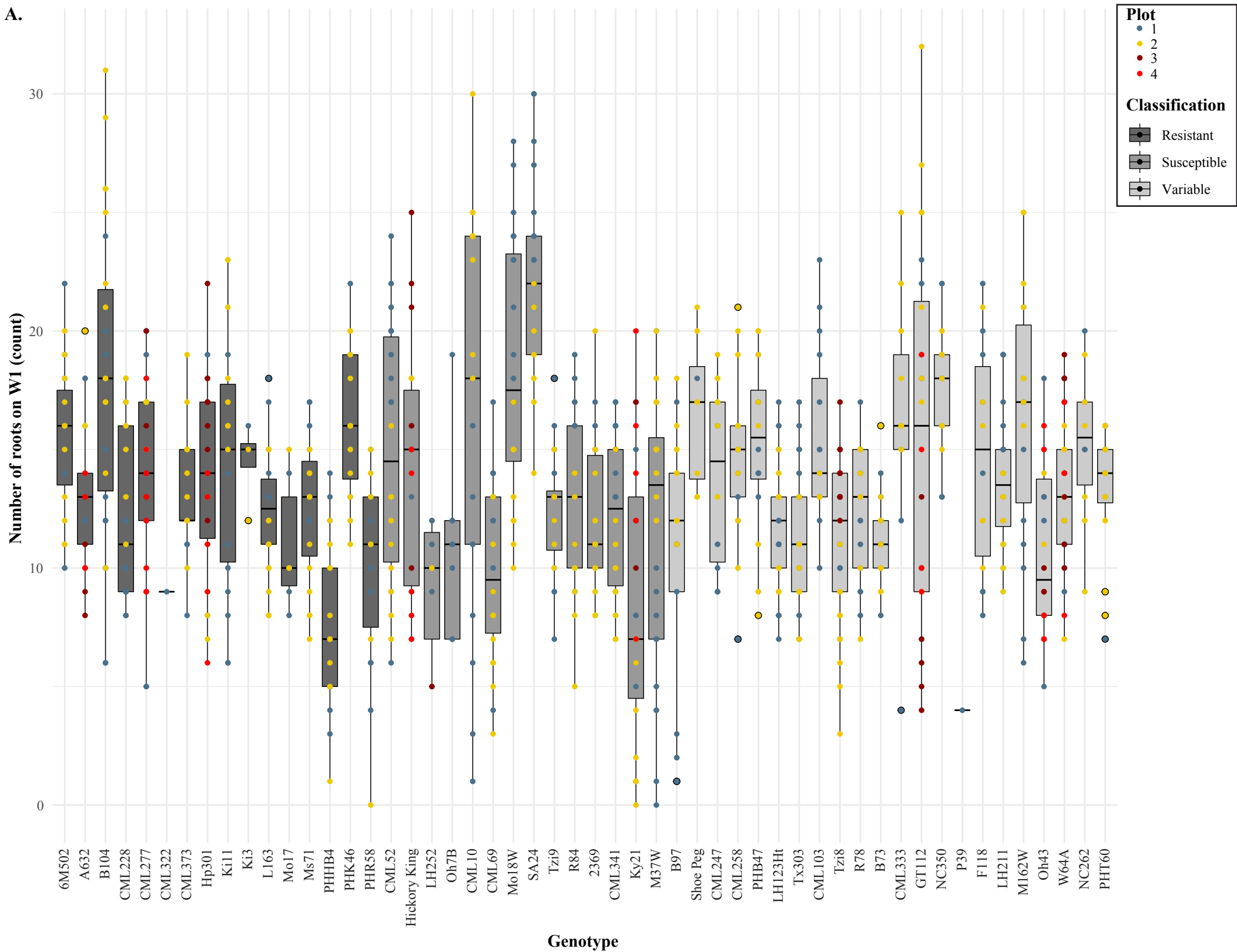
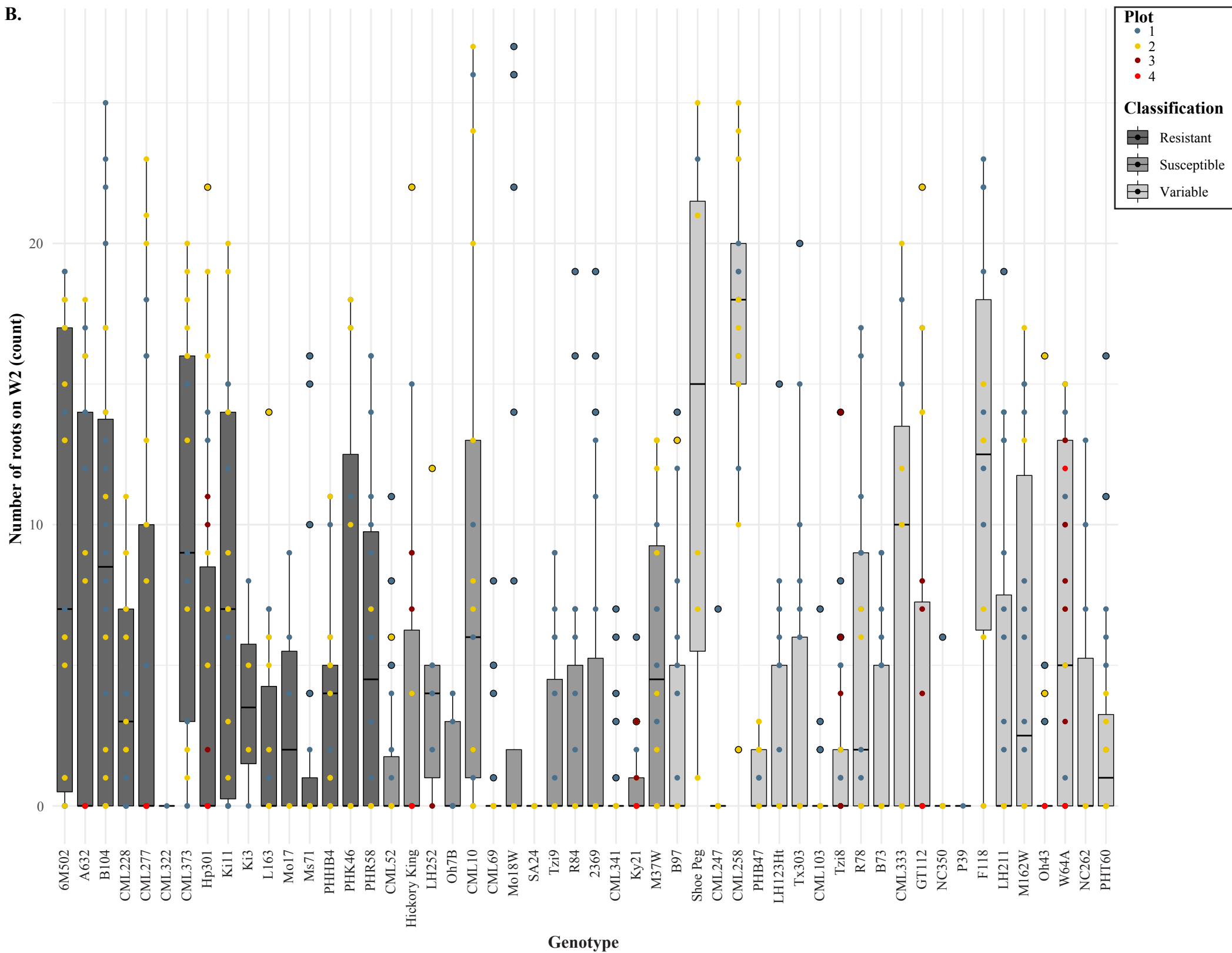


Figure S6. Root lodging was not related to field position. Winds from the West and North-West induced root lodging. (A) Plots were colored according to the susceptibility of root lodging within each plot. Plots with 100% root lodging were highlighted with white, whereas plots that had 0% root lodging were highlighted with dark red. (B) Plots were colored according to the average severity score within the plot. Plots that are highlighted in white are plots where plants were on the ground (90 degree displacement), whereas plots that are highlighted in dark red are plots where plants remained vertical (lack of root lodging). (C) Genotypes that are highlighted in dark red are genotypes that are root lodging resistant; genotypes that are highlighted in orange are genotypes that are root lodging susceptible; genotypes that are highlighted in white are genotypes that are variable among the two plots. (A-C) Plots with a grid overlaid indicate plots that did not germinate.

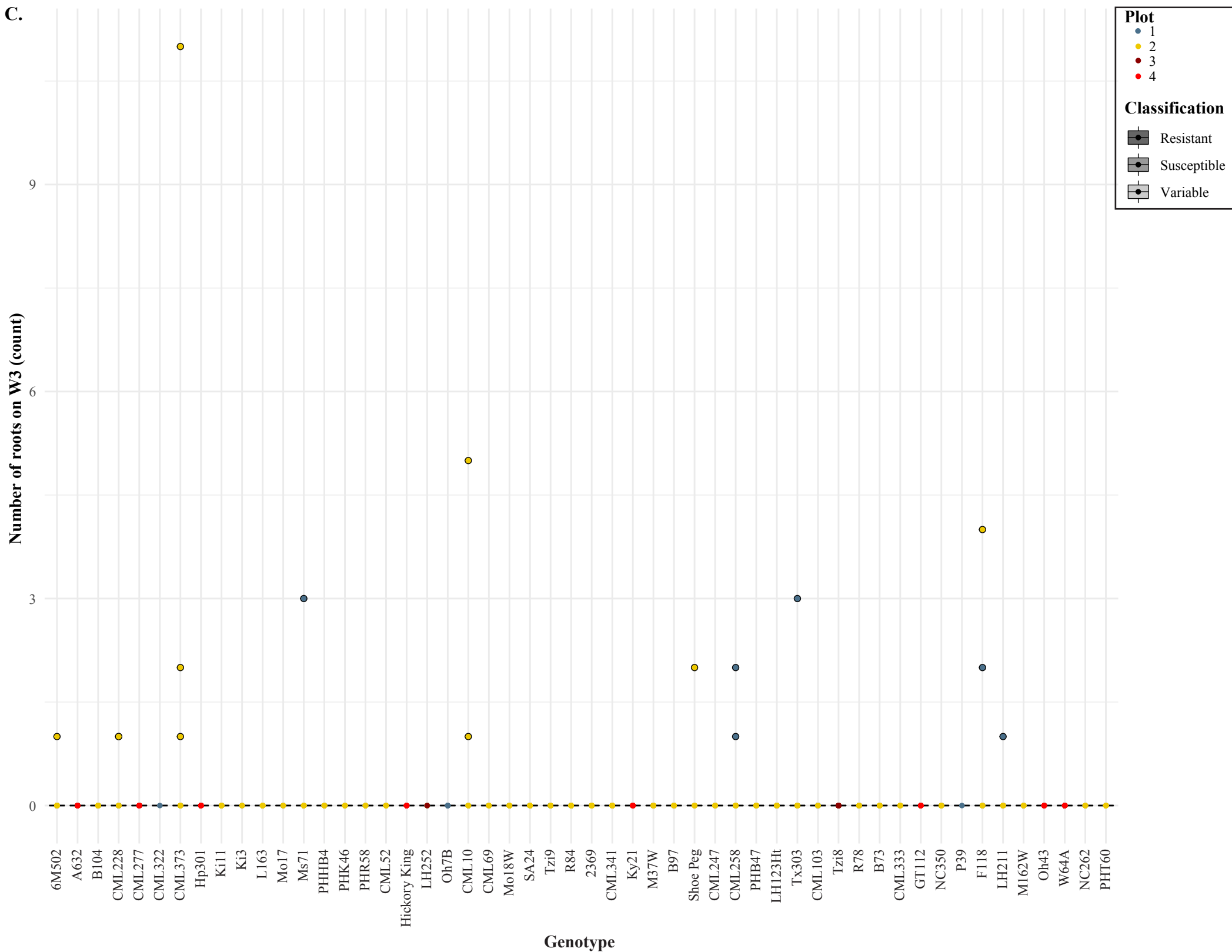
A.



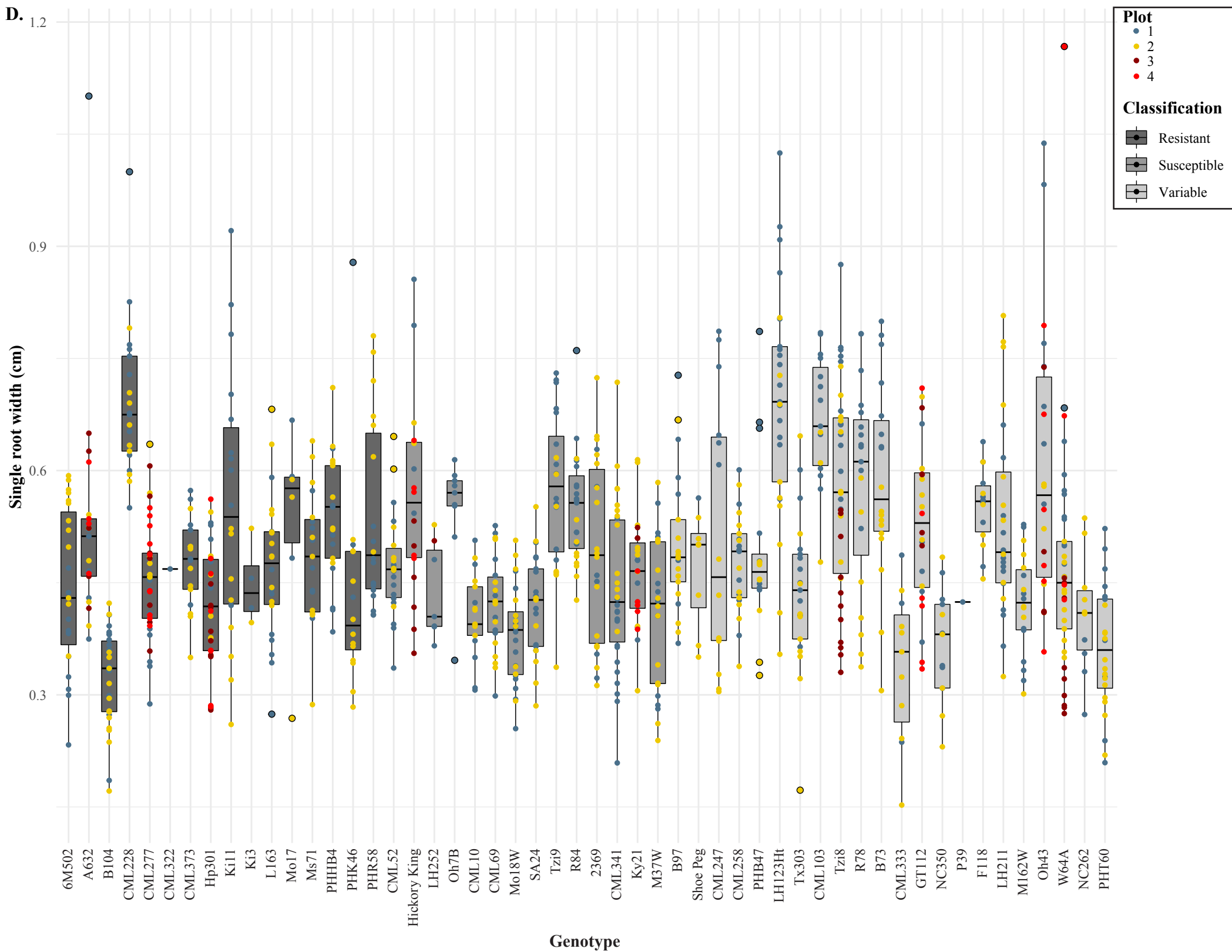
B.



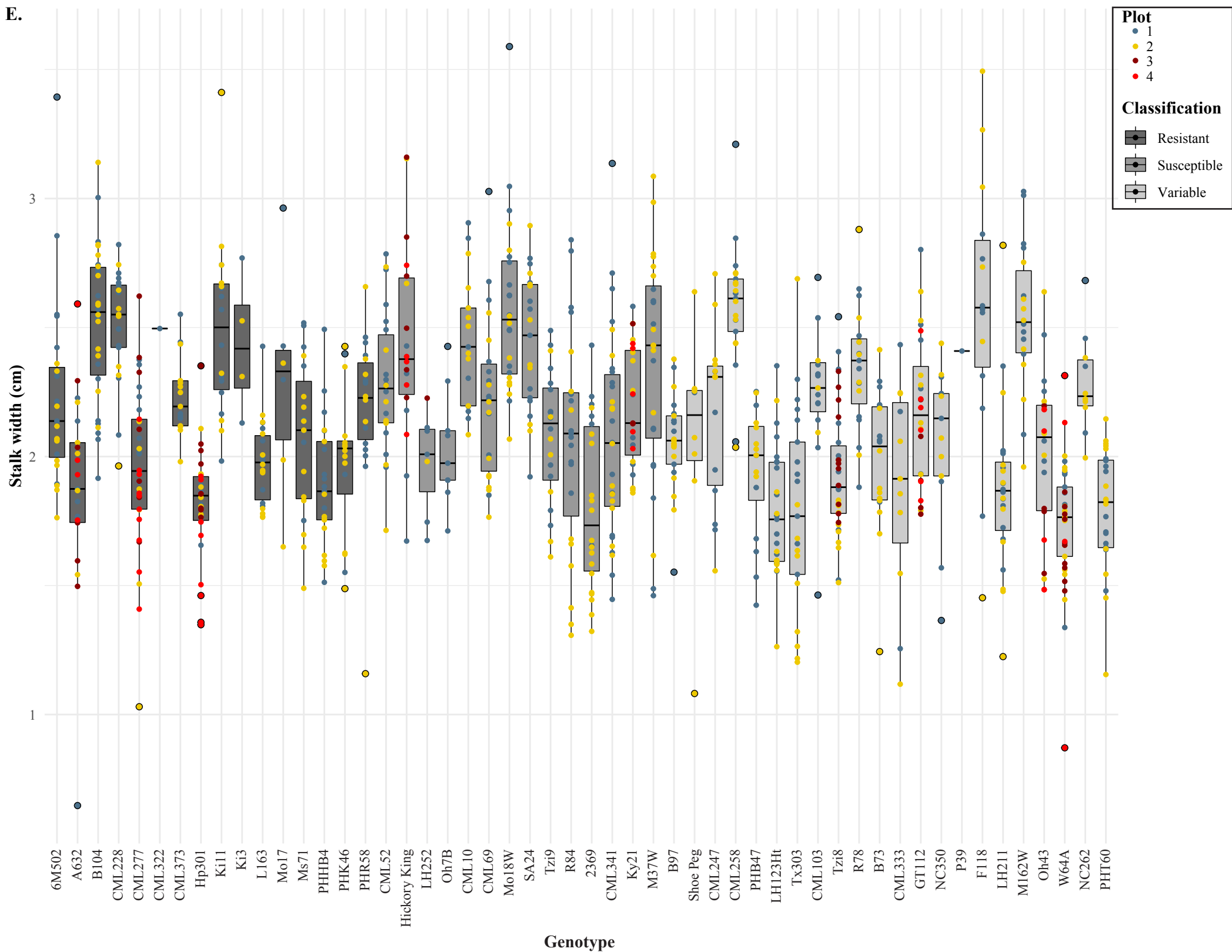
C.



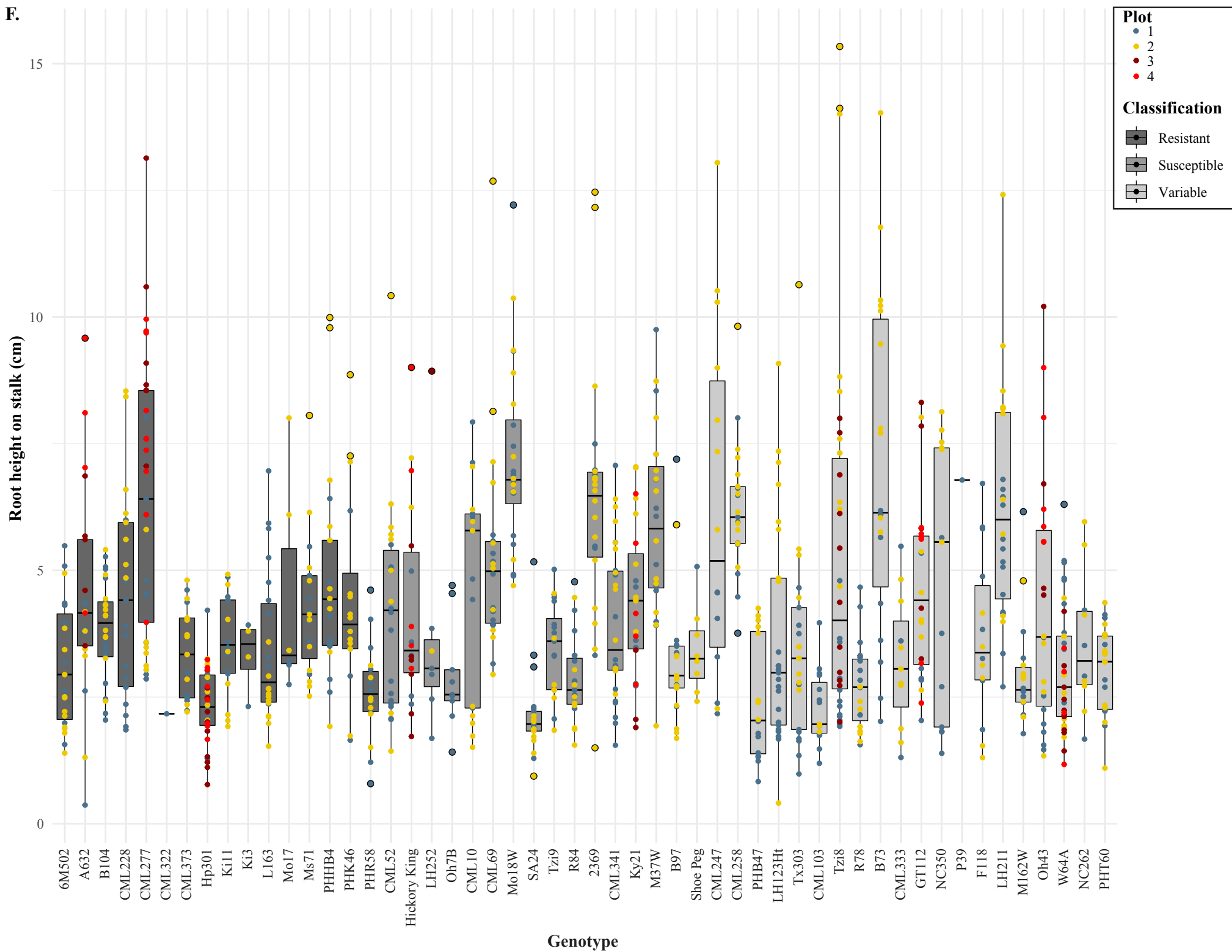
D.



E.

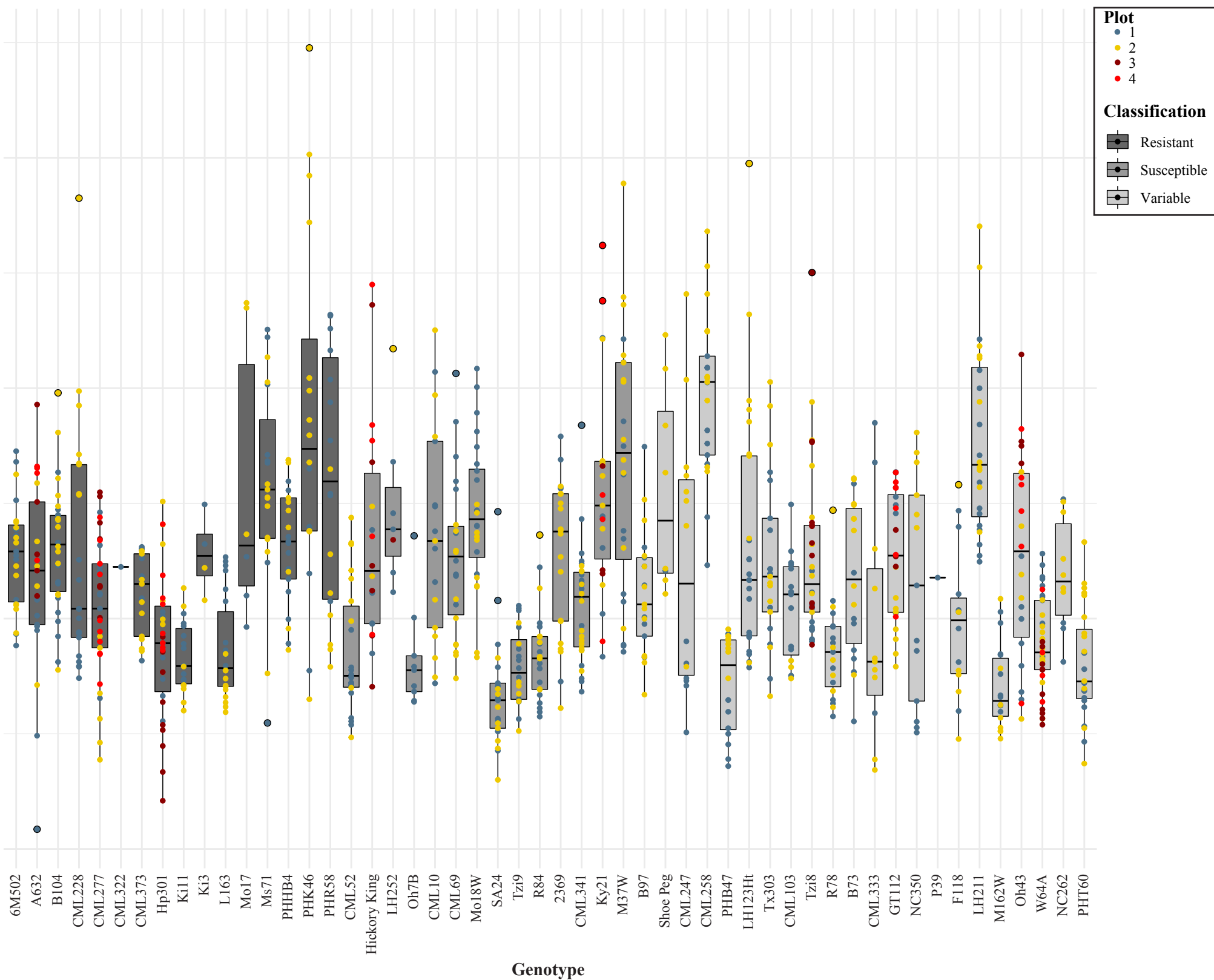


F.



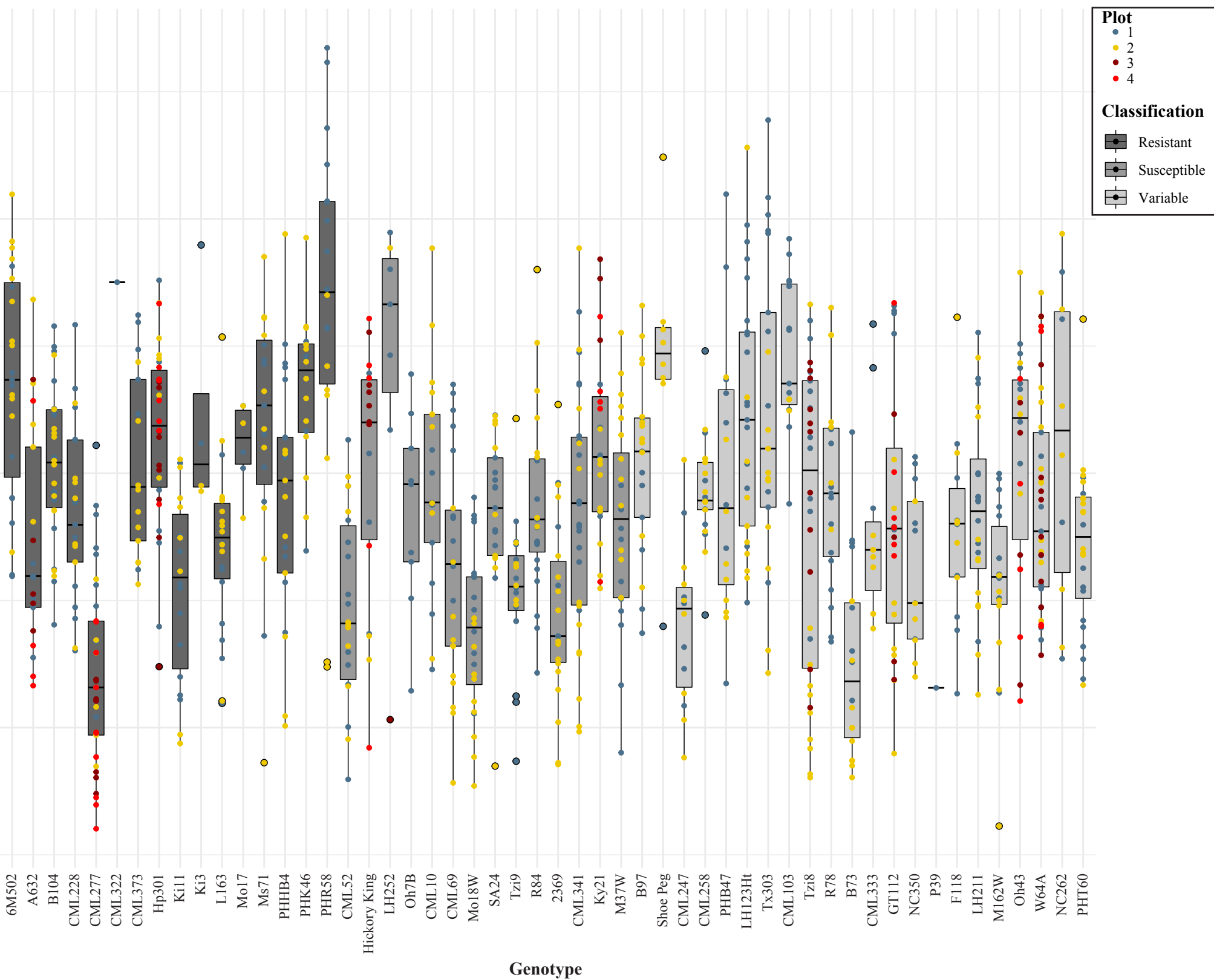
G.

Stalk-to-root grounding (cm)

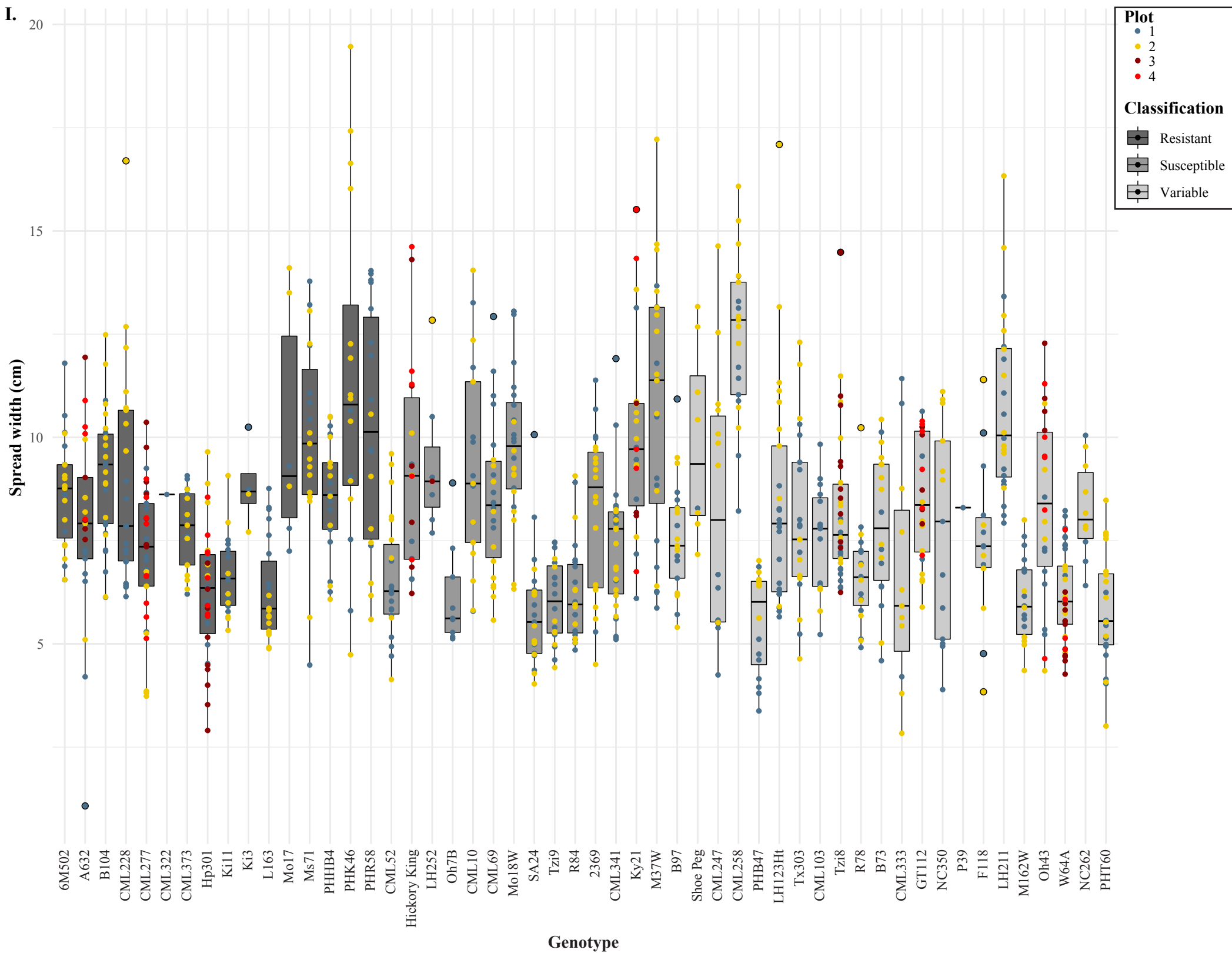


H.

Root angle (degrees)

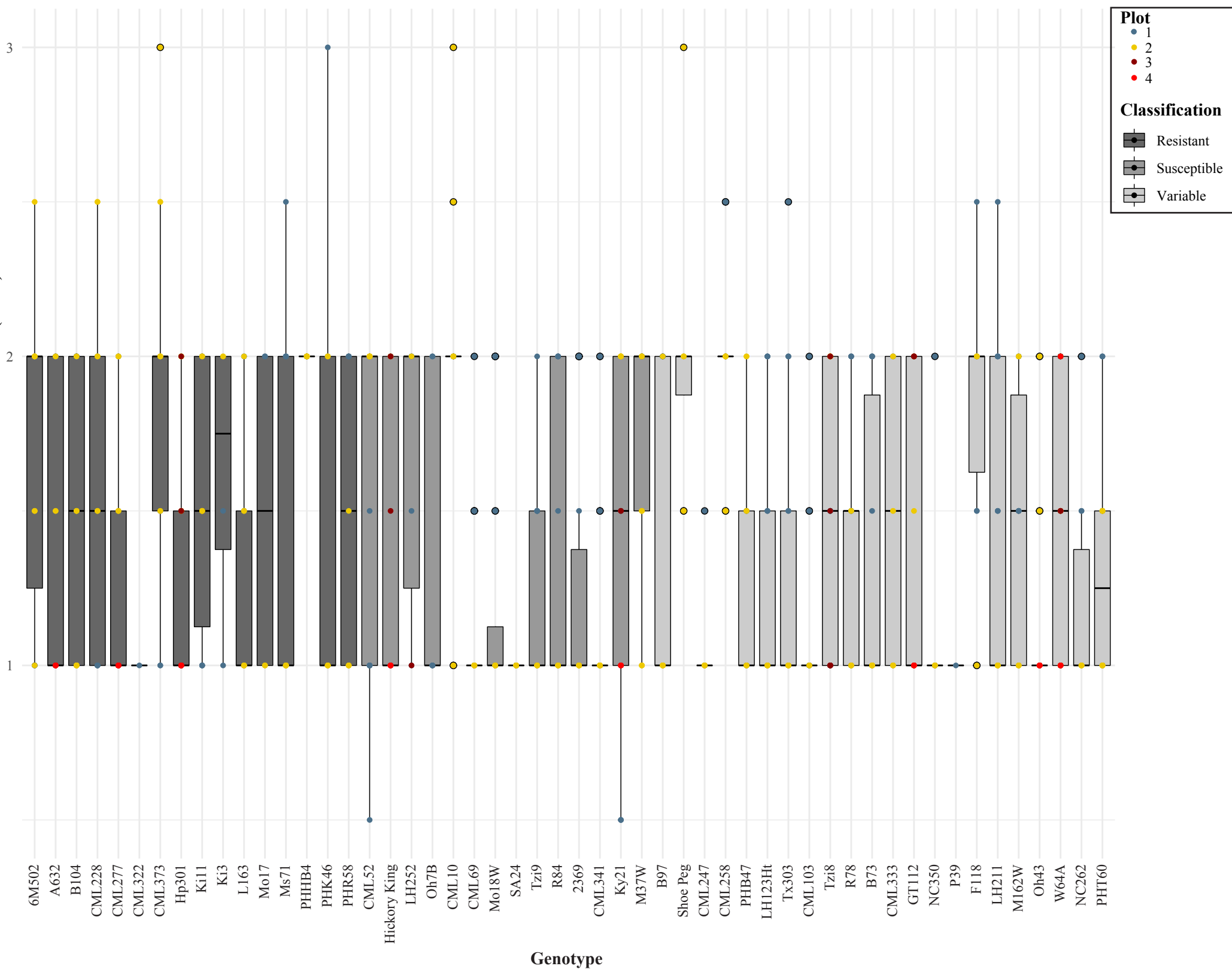


I.



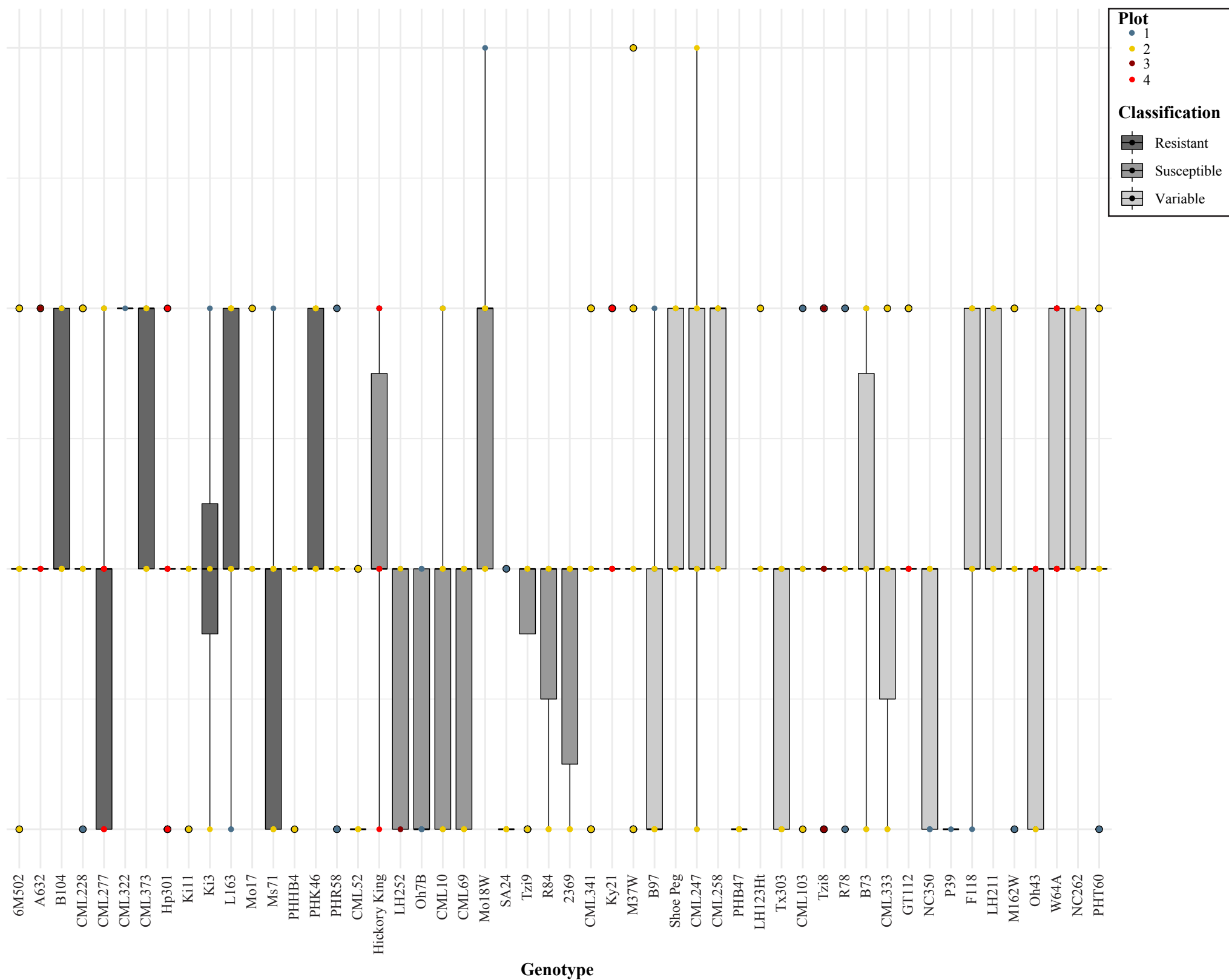
J.

Number of whorls in the soil – GUI (count)



K.

Number of whorls in the soil – Manual (count)



L.

Plant height (cm)

Genotype

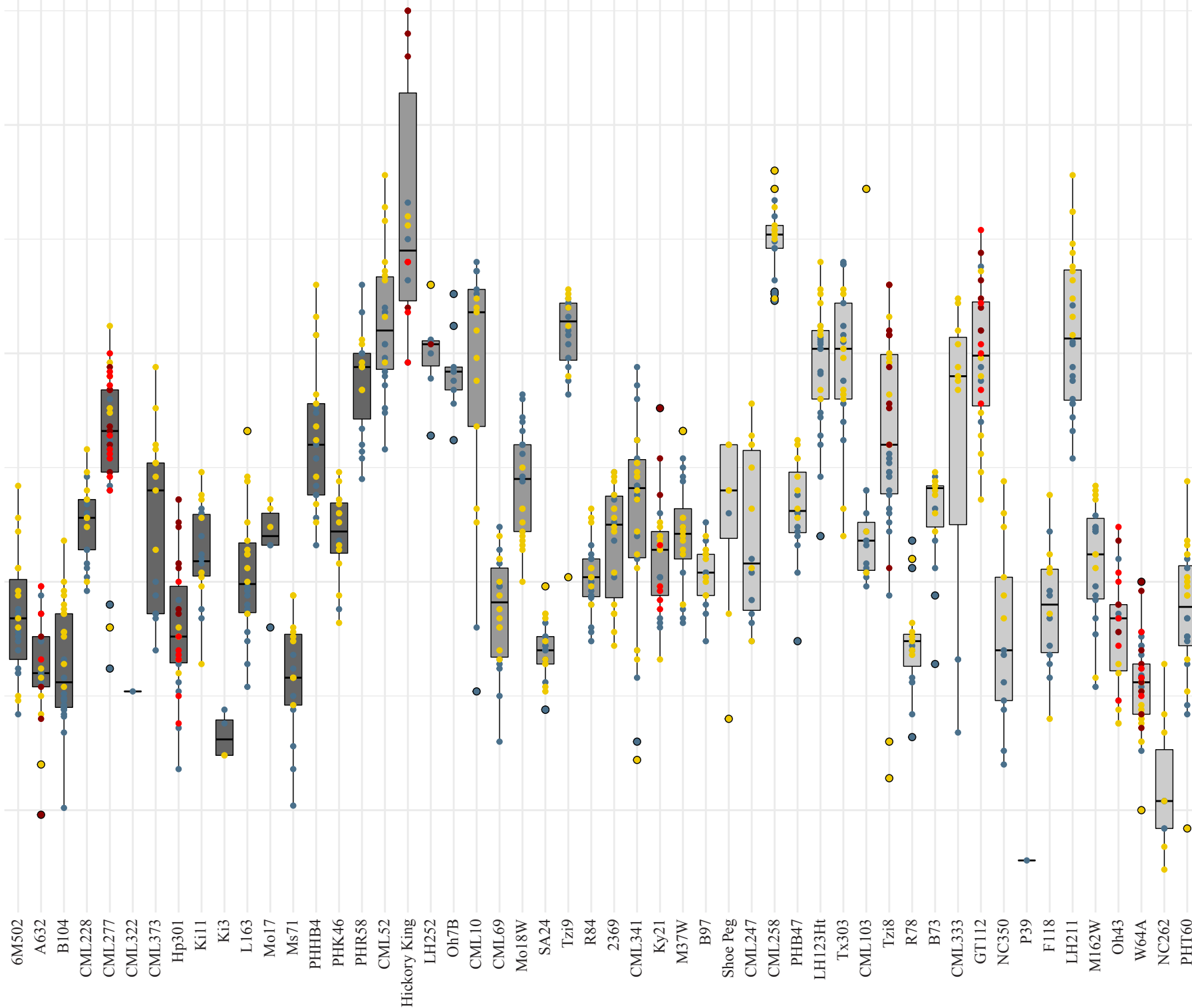
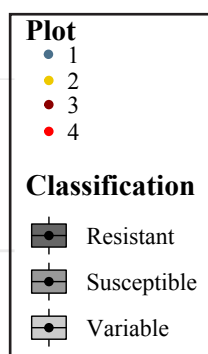


Figure S7. Plant phenotypes vary among genotypes. The following phenotypes vary among genotypes: (A) The number of roots on whorl 1 (the whorl closest to the soil, bottom whorl), (B) the number of roots on whorl 2 (middle whorl), (C) the number of roots on whorl 3 (top whorl), (D) the single root width, (E) the stalk width, (F) the root height on stalk, (G) the stalk-to-root grounding, (H) the root angle, (I) the spread width, (J) the number of whorls in the soil, quantified from the GUI, (K) the number of whorls in the soil, quantified manually, and (L) plant height. (A-L) Genotypes are ordered by root lodging classification as in Figure 2. The shade of each boxplot highlights whether the genotype was identified as root lodging resistant, root lodging susceptible, or variable. The color of each dot illustrates the replicate plot where the phenotype data is from. Outliers are outlined in black.

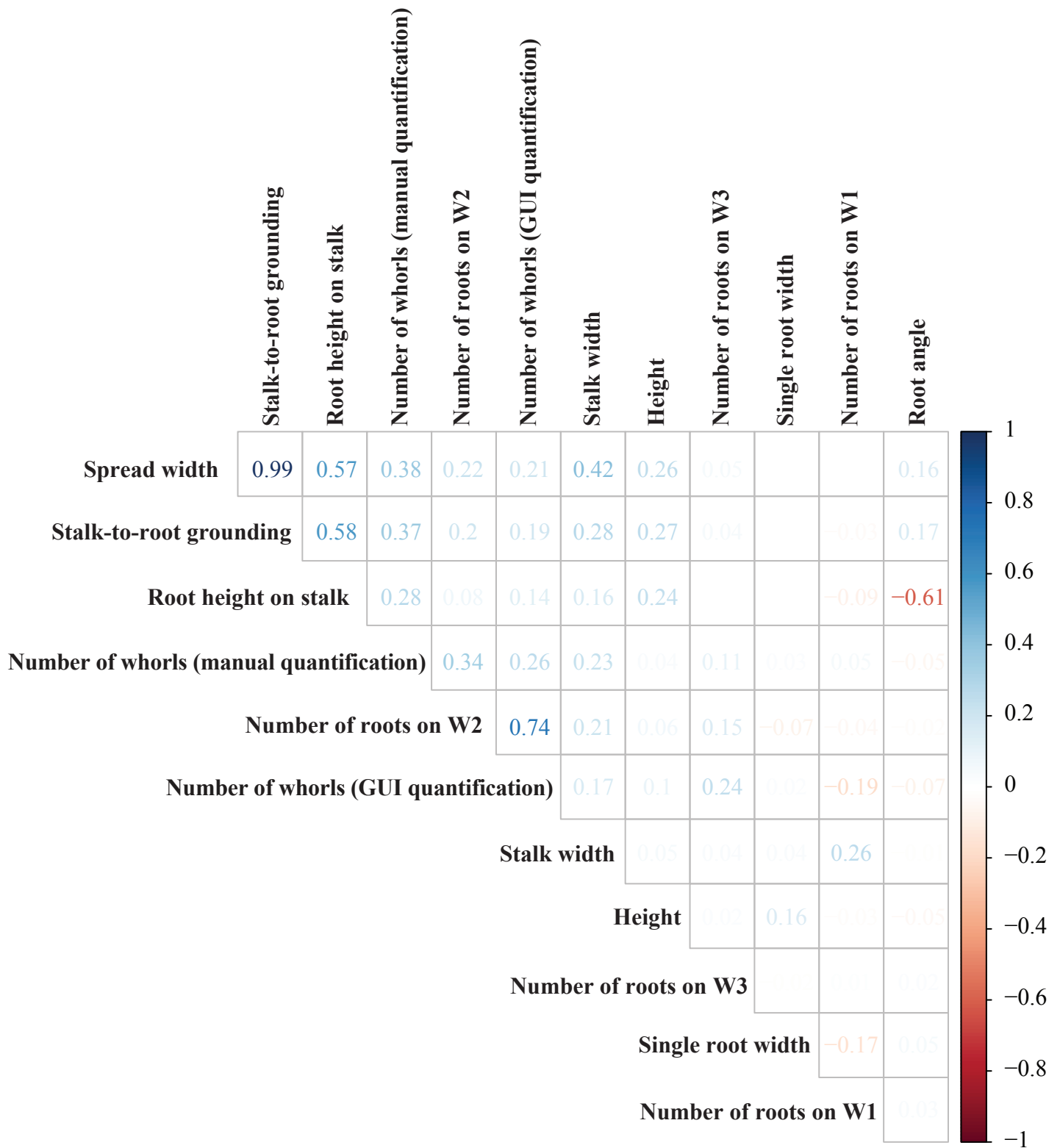


Figure S8. Brace root phenotypes are correlated with each other. A Pearson correlation analysis shows which phenotypes are highly correlated.

Predictor	EI		Ratio=None/All		Lodging susceptibility				Lodging severity				
	Flexural stiffness (EI)						13.365	32.178			16.978	27.592	
	Ratio = None/All						10.161	25.019			23.658	22.064	
	Height	42.194		33.742		65.84	31.006		65.74		41.74		
	Stalk width	41.624	48.468	17.22	19.389	42.108	52.141	10.461	29.143	40.588	50.548	13.3	24.438
	Single root width	19.922	21.836	12.202	16.972	34.11	36.311	13.561	18.056	43.418	54.847	15.048	29.187
	Number of roots on W1	20.931	23.721	24.449	28.072	26.024	32.835	8.988	18.081	27.643	30.748	10.629	18.657
	Root height on stalk	6.825	10.193	14.13	14.858	30.202	39.228	8.171	20.561	41.161	49.888	17.036	30.511
	Root angle	6.363	7.936	8.901	12.318	30.113	39.262	7.727	20.005	33.149	39.282	8.825	17.273
	Stalk-to-root grounding	7.702	9.741	26.072	24.845	33.218	33.225	7.867	17.61	30.748	33.032	8.069	16.399
	Spread width	7.352	10.213	14.063	11.265	25.029	31.238	6.751	17.358	24.809	33.334	5.617	13.217
	Number of roots on W2	8.561	14.473	7.118	12.954	22.665	21.165	6.471	14.192	17.683	19.222	5.407	13.352
	Number of whorls (manual)	4.065	4.879	9.105	8.086	16.931	20.945	10.511	13.94	19.246	17.583	13.265	13.509
	Number of whorls (GUI)	1.233	1.255	8.007	7.306	7.779	9.271	2.42	4.957	7.414	9.321	1.627	3.424
	Number of roots on W3	0	0.198	0.093	0.134	0.308	0.253	0.035	0.212	1.638	1.26	1.178	0.589
	82.5%	80.4%	78.4%	76.3%	68.0%	55.7%	68.0%	66.0%	55.7%	52.6%	63.9%	66.0%	
	EI		Ratio=None/All		Lodging susceptibility				Lodging severity				
	Outcome												

Literature Cited

- Flint-Garcia, S. A., A.-C. Thuillet, J. Yu, G. Pressoir, S. M. Romero, S. E. Mitchell, J. Doebley, et al. 2005. Maize association population: a high-resolution platform for quantitative trait locus dissection. *The Plant Journal* 44: 1054–1064.
- Liu, K., M. Goodman, S. Muse, J. S. Smith, E. Buckler, and J. Doebley. 2003. Genetic Structure and Diversity Among Maize Inbred Lines as Inferred From DNA Microsatellites. *Genetics* 165: 2117–2128.