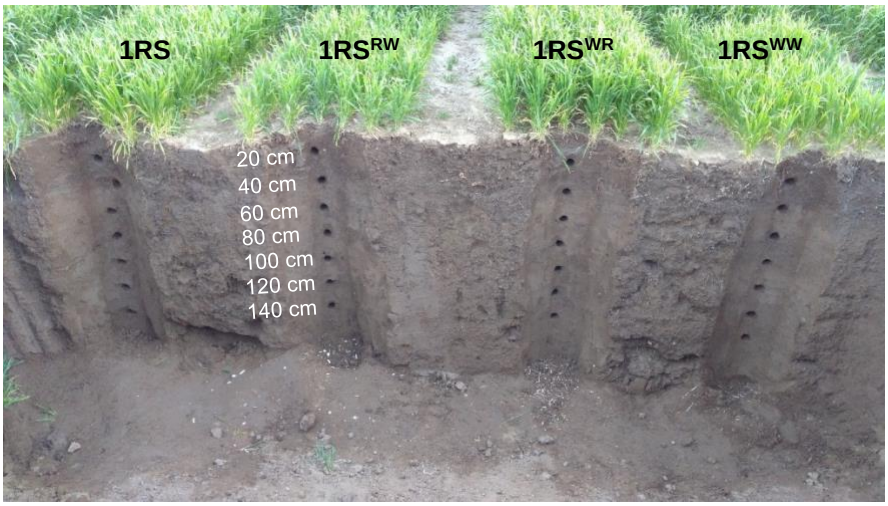
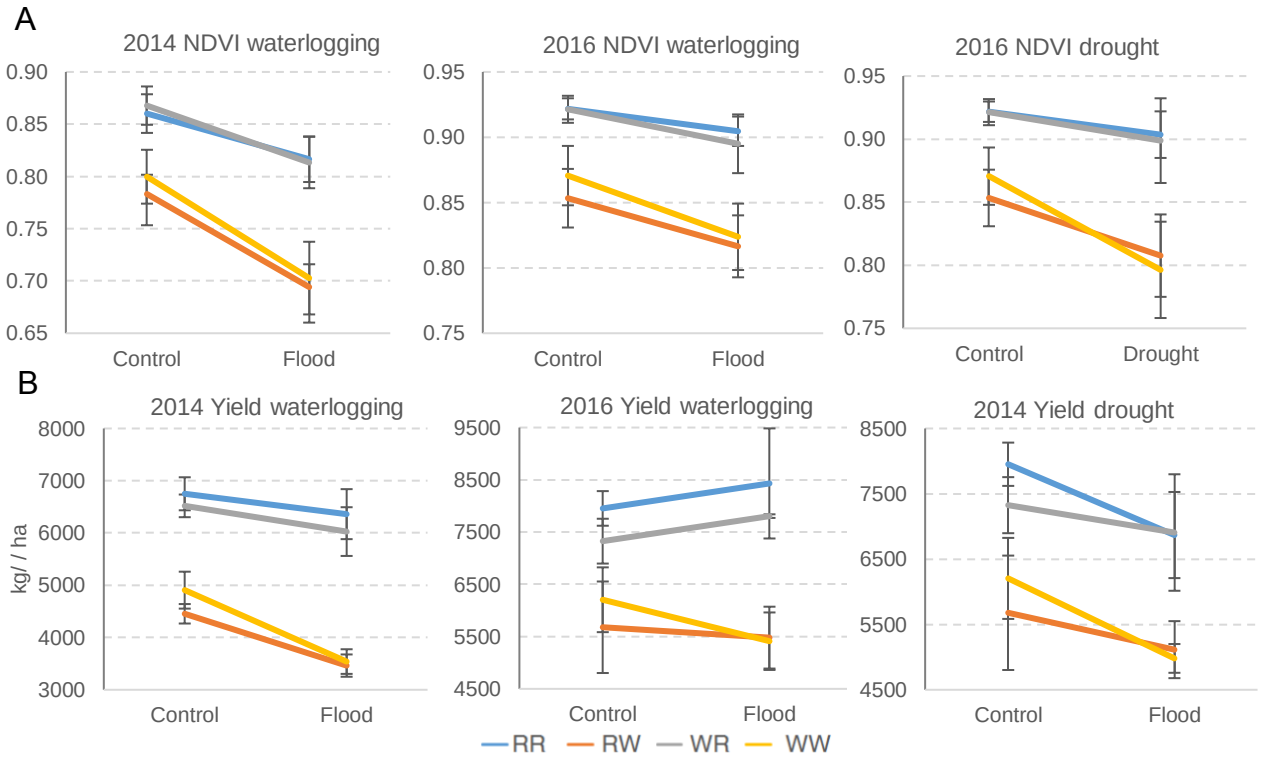
SUPPLEMENTARY FILE

Supplementary Figures

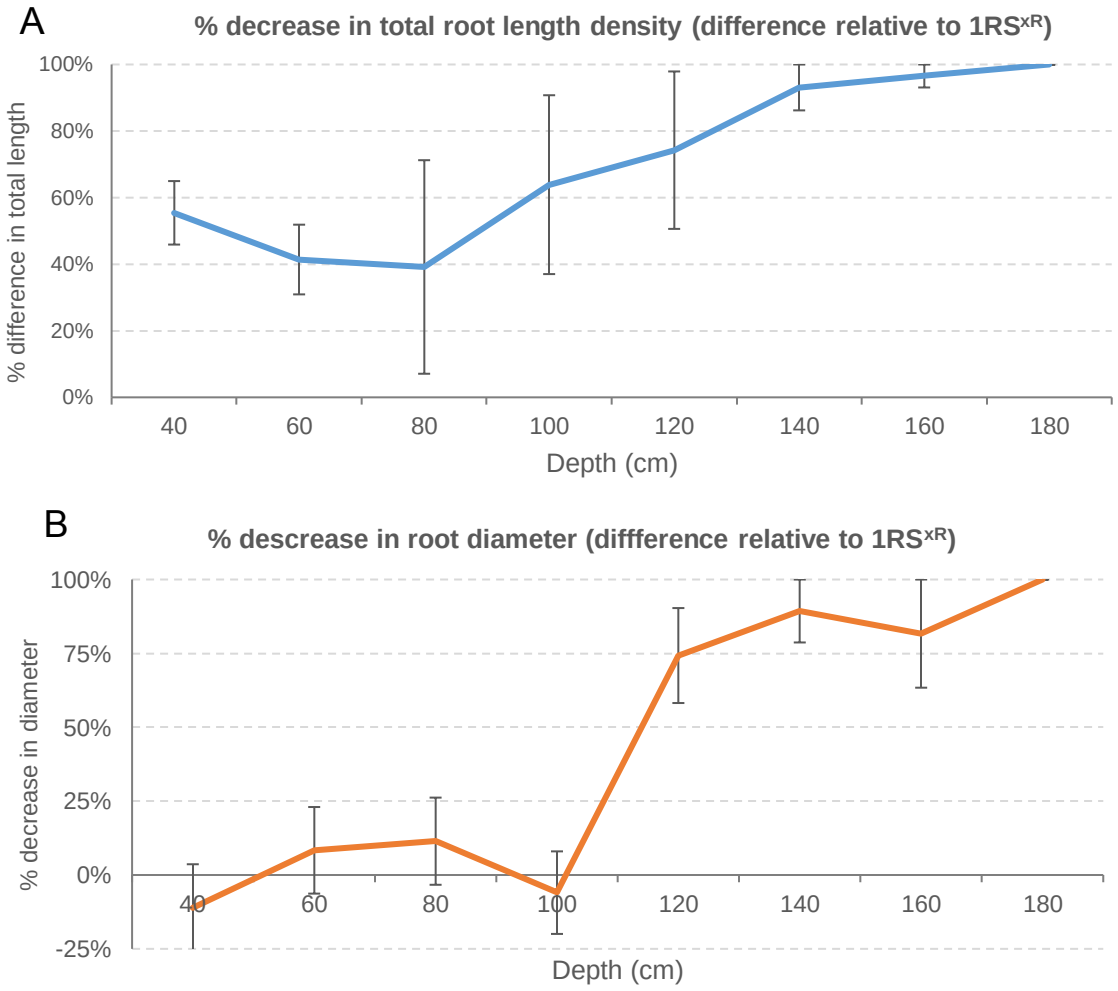
Supplementary Fig. S1. Excavation of the first block including the four genotypes (1RS, $1RS^{RW}$, $1RS^{WR}$, $1RS^{WW}$). The holes below the center of each plot (20 to 140 cm deep) indicate were the horizontal soil core samples were taken. Samples from 160 cm and 180 cm were taken from the other two blocks after we discovered the presence of roots at 140 cm in block 1. Plants were at the tillering stage at the time of the root sampling.



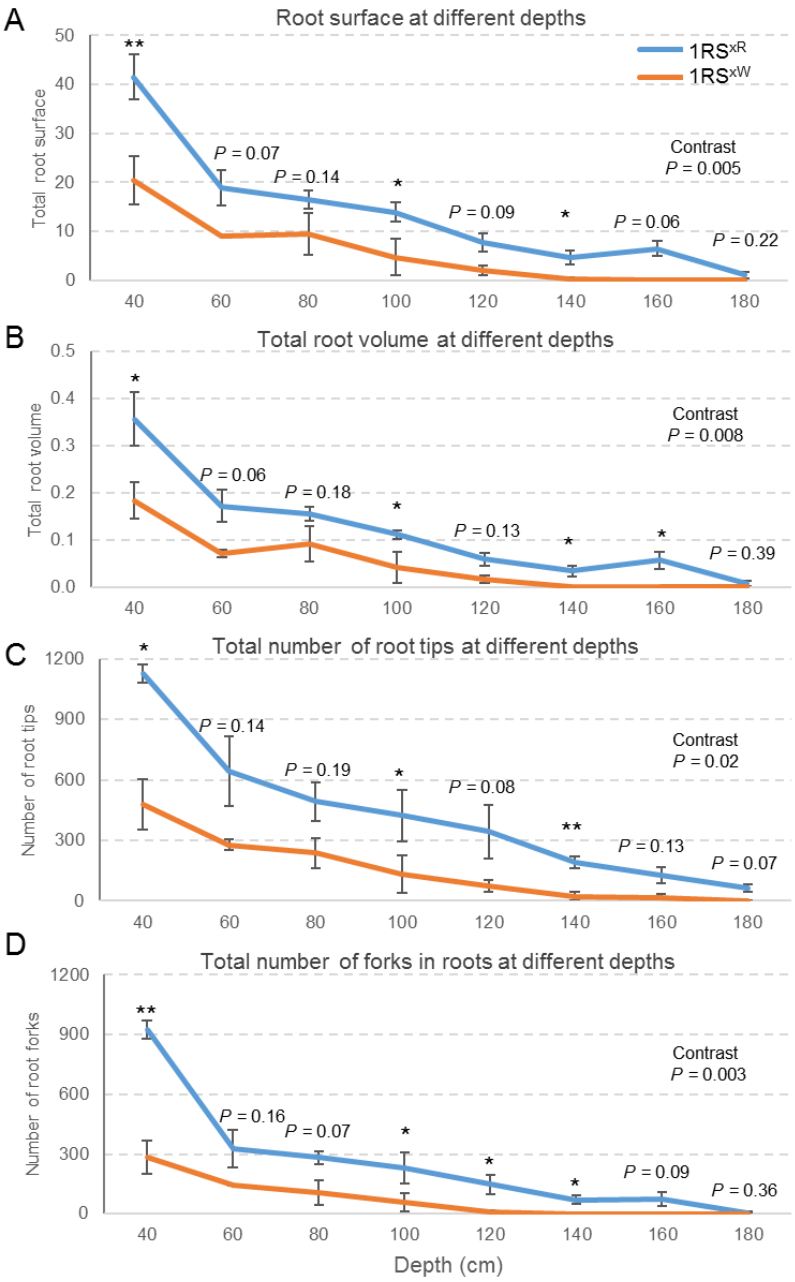
Supplementary Fig. S2. Plant biomass and grain yield. Plant biomass estimated by NDVI (A) and grain yield (B) under normal irrigation (control), waterlogging and drought conditions. Plants with the distal rye segment include the original Hahn-1RS lines (RR) and the line with the proximal wheat segment (WR =1RS^{WR}). Plants with the distal wheat segment include the line with both wheat segments (WW= 1RS^{WW}) and with only the distal wheat segment (RW =1RS^{RW}). Note the similar response of the lines with a similar distal chromosome segment. Differences between genotypes were generally larger under water stress conditions.



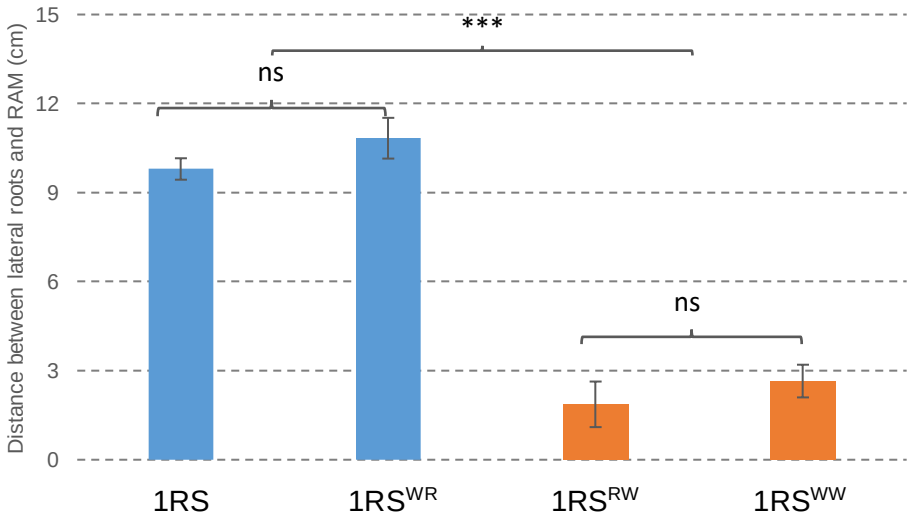
Supplementary Fig. S3. Percent difference in total root length density and diameter. Differences between genotypes expressed as a percent of the $1RS^{xR}$ values (($(1RS^{xR} - 1RS^{xW}) / 1RS^{xR}) *100$). **A**) Percent difference in total root length. **B**) Percent difference in average root diameter. Error bars represent standard errors of the means across blocks.



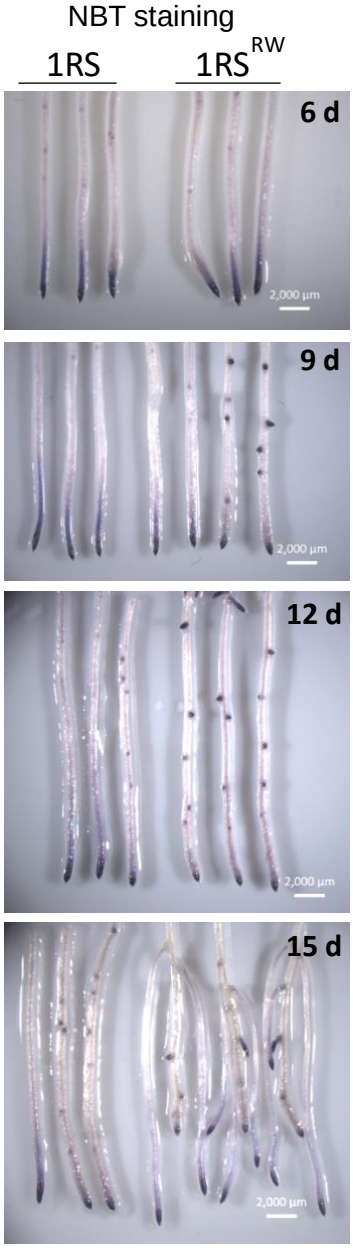
Supplementary Fig. S4. Differences between 1RS^{xR} and 1RS^{xW} in different root parameters with depth. The experiment is organized in a split plot design with 4 genotypes as main plots and depths as subplots. **A**) Root surface (SQRT (x+0.5)). **B**) Root volume ((x+1)$^{-4}$). **C**) Number of root tips (SQRT (x+0.5)). **D**) Number of root forks (SQRT (x+0.5)). *P* values were calculated using contrasts between 1RS^{xR} and 1RS^{xW} lines in individual ANOVAs by depth. Error bars represent standard errors of the means across blocks. *P* values are from transformed data (to meet the assumptions of the ANOVA) and means and SE are untransformed.



Supplementary Fig. S5. Comparison of the distances between the primary root apex and the first lateral root in lines carrying the distal rye (1RS and $1RS^{WR}$) versus the ones carrying the distal wheat segment ($1RS^{WW}$ and $1RS^{RW}$) in plants grown in hydroponics conditions for 22 days after germination under 2 mM nitrate. Lateral roots were significantly closer to the primary root apical meristem in lines with the distal wheat segment than in the lines with the distal rye segment (P = 2.5 E^{-7}, total n=12). No significant differences were detected between the lines. differing in the proximal wheat and rye segments.



Supplementary Fig. S6. Distribution of nitro blue tetrazolium (NBT) staining during root development in plants grown under low nitrate conditions (0.2 mM nitrate). No differences were detected between 1RS and 1RS^{RW} lines six days after germination (DAG). At 9 DAG, stained primordia of the lateral roots close to the root apical meristem (RAM) are evident in 1RS^{RW} but not in the 1RS. By 15 DAG, lateral roots are formed close to the RAM in 1RS^{RW} but no lateral root initiation is observed in the 1RS line close to the RAM (experiment performed at Davis).



Supplementary Tables

Supplementary Table S1a. NDVI waterlogging 2013-2014. Split plot RCBD repeated in time ANOVA table for the Normalized Difference Vegetation Index (NDVI). Main plots are waterlogging and normal irrigation, subplots are the genotypes $1RS^{xR}$ (averaged 1RS and $1RS^{WR}$) and $1RS^{xW}$ (averaged $1RS^{RW}$ and $1RS^{WW}$), and sub-sub plots are days. Since there is only one degree of freedom for days, we did not calculated conservative degrees of freedom.

Source	dF	Type III SS	Mean Sq.	F Value	Pr > F
Main plot					
Treat	1	0.04034	0.04034	90.30	0.0025
Block	3	0.00447	0.00149	3.34	0.1743
Error: Block*Treat	3	0.00134	0.00045		
Genot.	1	0.07140	0.07140	407.17	<.0001
Treat*Genot	1	0.00386	0.00386	22.02	0.0034
Error: Block*Treat*Genot.	6	0.00105	0.00018		
day	1	0.13770	0.13770	1130.10	<.0001
day*Treat	1	0.00100	0.00100	8.25	0.0232
day*Genot	1	0.00618	0.00618	50.75	<.0001
day*Treat*Genot	1	0.00007	0.00007	0.60	0.4518
Error	12	0.00146	0.00012		

R-square= 0.99456

Supplementary Table S2. NDVI waterlogging 2013-2014 by day. Because the day * genotype was significant we also analyzed the individual split plot ANOVAs by individual days. The significance of treatment and block effects was tested using the Block*Treatment as error (df = 3) and the significance of Genotype and Treatment * Genotype using the residual error (df = 6)

Source	df	April 17 *P*	April 30 *P*
Treat	1	<.0001	0.0083
Block	3	0.0126	0.3453
Genotype	1	<.0001	<.0001
Treat*Genot	1	0.0032	0.0097

Supplementary Table S3. Yield waterlogging 2013-2014. Split plot RCBD ANOVA table for grain yield. 2013-2014 field experiments at Davis, CA. Main plots are waterlogging and normal irrigation and subplots are the genotypes $1RS^{xR}$ (averaged 1RS and $1RS^{WR}$) and $1RS^{xW}$ (averaged $1RS^{RW}$ and $1RS^{WW}$).

Source	dF	Type III SS	Mean Sq.	F Value	Pr > F
Treat	1	2623671	2623671	3.56	0.1556
Block	3	510586	170195	0.23	0.8701
Block*Treat	3	2210786	736929		
Genot	1	21593447	21593447	138.98	<.0001
Treat*Genot	1	545493	545493	3.51	0.1101
Error	6	932244	155374		

R-square= 0.967193

Supplementary Table S4. NDVI waterlogging 2015-2016. Split plot RCBD repeated in time ANOVA table for the Normalized Difference Vegetation Index (NDVI). Main plots are waterlogging and normal irrigation, subplots are genotypes with $1RS^{xR}$ (averaged 1RS and $1RS^{WR}$) and $1RS^{xW}$ (averaged $1RS^{RW}$ and $1RS^{WW}$), and sub-sub plots are the four days. Since it is not possible to randomize days, *P* values were calculated using conservative degrees of freedom (indicated in blue, and calculated by dividing by df days= 3).

Source	**df**	**Type III SS**	**Mean Sq.**	**F Value**	**Pr > F**	
Main plot						
Treat	1	0.01214	0.01214	960.33	0.0010	
Block	2	0.00307	0.00154	121.57	0.0082	
Error: Block*Treat	2	0.00002	0.00001			
Genot	1	0.05815	0.05815	266.34	<.0001	
Treat*Genot	1	0.00121	0.00121	5.53	0.0783	
Error: Block*treat*Genot.	4	0.00087	0.00022			
	df	Con.df				*P* Con.df
day	3	1	0.14303	0.14303	894.6	<.0001	<.0001
day*Treat	3	1	0.00161	0.00161	10.1	0.0002	0.0130
day*Genot	3	1	0.02108	0.02108	131.5	<.0001	<.0001
day*Treat*Genot	3	1	0.00059	0.00059	3.7	0.0253	0.0906
Error	24	8	0.00128	0.00005			

R-square= 0.9947

Supplementary Table S5. NDVI waterlogging 2015-2016 by day. Because the day * genotype was significant we also analyzed the individual split plot ANOVAs by individual days. The significance of treatment and block effects was calculated using the Block*Treatment as error (df = 2) and the significance of Genotype and Treatment * Genotype using the residual error (df = 4). The four NDVI measurement were taken before the breakage of the irrigation pipeline, so are not affected by this accident.

Source	df	March 24	April 6	April 13	April 28
		P	*P*	*P*	*P*
Treat	1	0.0110	0.0057	0.0133	0.0146
Block	2	0.0383	0.0291	0.2118	0.0972
Genotype	1	0.0080	0.0008	0.0009	<.0001
Treat*Genot	1	0.9137	0.0595	0.0687	0.1587

Supplementary Table S6. Yield waterlogging 2015-2016. Split plot RCBD ANOVA table for grain yield. Main plots are waterlogging and normal irrigation, whereas subplots are genotypes with $1RS^{xR}$ (averaged 1RS and $1RS^{WR}$) or $1RS^{xW}$ (averaged $1RS^{RW}$ and $1RS^{WW}$). The broken irrigation pipeline increased variability and contributed to the absence of treatment differences.

Source	df	Type III SS	Mean Sq.	F Value	Pr > F
Treat	1	218	218	0.00	0.9772
Block	2	4048744	2024372	9.58	0.945
Block*Treat	2	422468	211234	0.44	0.6702
Genot	1	14317650	14317650	30.03	0.0054
Treat*Genot	1	711945	711945	1.49	0.2888
Error	4	1906973	476743		

R-square= 0.91092

Supplementary Table S7. NDVI drought 2015-2016. Split plot RCBD repeated in time ANOVA table for the Normalized Difference Vegetation Index (NDVI). Main plots are terminal drought and normal irrigation, subplots are the genotypes $1RS^{xR}$ (averaged 1RS and $1RS^{WR}$) and $1RS^{xW}$ (averaged $1RS^{RW}$ and $1RS^{WW}$), and sub-sub plots are the four days. Since it is not possible to randomize days, P values were calculated using conservative degrees of freedom (in blue, calculated by dividing by df days= 3).

Source	df	Type III SS	Mean Sq.	F Value	Pr > F	
Main plot						
Treat	1	0.019388	0.019388	7.38	0.1130	
Block	2	0.000617	0.000309	0.12	0.8949	
Error: Block*Treat	2	0.005254	0.002627			
Genot.	1	0.075617	0.075617	171.84	0.0002	
Treat*Genot	1	0.004705	0.004705	10.69	0.0308	
Error: Block*treat*Genot.	4	0.001760	0.000440			
	df	Con.df				*P* Con.df
day	3	1	0.231431	0.077144	434.07	<.0001	<.0001
day*Treat	3	1	0.017311	0.005770	32.47	<.0001	0.0005
day*Genot	3	1	0.025883	0.008628	48.55	<.0001	0.0001
day*Treat*Genot	3	1	0.000529	0.000176	0.99	0.4134	0.35
Error	24	8	0.004265	0.000178			

R-square= 0.98897

Supplementary Table S8. NDVI drought 2015-2016 by day. Because the day * genotype was significant we also analyzed the individual split plot ANOVAs by individual days. The significance of treatment and block effects was calculated using the Block*Treatment as error (df = 2) and the significance of Genotype and Treatment * Genotype using the residual error (df = 4)

Source	df	March 24 *P*	April 6 *P*	April 13 *P*	April 28 *P*
Treat	1	0.2372	0.3538	0.2148	0.0562
Block	2	0.8203	0.7723	0.8138	0.7577
Genotype	1	0.0128	0.0049	0.0011	<.0001
Treat*Genot	1	0.0984	0.1885	0.1559	0.0005

Supplementary Table S9. Yield drought 2015-2016. Split plot RCBD ANOVA table for grain yield. Main plots are waterlogging and normal irrigation and subplots are the genotypes $1RS^{xR}$ (average 1RS and $1RS^{WR}$) and $1RS^{xW}$ (average $1RS^{RW}$ and $1RS^{WW}$).

Source	df	Type III SS	Mean Sq.	F Value	Pr > F
Treat	1	2030506	2030506	0.83	0.4587
Block	2	961895	480948	0.20	0.8359
Block*treat	2	4900514	2450257		
Genot	1	9389144	9389144	22.81	0.0088
Treat*Genot	1	15394	15394	0.04	0.8561
Error	4	1646190	411547		

R-square= 0.9131

Supplementary Table S10. Total root length density. Split plot – RCBD ANOVA for the excavation experiment including genotypes as main plots (averages of 1RS and $1RS^{WR}$ and $1RS^{RW}$ and $1RS^{WW}$) and depths as subplots. Conservative degrees of freedom (in blue) were used to test the significance of Depth and Genotype * Depth because depths cannot be randomized. This approach is very conservative and the correct *P* value is between the two estimates. This does not affect the significance of the differences between genotypes, which is the main objective of this analysis.

Source	**df**		**Type III SS**	**Mean Sq.**	**F Value**	**Pr > F**	
Main plot							
Genot	1		64176	64176	36.58	0.0263	
Block	2		13794	6897	3.93	0.2028	
Main error Block*Genot	2		3509	1754			
Subplots	df	Cons.df					*P* Cons.df
Depth	7	1	327458	46780	25.07	<.0001	0.007
Genot*Depth	7	1	31705	4529	2.43	0.0497	0.190
Subplot Error	24	3.4	44781	1866			

R-square= 0.806

Supplementary Table S11. Total root length density contrast. The previous analysis (Table S10) has limited degrees of freedom for genotype (ν_1= 1ν_2= 2), so we performed an alternative ANOVA including the four genotypes and a statistical contrast between the lines with distal rye and wheat segments (ν_1= 1,ν_2= 6).

Source	df	Type III SS	Mean Sq.	F Value	Pr > F
Contrast distal R *vs.* W	1	504.23	504.23	19.64	**0.004**
Error Block*Gen	6	154.07	25.68		

[1] Data transformed (SQRT(length +0.05) to optimize homogeneity of variances and normality of residuals.

Supplementary Table S12. Total root length density contrast by depth. Alternative ANOVA including the four genotypes and a statistical contrast between the lines with distal rye and wheat segments by depth.

Source	40 cm	60 cm	80 cm	100 cm	120 cm	140 cm	160 cm	180 cm
Contrasts distal R *vs.* W	0.0020	0.1279	0.1313	0.0347	0.0844	0.0180	0.0678	0.1687

[1] Data transformed (SQRT(length +0.05) to optimize homogeneity of variances and normality of residuals.

Supplementary Table S13. Average root diameter. Split plot RCBD ANOVA table for the excavation experiment using genotypes as main plots (averages of 1RS and 1RS^{WR} and 1RS^{RW} and 1RS^{WW}) and depths as subplots. Conservative degrees of freedom (in blue) were used to test the significance of Depth and Genotype * Depth. This is a very conservative approach and the correct *P* value is between the two estimates. This does not affect the significance of the differences between genotypes, which is the main objective of this analysis.

Source	df		Type III SS	Mean Sq.	F Value	Pr > F	
Main plot							
Genot	1		0.0083442	0.0041721	13.92	0.0649	
Block	2		0.1096073	0.1096072	0.53	0.6536	
Main error Block*Genot	2		0.0157427	0.0078713			
Subplots	**df**	**cons.df**				***p***	**cons.df**
Depth	7	**1**	0.3666487	0.0523784	19.45	<0.0001	**0.02**
Gen*Depth	7	**1**	0.1232096	0.0176014	6.54	0.0002	**0.08**
Subplot Error	24	3.4	0.0646355	0.0026931			

R-square= 0.68817

Supplementary Table S14. Average root diameter contrast. Alternative split plot ANOVA including the four genotypes and a statistical contrast between the lines with distal rye and wheat segments (ν_1= 1ν_2= 6). More sensitive than Table S13.

Source	df	Type III SS	Mean Sq.	F Value	Pr > F
Contrast distal R *vs.* W	1	0.21715	0.21715	16.70	**0.006**
Main error Block*Genot	6	0.07800	0.01300		

Supplementary Table 15. Average root diameter contrast by depth. Alternative ANOVA including the four genotypes and a statistical contrast between the lines with distal rye and wheat segments by depth.

Source	40 cm	60 cm	80 cm	100 cm	120 cm	140 cm	160 cm	180 cm
Contrasts distal R *vs.* W	0.3596	0.7262	0.5726	0.8369	0.0735	0.0012	0.0256	0.0287

Supplementary Table 16. Repeated measures for root length (mm). ANOVA between 1RS and 1RS^{RW} of the second longest main root measured daily between 6 and 16 d. Within each experiment, data was analyzed as a split plot with genotype as main plot and days as subplot (using conservative df, indicated in blue). The three experiments were combined in a single analysis using experiment as block and reps nested in experiment (Fig. 4A).

Source	df	Type III SS	Mean Sq.	F Value	Pr > F
Exp	2	143301.9	71650.9	22.35	<.0001
genot	1	97054.5	97054.5	30.27	<.0001
genot*rep(Exp)	47	150674.8	3205.8		

	df	Cons.df				*P* cons.df
day	10	1	1688462	168846	1225.49	<.0001 <.0001
genot*day	10	1	101971	10197	74.01	<.0001 <.0001
Error	490	49	67512	137.8		

R-square= 0.96986

Supplementary Table S17. Root length (mm) by day. ANOVAs for each time point combining the three experiments (used as blocks). Untransformed LS means (n=10) obtained from ANOVAs by day.

	6	7	8	9	10	11	12	13	14	15	16
1RS	90.9	104.8	122.2	139.5	160.2	183.5	206.3	231.9	253.0	274.3	299.6
1RSRW	88.6	102.8	118.8	133.3	150.9	169.3	184.9	197.9	204.9	210.6	214.6
ANOVA *P*	0.446	0.581	0.420	0.173	0.053	0.006	0.0002	<.0001	<.0001	<.0001	<.0001

Supplementary Table S18. Repeated measures for root elongation rate (mm/ h^{-1}). ANOVA between 1RS and $1RS^{RW}$ for the second longest main root measured daily between 6 and 16 d. Within each experiment, data was analyzed as a split plot with genotype as main plot and days as subplot (using conservative df, indicated in blue). The three experiments were combined in a single analysis using experiment as block and replications nested within experiment (Fig. 4B).

Source		df	Type III SS	Mean Sq.	F Value	Pr > F	
Exp		2	3.3099	1.6550	8.95	0.0005	
genot		1	22.9752	22.9752	124.23	<.0001	
genot*rep(Exp)		47	8.6921	0.1849			
		Cons.df					*P* Cons.df
day	9	1	5.9958	0.6662	13.18	<.0001	0.0007
genot*day	9	1	13.5174	1.5019	29.72	<.0001	<.0001
Error	441	49	22.2881	0.0505			

R-square=0.7109

Data was transformed to the power of **1.7 to improve normality of residuals

Supplementary Table S19. Root elongation rate by day. ANOVAs for each time point combining the three experiments (used as blocks). Untransformed LS means (n=10) obtained from ANOVAs by day.

	6.5	7.5	8.5	9.5	10.5	11.5	12.5	13.5	14.5	15.5
1RS	0.5794	0.7116	0.7418	0.8622	0.9449	0.9441	1.0136	0.8946	0.9556	1.0128
$1RS^{RW}$	0.5997	0.6545	0.6245	0.7335	0.7487	0.6455	0.5141	0.2973	0.2602	0.1630
ANOVA *P*	0.7085	0.2923	0.0328	0.0021	0.0006	<.0001	<.0001	<.0001	<.0001	<.0001

P values from transformed data to the power of **1.7 to improve normality of residuals.