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Association Study of SNPs Markers to Traits Linked to Drought Stress Tolerance in Potato

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ABSTRACT: Potato (*Solanum tuberosum* L.) is the third most important food crop in the world and its production is constantly threatened by periods of drought. In this study, 115 potato genotypes were evaluated among 56 of the andigena group and 59 phureja to observe genetic variation in physiological traits that may be linked to drought tolerance. Eleven attributes were evaluated in genotypes tolerant and susceptible to drought stress. The genotypic variation of the materials was evaluated with a total of 968 SNP-type molecular markers, subjected to two soil moisture conditions. Association analysis was performed using the GWASpoly program to determine possible allelic interactions between genotypes with different ploidy levels. Analyses were corrected using population structure and parentage matrix as fixed cofactors. Significant SNPs were associated with phenotypic characteristics under contrasting water conditions for traits such as days to flowering, relative water content, tuber number and plant height. MYC-type transcription factors were associated with plant height, number of tubers per plant and plant water balance, demonstrating the multifunctionality of these regulatory proteins. While the HOS1 gene could be linked to the reduction of flowering time. These results will be the starting point for future studies for the validation of the markers, so that they can be used in potato drought stress breeding programs.

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KEYWORDS: Genetic polymorphisms, Phenotypic traits, GWASpoly analysis, Population structure. Allelic interactions and Genotypic variation

INTRODUCTION

After cereals, potato (*S. tuberosum* L.) is the most important crop worldwide. The most valued tuber, it is also the main candidate to address food security in a continuously growing population. It is projected that by 2050 we will need to feed more than 9.7 billion people, which will require a 70% increase over current food yields (Devaux et al., 2021). Such growth in food production must be sustainable. Therefore, it is necessary to generate more water-efficient cultivars that can mitigate the negative effects of climate change. In this regard, potato is an extremely sensitive species to water deficit stress, with a reduction in production of up to 91% under severe conditions (Gervais et al., 2021). Thus, the absence of cultivars adapted to dry season could cause yield losses of up to 32% by 2050 (Alvarez-Morezuelas et al., 2023). Public and private sector research centers are developing breeding strategies to make potato crop production less vulnerable to drought stress.

Drought tolerance is a complex attribute of a quantitative nature that hinders breeding progress. Yield is the trait that normally defines the tolerance levels of genotypes exposed to low soil moisture conditions. On the other hand, the most important physiological traits that determine tuber production are mainly associated with photosynthesis, stomatal conductance and leaf area (Aliche et al., 2020). In addition, earliness has been a useful strategy for crops to escape dry-seasons at the end of the growing steppes (Kebede et al., 2019). The molecular basis of water stress tolerance is quite complex, although molecular advances have revealed many of the underlying mechanisms.

The Genome Wide Association Study (GWAS) is a powerful tool to reveal the molecular mechanisms of complex traits. The association of phenotypic and genotypic characteristics evidences genetic effects as well as interactions between alleles involved in the traits of interest. The possibility of selecting, through markers, genes and gene families simultaneously accelerates breeding processes for contributions from multiple *loci*. In addition, associative mapping techniques have contributed to breeding in plants of a tetraploid nature, where polygenic traits and their genetic effects become more complex. Recently, regions of the potato crop genome associated with tuber quality (Pandey et al., 2022), efficient use of nitrogen (Ospina et al., 2021), common scab resistance

(Koizumi et al., 2021), resistance to *P. infestans* attack (Mosquera et al., 2016) and drought tolerance (Alvarez-Morezuelas et al., 2023; D'hoop et al., 2014; Saidi & Hajibarat, 2020; Tagliotti et al., 2021; Tiwari et al., 2022). The sequencing of the potato genome (PGSC, 2011), reduction in sequencing costs and increase in number of reads has favored the identification of single nucleotide polymorphisms (SNPs). SNP-type marker platforms such as Infinium 8303 (Felcher et al., 2012) and DroughtDB databases (Alter et al., 2015) have boosted association studies of complex potato crop traits for drought resistance..

The identification of SNPs associated with drought resistance traits is extremely useful in potato breeding programs. In this study, we studied the relationship between molecular markers of SNPs and traits linked to potato tolerance to water deficit stress.

METHODOLOGY

Association analysis

Phenotypic information was obtained from eleven quantitative traits evaluated in 115 potato (*Solanum tuberosum*) genotypes under moisture deficit stress conditions, including days to flowering (DF), stem number (NT), plant height (AP), leaf water potential (Ψh) 2 am (PH2), leaf water potential (Ψh) 10 am (PH10), total chlorophyll content (CCI), relative water content (CRA), foliar dry matter accumulation (MSH), tuber dry matter accumulation (MST), number of tubers per plant (NTP) and yield per plant (RTO). Genotypic data were obtained by massive sequencing of libraries constructed from 87 candidate genes identified for water stress tolerance or resistance traits with relevant biological significance (Table S1).

With the sequencing data, population structure was determined through Bayesian modeling implemented in STRUCTURE software (Pritchard et al., 2000) without a priori population information using a tetraploid model (Andigena: 1 = AAAA, 2 = AAAB, 3 = AABB, 4 = ABBB, 5 = BBBB; Phureja: 1 = AA, 3 = AB, 5 = BB) and determined using the method of Evanno et al. (2005) in STRUCTURE Harvester python script version 0.6.8 (Earl & von Holdt, 2012). These results were analyzed using the CLUMPP program (Jakobsson & Rosenberg, 2007) that minimizes variance in all interactions and corroborated with the methodology proposed by Puechmaille (2016) that determines four additional metrics (Mean LnP (K), Stdev LnP (K), Ln'(K), (|Ln"(K)|) implemented in StructureSelector (Li & Liu, 2018). The number of subpopulations was confirmed with a principal component analysis (PCoA) performed in the GeneAlEx version 6.5 package (Peakall and Smouse, 2006).

A total of 968 markers were used to assess the associations between marker and phenotypes using linear mixed models (MLM). The MLMs considered the results of the population structure and the kinship matrix as fixed effects. The analysis was performed using the library developed for association studies in autopolyploids GWASpoly (Rosyara et al., 2016) in the statistical program R v4.2.0 (www.r-project.org). The obtained sequences were filtered by allele frequency less than 0.05. Potato plant genotypes were characterized according to allele dosage, including nuliplex (0=AAAAAA), simplex (1=AAAB), duplex (2=AABB), triplex (3=ABBB) and quadriplex (4=BBBB), considering that the study materials were tetraploid (Rosyara et al., 2016). The general genetic model allows the fixed effect for each genotype class to be arbitrary. In this case, the model presented four degrees of freedom for the F-test (one less than the number of genotype classes). In addition to the general model, four different single-parameter genetic models were used: additive, single dominant, double dominant and additive diploidized. The additive model considers the effect of each SNP as proportional to the reference allele dose. The dominance models correspond to single dominant and double dominant. Basically, the single dominant model evaluates the hypothesis that the presence of one or more copies of a reference allele had similar performance among them and different performance of the homozygous genotype of the alternative allele. In this case, the three heterozygotes are equivalent to one of the homozygotes, thus two non-equivalent single dominant parameterizations for each marker. The double dominant model is specific for tetraploids and evaluates whether the duplex genotype performed similarly to the simplex and nulliplex, or to the triplex and quadriplex, depending on the dominant allele (A or B). To identify the significance of marker-phenotype associations, a threshold of 0.05 corrected by the Bonferroni method and by the permutation test was taken. Results were presented in Manhattan-charts and plots (Rosyara et al., 2016).

Gene annotation

The sequence of nucleotide positions of the two sides of significant SNPs were selected from the respective reference genome and compared with other species using the BLAST program (Altschul et al., 1990) to determine candidate genes. The homology and function of the genes were annotated using the NCBI databank (https://www.ncbi.nlm.nih.gov/).

RESULTS AND DISCUSSION

Population structure analysis discriminated two main populations (K = 2; Table 1 and Figure 1), which was corroborated by Neighbor cluster analysis (Figure 2a) using a dendogram obtained from an identity analysis where two groups were separated with a dissimilarity coefficient of 0.30, and principal component analysis (PCA) for which a linkage disequilibrium value was established with a DL=0.001 in which 45.5% of the variability was explained by the first two components (Figure 2b). The first component explains 34.3% of the total variance and separates the Phurejas group (red) from the Andigenas (blue). The first population, named

Andigena, contains 62 accessions (62.6% of the population). 37 genotypes of the population (37.4%) constituted the second population, named Phureja.

In contrast, Sanchez (2017), obtained seven clusters of the evaluated accessions, with similarity indices ranging from 0.452 to 0.841. While Deperi (2019), detected five clusters within the evaluated population of potatoes of 231 genotypes from the germplasm collection of the INTA Balcarce Breeding Program, with coefficients between 0.721 and 1, which evidences a structured population. And Tagliotti et al (2021), in their genetic diversity study with 183 genotypes which tended to form five clusters.

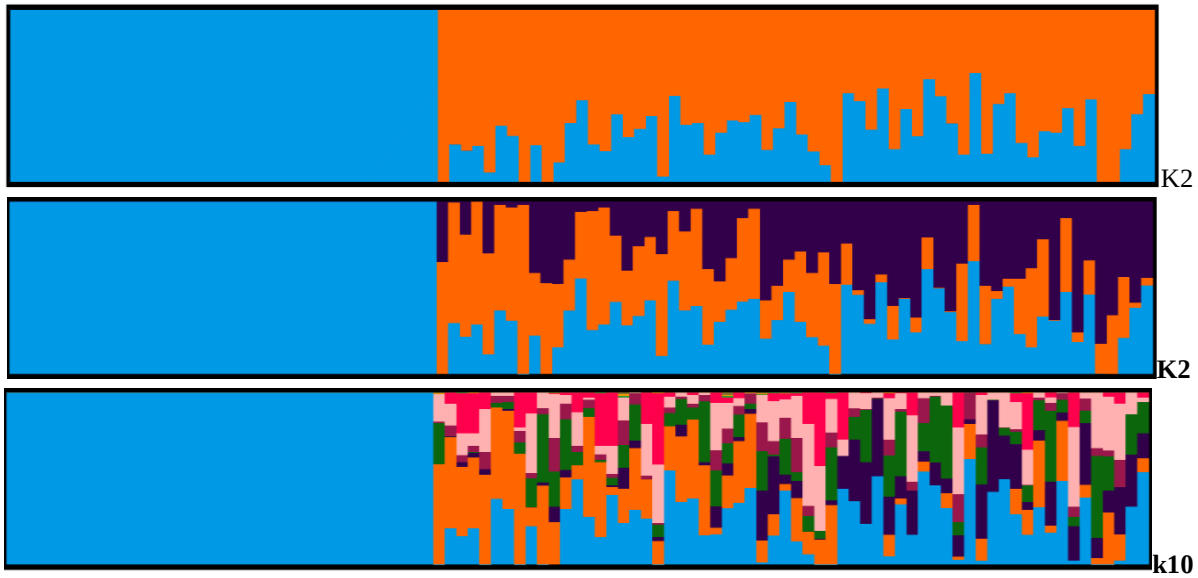
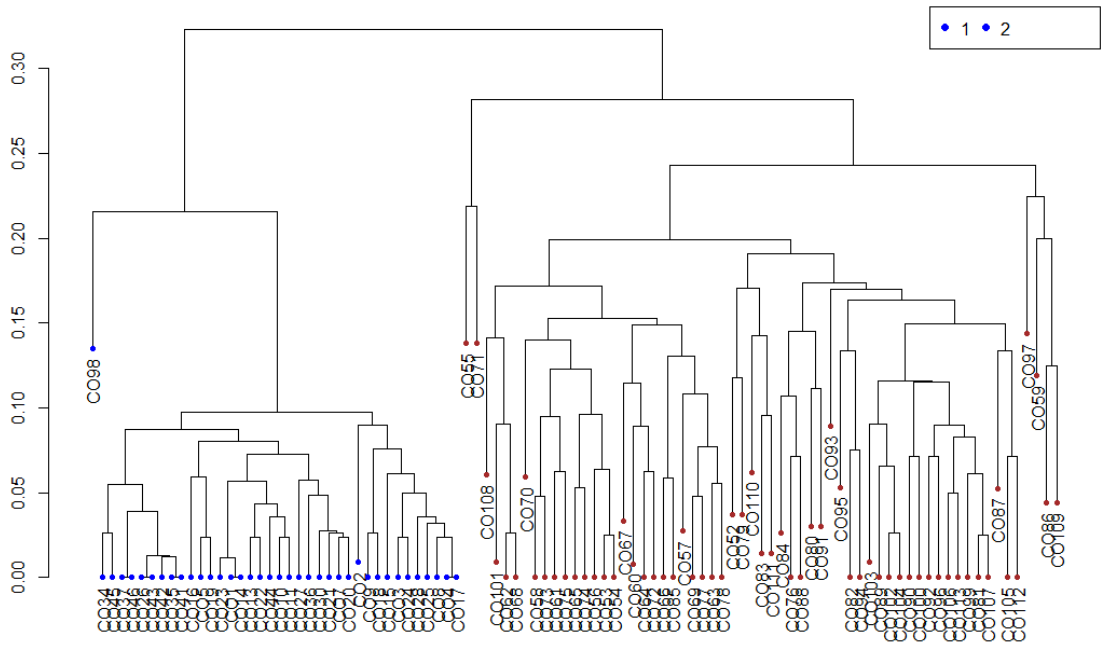


Figure 1. Graphical representation of the results of the genetic structure analysis of 2 potato populations *(S. tuberosum* Phureja group and *S. tuberosum* Andigena group) (K= 2 to k10) using 968 (SNPs).



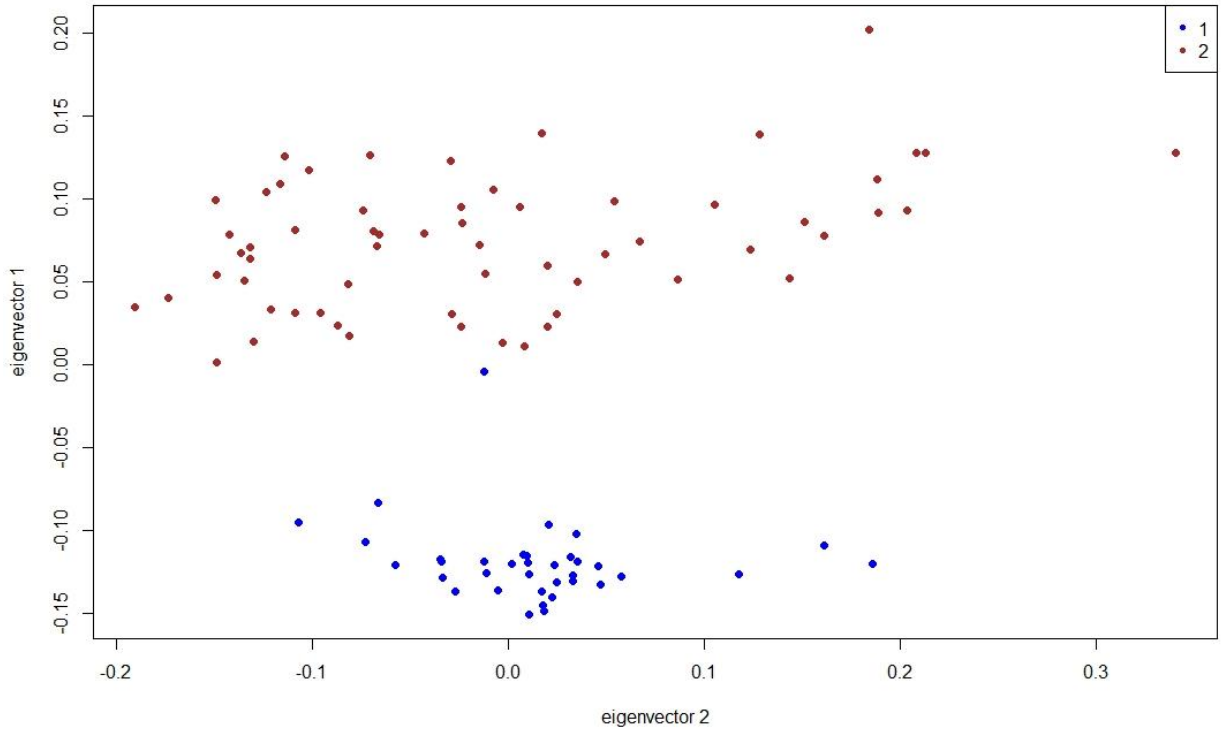


Figure 2. A. Damerogram based on IBS in two potato varieties B. Principal Component Analysis for the two potato groups studied (*S. tuberosum* Phureja group and *S. tuberosum* Andigena group). PC1 vs. PC2 are shown.

Table 1. STRUCTURE statistics resulting from the analysis of 2 potato populations (S. tuberosum Phureja group and S. tuberosum Andigena group) using 968 SNPs. Delta K analysis selected K = 2 optimal clusters (Blue).

K	Reps	Mean LnP(K)	Stdev LnP(K)	Ln'(K)	|Ln''(K)|	Delta K
1	10	-9.766.101.000	336.269	NA	NA	NA
2	10	-6.579.998.000	1.089.534	3.186.103.000	2.900.421.000	266.207.467
3	10	-6.294.316.000	110.280.811	285.682.000	17.318.000	0.15704
4	10	-6.025.952.000	19.258.785	268.364.000	148.142.000	769.218
5	10	-5.905.730.000	76.795.321	120.222.000	37.948.000	0.49414
6	10	-5.747.560.000	63.837.948	158.170.000	119.500.000	187.193
7	10	-5.708.890.000	30.298.121	38.670.000	13.784.000	0.45495
8	10	-5.656.436.000	52.727.151	52.454.000	45.368.000	0.86043
9	10	-5.649.350.000	57.121.098	7.086.000	21.439.000	0.37533
10	10	-5.663.703.000	77.265.451	-14.353.000	NA	NA

The results of the analysis of observed versus expected values indicate some degree of deviation of p-values from the null hypothesis for all phenotypic variables (Supplementary Figure **2**-S11). The results were similar for all models evaluated (general, additive, diplo-general, diplo-additive), with, in general, the additive and diplo-additive models showing the lowest degree of inflation of p-values (Supplementary Figure **2**-S11). Population structure (population 1 and 2) and kinship matrix were factors incorporated in the GWAS study (Rosyara et al., 2016), however, no substantial improvements in data distribution were presented. Quantile-quantile plots (*qqplot*) showed certain deviations from normal distribution for phenotypic variables (Supplementary Figure **2**-S11). These results could be influenced by the low number of markers and the small population size of this study. Furthermore, Ray & Chatterjee (2020) assert that kinship adjustment does not guarantee normal distribution of residuals, when outliers exist and/or are strongly correlated. Nevertheless, it was assumed that the variables are normally distributed, even though there may be a loss in statistical power and/or the false positive rate (type I error) may increase(Verhulst & Neale, 2021). Threshold correction for the identification of QTLs was performed using the Multiple Testing test for GWAS proposed by Moskvina & Schmidt (2008), with a confidence level of $p<0.05$ (

Table **2**2). SNP-type markers were associated with traits such as days to flowering, plant height, relative water content and number of tubers per plant (

Table **2**2). The additive and diplo-additive allelic interaction models showed the highest effect values for all phenotypic variables, while no significant effects were evident in the general and diplo-general models (

Table **2**2).

SNPs significantly associated with the variable days to flowering were identified on chromosomes 7 and 8 (

Table **2**2). The marker mapped on chromosome 7 has a larger effect compared to that detected on chromosome 8 (

Table **2**2). Manhattan plots revealed the quantitative nature of the days to flowering trait that could be governed by genetic polymorphisms on chromosomes 3, 6, 7 and 8, although some showed no significant effects (Figure **3**3). Significant SNPs were associated with genes with functions as negative regulators of salicylic acid (G1-2; XM_006343633.2) and common regulators of signaling pathways controlling flowering time (HOS1; BG591826.1), chromosomes 7 and 8, respectively (Figure **3**3). These markers showed contrasting effects (negative for HOS1).

Table 2. Significant associations between molecular markers and phenotypic traits.

Method	Trait	Model	Threshold -log(p)	Marker	Chromosome	Position	Value -log(p)	Effect
	DF	general	2,98	Chr07_G1-2_335138	Chr07	335138	3,34	NA
	DF	general	2,98	Chr08_HOS1_51442067	Chr08	5144206 7	3,2	NA
	DF	additive	2,98	Chr07_G1-2_335138	Chr07	335138	3,34	13,29
	DF	additive	2,98	Chr08_HOS1_51442067	Chr08	5144206 7	3,2	-9,29
	DF	diplo-general	2,98	Chr07_G1-2_335138	Chr07	335138	3,34	NA
	DF	diplo-general	2,98	Chr08_HOS1_51442067	Chr08	5144206 7	3,2	NA
	DF	diplo-additive	2,98	Chr07_G1-2_335138	Chr07	335138	3,34	13,29
	DF	diplo-additive	2,98	Chr08_HOS1_51442067	Chr08	5144206 7	3,2	-9,29
	AP	general	2,98	Chr03_EST710511_753647	Chr03	753647	3,57	NA
	AP	general	2,98	Chr08_RAP4_35655810	Chr08	3565581 0	3,43	NA
	AP	general	2,98	Chr08_RAP4_35655872	Chr08	3565587 2	3,43	NA
	AP	additive	2,98	Chr03_EST710511_753647	Chr03	753647	3,57	-18,46
	AP	additive	2,98	Chr08_RAP4_35655810	Chr08	3565581 0	3,43	-15,07
	AP	additive	2,98	Chr08_RAP4_35655872	Chr08	3565587 2	3,43	-15,07
	AP	diplo-general	2,98	Chr03_EST710511_753647	Chr03	753647	3,57	NA
	AP	diplo-general	2,98	Chr08_RAP4_35655810	Chr08	3565581 0	3,43	NA
	AP	diplo-general	2,98	Chr08_RAP4_35655872	Chr08	3565587 2	3,43	NA
	AP	diplo-additive	2,98	Chr03_EST710511_753647	Chr03	753647	3,57	-18,46

	AP	diplo-additive	2,98	Chr08_RAP4_35655810	Chr08	35655810	3,43	-15,07
	AP	diplo-additive	2,98	Chr08_RAP4_35655872	Chr08	35655872	3,43	-15,07
	CRA	general	2,98	Chr01_RAP3_39237245	Chr01	39237245	3,4	NA
	CRA	additive	2,98	Chr01_RAP3_39237245	Chr01	39237245	3,4	10,59
	CRA	diplo-general	2,98	Chr01_RAP3_39237245	Chr01	39237245	3,4	NA
	CRA	diplo-additive	2,98	Chr01_RAP3_39237245	Chr01	39237245	3,4	10,59
Multiple Testing Correction for GWAS Moskvina and Schmidt (2008)	NTP	general	2,98	Chr01_RAP3_40201293	Chr01	40201293	3,17	NA
	NTP	additive	2,98	Chr01_RAP3_40201293	Chr01	40201293	3,17	5,19
	NTP	diplo-general	2,98	Chr01_RAP3_40201293	Chr01	40201293	3,17	NA
	NTP	diplo-additive	2,98	Chr01_RAP3_40201293	Chr01	40201293	3,17	5,19

Association studies between marker and phenotypic trait were performed using the methodology described by Rosyara et al., 2016, threshold correction was performed for multiple tests following Moskvina & Schmidt (2008). Days to flowering (DF), plant height (PA), relative water content (CRA), number of tubers per plant (NTP).

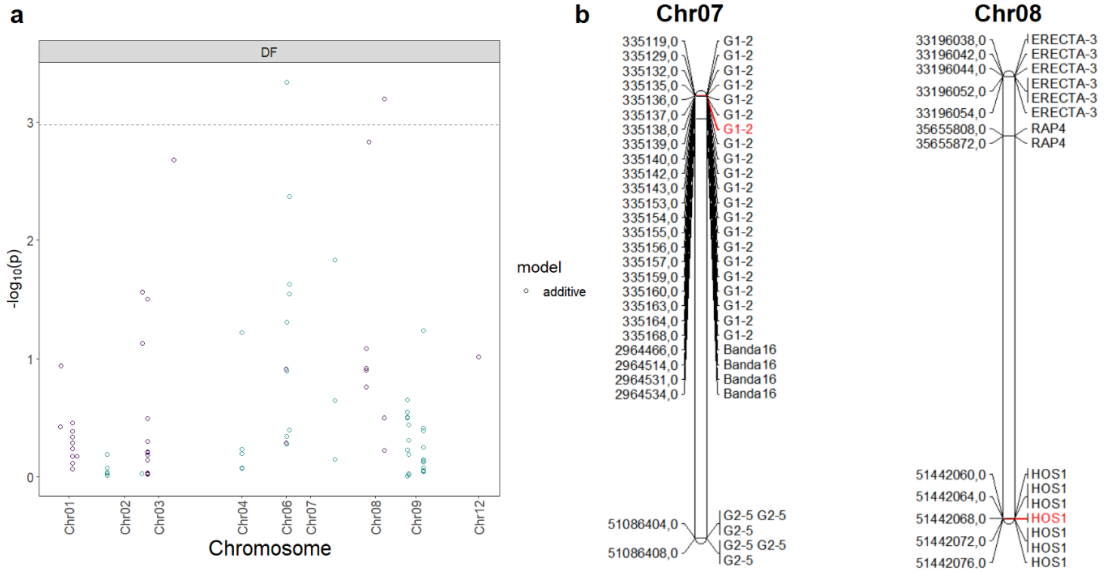


Figure 3. Manhattan plots for the variable days to flowering (DF). (a) Significant markers were identified under the threshold correction for multiple testing proposed by Moskvina & Schmidt (2008), with a significance level p<0.05. (b) Relative position regions with the presence of at least one polymorphic nucleotide (SNP) identified over the reference genome. (b) Significant SNPs are highlighted in red. Chromosome 7 and 8 (Chr07 and Chr08, respectively).

The role of HOS1 genes in the flowering response has been extensively studied. *HOS1 (HIGH EXPRESSION OF OSMOTICALLY RESPONSIVE GENES 1)* proteins regulate multiple aspects of plant growth through ubiquitin-dependent proteolysis, including physiological processes such as leaf development, seed dormancy, cell cycle, vegetative to reproductive growth transition (Lee et al., 2012). Thus, HOS1 proteins regulate the transcriptional activity of genes in a large protein complex that together control flowering through chromatin modifications. Ubiquitination of specific histones affects the activity of genes such as *FLOWERING LOCUS C (FLC*), which encodes a transcription factor that represses flowering (Linden & Callis, 2020). Consequently, *hos1* mutants of *Arabidopsis thaliana* flower earlier than non-transformed *(wild type*) plants (Lazaro et al., 2012). Therefore, genetic polymorphisms associated with earlier potato cultivars may be related to the functional loss of *HOS1* genes. In favor of this

hypothesis, QTLs in *HOS1* regions that are associated with early flowering time have been identified and mapped in species such as *Cucurbita pepo* (Qu et al., 2022) and *A. thaliana* (Ehrenreich et al., 2009). Inactivation of HOS1 genes via CRISPR-Cas9 could be an interesting strategy for the generation of earlier cultivars, once the potato genome editing technique has been well established (Tiwari et al., 2022)_

On the other hand, G1-2 markers are associated with *EDR2 (ENHANCED DISEASE RESISTANCE 2-like*) genes involved in salicylic acid-mediated pathogen response regulatory functions. QTLs for *EDR2* have been candidates for use in marker-assisted breeding for resistance to anthracnose (Bredeson et al., 2022) and mildew (Wang et al., 2011). Association studies by GWAS indicate that *EDR2* provides ability to enhance developmental processes probably due to control of several hormonal regulatory pathways (Yeboah et al., 2021).

The results indicate that selection for genetic traits can contribute to the genetic improvement and/or genetic transformation of potato plants oriented to the generation of earlier cultivars. Currently, the induction of mutations for the *HOS1* gene could be a useful alternative for the generation of potato varieties with reduced flowering time, which is correlated with tuber formation time.

SNPs significantly associated with the variable Relative Water Content were identified on chromosome 1 (

Table **2**2). Manhattan plots revealed that this variable could be governed by genetic polymorphisms on chromosomes 6 and 9, although they did not show significant effects (Figure 4). The significant SNP was associated with genes encoding MYC-type *Helix-Loop-Helix* (HLH) transcription factors (RAP-3; JZ168070.1). These proteins are part of a large family of transcription factors, which possess basic amino acid residues with high DNA binding capacity (Boter et al., 2004). *In silico* analyses show that the DNA-binding element presents the conserved -CANNTG- motif, which has affinity for a 17 amino acid residue in the N-terminal region(Atchley & Fitch, 1997).

bHLH proteins are mainly located in the cell nucleus and are expressed in stems and new leaves. The literature indicates that potato plants tolerant to abiotic stress conditions significantly increase the transcriptional activity of *bHLH-type* genes compared to susceptible plants (Filiz & Kurt, 2021). Analysis of RNA-seq data revealed that *bHLH-type* transcription factors are differentially expressed when potato plants are subjected to drought stress In *S. lycopersicum*, *bHLH* gene expression renders transgenic plants more tolerant to drought and salinity compared to non-genetically modified plants (Waseem et al., 2019). Similarly, in *Arabidopsis*, plants transgenic for *bHLH* genes are more tolerant to cold, due to increased production of enzymes that are part of the antioxidant system (Pan et al., 2022). Overexpression of *bHLH* increases the production of enzymes such as peroxidases, superoxide dismutases, malondialdehyde content, soluble proteins and proline (Pan et al., 2022). The latter proteins play a key role in the catabolism of reactive oxygen species and cellular osmoregulation. Thus, there is a positive correlation between total proline content and relative water content values (Bayat & Moghadam, 2019). Consequently, relative water content and chlorophyll content increases in plants transgenic for *bHLH* genes , as demonstrated in *S. lycopersicum* (Waseem et al., 2019). For this reason, association studies have identified *bHLH* genes as responsible for resistance to abiotic factors such as low temperature and salinity in *Zoysia japónica* (Xu et al., 2022; Zuo et al., 2021). However, genes encoding bHLH transcription factors may be useful for improving the water status of potato plants.

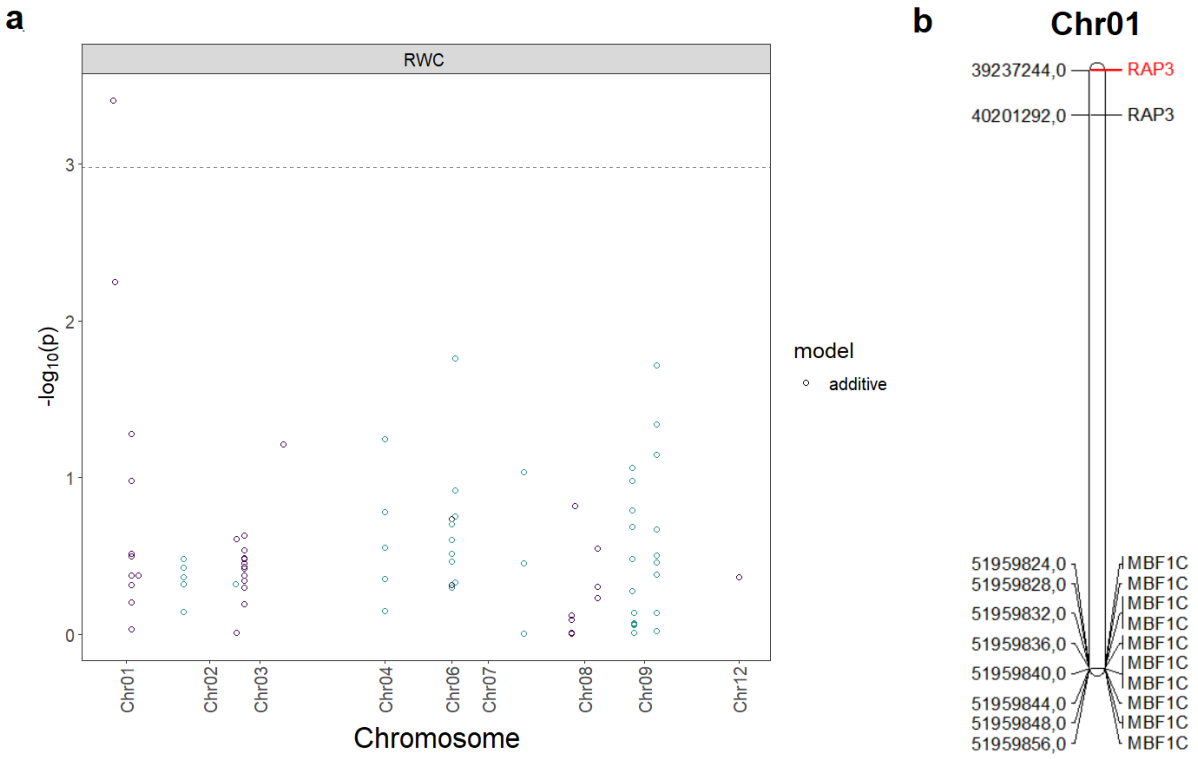


Figure 4. Manhattan plots for the relative water content (CRA) variable. (a) Significant markers were identified under the threshold correction for multiple testing proposed by Moskvina & Schmidt (2008), with significance level $p<0.05$. (b) Relative

position regions with the presence of at least one polymorphic nucleotide (SNP) identified over the reference genome. Significant SNPs are highlighted in red (b). Chromosome 1 (Chr01).

One SNP was significantly associated with the variable Number of Tubers per Plant mapped on chromosome 1 (

Table **2**2). Manhattan plots revealed that other genetic polymorphisms on the same chromosome could be influencing the behavior of this variable, however, they did not show significant effects (

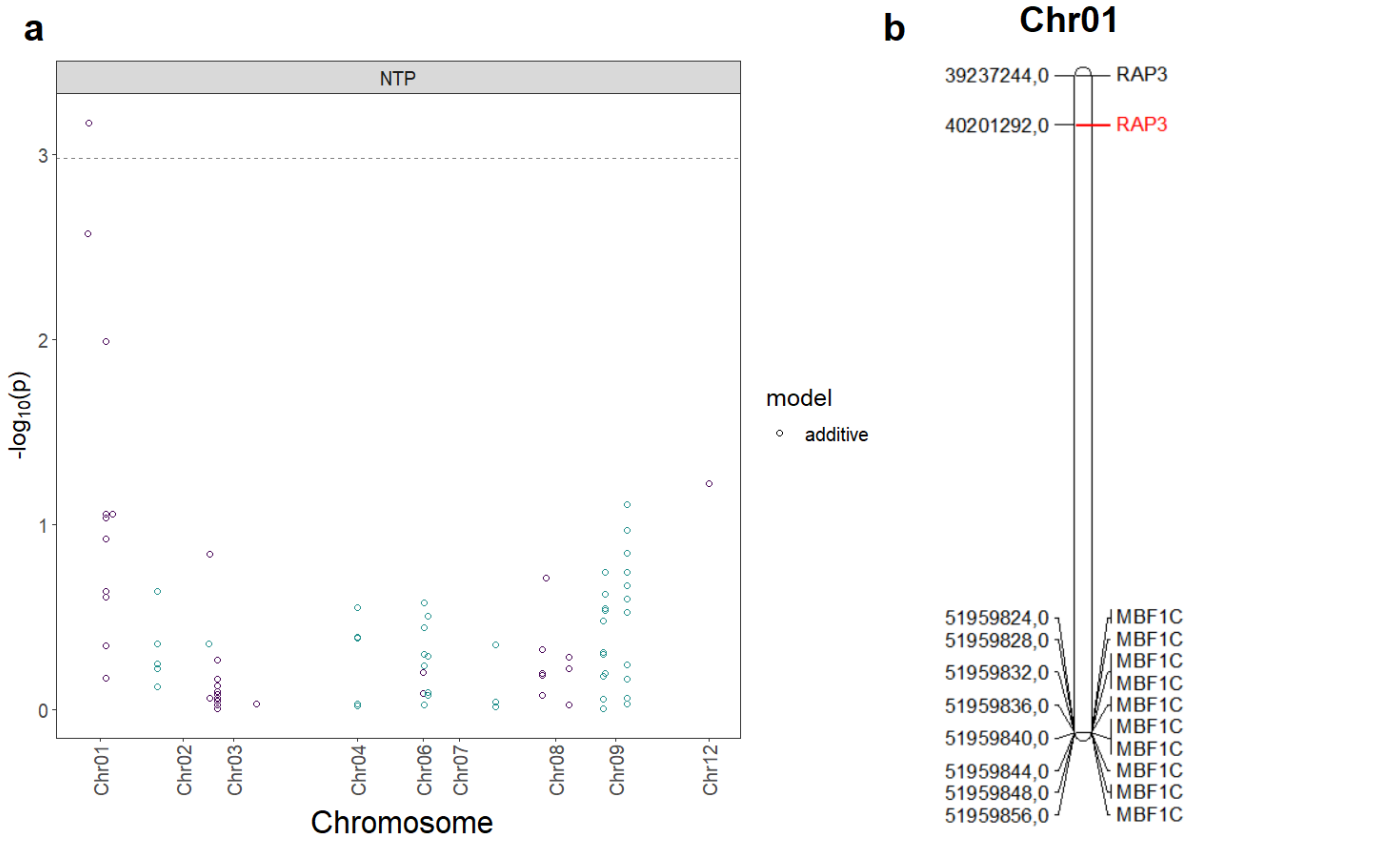


Figure 5). The significant SNP was associated with genes encoding MYC-type *bHLH* transcription factors, as described above. The variable number of tubers per plant could be indirectly influenced by plant water balance. Although mutations in distinct regions of the same gene suggest that multifunctional domains may exist in *bHLH* proteins, acting independently.

In potato, whole genome characterization of these genotypes revealed 259 *bHLH* genes transcription factors of which, could be phylogenetically divided into 15 groups(Wang et al., 2018). So that, *bHLH* genes can in various physiological processes, including in growth, influence plant development and abiotic stress response. The proteins have been implicated in processes such as floral (Hudson & Hudson, 2015) and root developmental response (Karas et al., 2009). Thus, *bHLH* gene mutants are affected in the production of root hairs in *Lotus japonicus* (Karas et al., 2009). On the other hand, studies show an expansion of *bHLH* genes in the genomes of *L. japonicus, Arabidopsis*, *O. sativa* and *S. tuberosum* (Filiz & Kurt, 2021; Karas et al., 2009; Zuo et al., 2021, 2023), which could favor the possibility of acquiring new functions, including participation in physiological processes oriented to the production of tubers per plant.

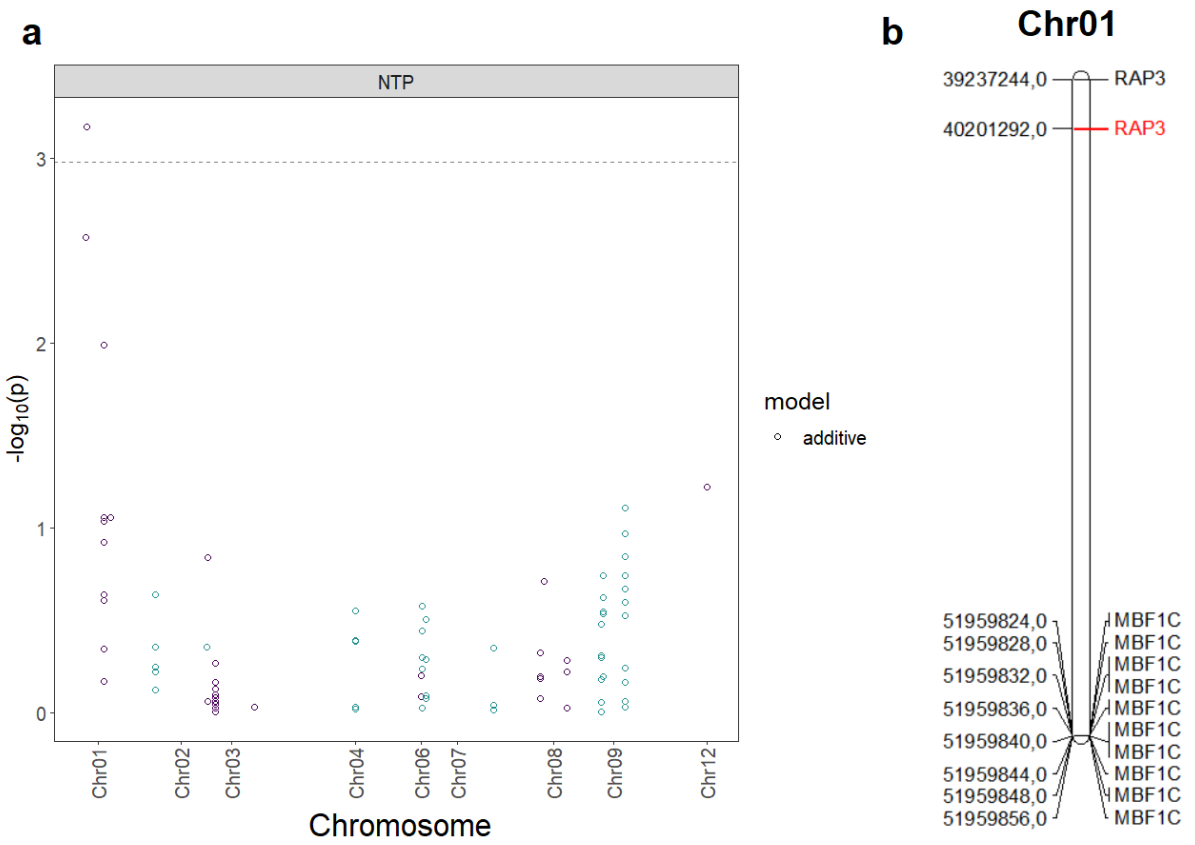


Figure 5. Manhattan plots for the variable number of tubers per plant (NTP). (a) Significant markers were identified under the threshold correction for multiple testing proposed by Moskvina & Schmidt (2008), with a significance level $p<0.05$. (b) Relative position regions with the presence of at least one polymorphic nucleotide (SNP) identified over the reference genome (b). Significant SNPs are highlighted in red. Chromosome 1 (Chr01).

In addition, MYC-type transcription factors act directly in the production pathways of jasmonic acid, which plays a critical role in tuberization processes in *S. tuberosum* (Begum et al., 2022). Jasmonic acid interacts antagonistically with gibberellic acid, stimulating tuber initiation and bulking (Aksenova et al., 2012). Even, exogenous application of jasmonic acid promotes tuberization in potato explants grown *in vitro* (Koda et al., 1991; Pervaiz et al., 2023). The response of potato plant tissue to jasmonate is directly dependent on the interaction between jasmonic acid co-receptors (JAZ proteins) and MYC-type transcription factors(Lorenzo et al., 2004). Thus, transcription factors play an indirect role in tuber production, due to the interaction with proteins involved in jasmonic acid signaling pathways. Hormonal imbalance caused by MYC-type transcription factors (RAP-4; BQ508783.2) could have favored

the growth of potato plants (

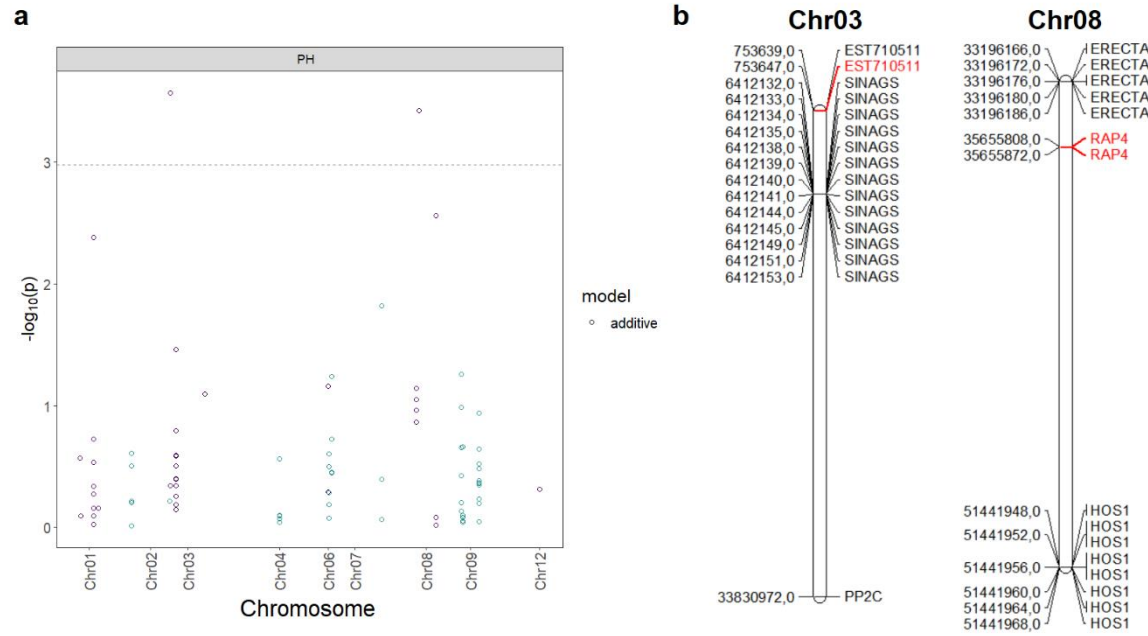


Figure). In rice, for example, Heang & Sassa (2012) showed that these regulatory proteins control organ growth by controlling cell length.

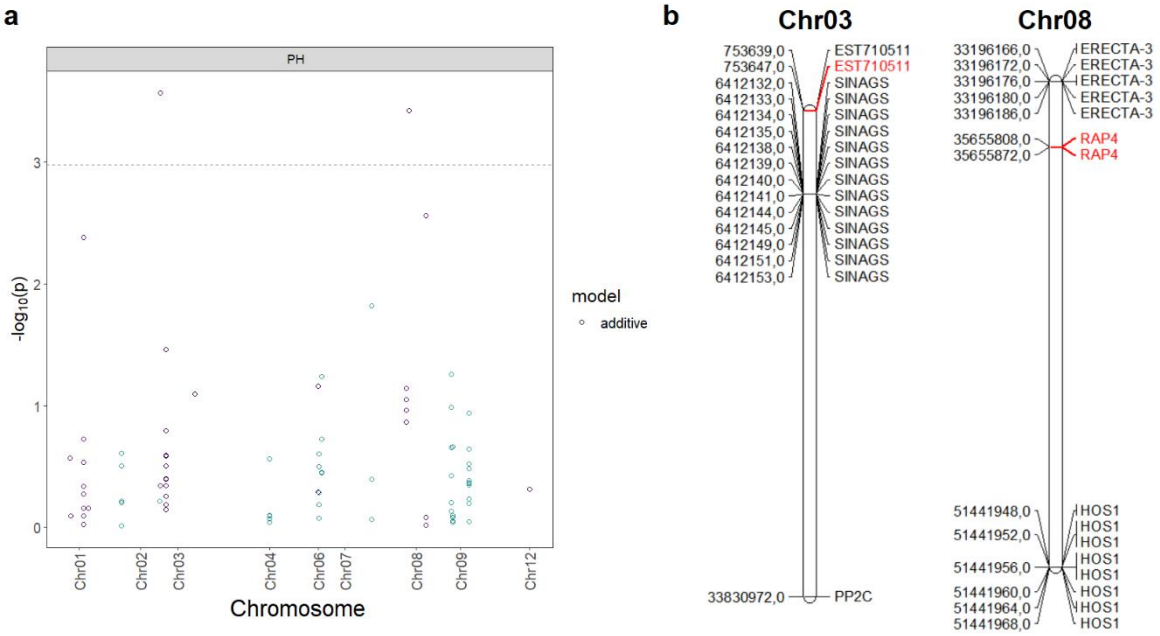


Figure 6. Manhattan plots for the plant height variable. (a) Significant markers were identified under the threshold correction for multiple testing proposed by Moskvina & Schmidt (2008), with a significance level *p*<0.05. (b) Relative position regions with the presence of at least one polymorphic nucleotide (SNP) identified over the reference genome. Significant SNPs are highlighted in red. Chromosome 3 (Chr03) and Chromosome 8 (Chr03).

SNPs encoding proteins involved in photosynthesis were significantly associated with the variable plant height. This SNP was mapped on chromosome 3 (

Table 22). Manhattan plots indicate that other genetic polymorphisms could be influencing the behavior of this variable, including on chromosome 1, 6 and 7, however, they did not present significant effects (Figure 6). Marker EST710511 (XM_006356011.2) was significantly associated with genes encoding chloroplastidial proteins. Alteration in these proteins causes subsequent reduction in plant vigor in terms of growth and yield. Stunting is caused by reduced stromal electron flow, which decreases photosynthetic efficiency (Yang et al., 2020). This trait is an important attribute to consider incorporating into potato

breeding programs for drought-resistant genotypes. However, this trait would be negatively selected when considering that larger plants are usually less efficient in the use of water and nutrients (Tolessa, 2019). For this reason, the effects of the above two attributes for plant height have negative effects, suggesting that these traits can be discarded in selection plans for water stress tolerance.

The relationship between the markers described here and phenotypic traits should be validated by crosses between populations with contrasting traits. Similarly, it is possible to take advantage of information available in public repositories such as SolCAP SNP, NCBI, in order to provide better resolution mappings in the identification of marker-phenotypic trait associations. Marker-assisted selection will be of great value in the challenging efforts undertaken for the systematic understanding of polygenic traits. Certainly, studies in this area will have a significant impact on potato breeding for complex traits.

CONCLUSIONS

Six markers were associated with four phenotypic traits linked to the resistance of potato genotypes resistant to drought stress. SNPs were associated with traits such as days to flowering, relative water content, number of tubers per plant and plant height. Negative regulators for earliness could be associated with HOS1 genes. The qualitative nature of the trait suggests that it can be directly introgressed into improved cultivars or inactivated by genetic engineering techniques. MYC-type transcription factors may be associated with plant water balance and vigor in terms of plant height and number of tubers per plant. The results suggest that the expansion of these genes contributes to the functional divergence of this gene family, which could undergo positive and negative selection processes. It is necessary to increase the number of genotypes evaluated and the genome coverage with a greater number of markers to discern with greater precision the molecular basis of the tolerance of potato genotypes to water deficit stress.

SUPPLEMENTARY MATERIAL

Supplementary Table 1. List of 87 candidate genes associated with water stress traits in potato

No.	Name	ID	Description	bp
1	AREB	CK262297.1	EST708375 potato abiotic stress cDNA library Solanum tuberosum cDNA clone POABF34 3' end, mRNA sequence	897
2	ASK-C3	NM_001287965.1	Solanum tuberosum AP2 domain CBF protein (CBF3), mRNA	1001
3	ASK-H1	XM_006351549.2	Solanum tuberosum 17.3 kDa class II heat shock protein-like (LOC102604529), mRNA	817
4	At1-1	XM_006350239.2	Solanum tuberosum probable pectate lyase 8 (LOC102587929), mRNA	2062
5	At1-2	XM_006350239.2	Solanum tuberosum probable pectate lyase 8 (LOC102587929), mRNA	2062
6	AtHB-7	XM_006339695.2	Solanum tuberosum homeobox-leucine zipper protein ATHB-12 (LOC102596484), mRNA	1290
7	B1	XM_006348054	Solanum tuberosum peroxidase 42-like (LOC102596217), mRNA	1294
8	B14	XM_006354292	Solanum tuberosum cell division cycle protein 48 homolog (LOC102600837), mRNA	2696
9	B15	XM_006364243.2	Solanum tuberosum uncharacterized LOC102579399 (LOC102579399), mRNA	1021
10	B16	XM_006359474	Solanum tuberosum metal tolerance protein 1-like (LOC102592960), transcript variant X2, mRNA	1951
11	B18bis	XM_006337933.2	Solanum tuberosum beta-glucosidase-like SFR2, chloroplastic (LOC102593280), mRNA	2582
12	B19sb	XM_006348054.1	PREDICTED: Solanum tuberosum peroxidase 42-like (LOC102596217), mRNA	1294
13	B20	XM_006355435.2	Solanum tuberosum hsp70-Hsp90 organizing protein 2-like (LOC102583884), mRNA	2163

14	B8	XM_006354542.2	Solanum tuberosum 3-hydroxyisobutyryl-CoA hydrolase 1-like (LOC102602750), mRNA	1636
15	CBF-1	EU849679.1	Solanum tuberosum AP2 domain CBF protein (CBF3) mRNA, complete cds	1001
16	CBL-2	DQ222487.1	Solanum tuberosum clone 097G01 unknown mRNA	1040
17	ERD	U69633.1	Solanum tuberosum cold-stress inducible protein (C17) gene, complete cds	6251
18	ERECTA-1	AC239300.1	Solanum tuberosum strain Diploid genotype RH89-039-16 chromosome 9 clone RH047D01, *** SEQUENCING IN PROGRESS ***, 5 unordered pieces	175546
19	ERECTA-2	AC239300.1	Solanum tuberosum strain Diploid genotype RH89-039-16 chromosome 9 clone RH047D01, *** SEQUENCING IN PROGRESS ***, 5 unordered pieces	175546
20	ERECTA-3	BQ113052.2	EST598628 mixed potato tissues Solanum tuberosum cDNA clone STMCL05 5' end, mRNA sequence	646

No.	Name	ID	Description	bp
21	EST710511	XM_006356011.2	Solanum tuberosum chlorophyll a-b binding protein 3C, chloroplastic-like (LOC102603980)	1074
22	F14J22.7-2	CK265320.1	EST711398 potato abiotic stress cDNA library Solanum tuberosum cDNA clone POABY07 5'	975
23	G1-2	XM_006343633.2	Solanum tuberosum protein ENHANCED DISEASE RESISTANCE 2 (LOC102603249)	2722
24	G1-4	XM_015306580.1	Solanum tuberosum protein ENHANCED DISEASE RESISTANCE 2 (LOC102592298), transcript variant X3	2492
25	G2-2	XM_006358041.2	PREDICTED: Solanum tuberosum probable alpha,alpha-trehalose-phosphate synthase [UDP-forming] 7 (LOC102597579)	3656
26	G2-4	XM_006348886.2	PREDICTED: Solanum tuberosum peroxisomal and mitochondrial division factor 2-like (LOC102599271)	1372
27	G2-5	XM_006348886.2	PREDICTED: Solanum tuberosum peroxisomal and mitochondrial division factor 2-like (LOC102599271)	1372
28	G3-1	XM_006347685.2	PREDICTED: Solanum tuberosum mediator of RNA polymerase II transcription subunit 14 (LOC102583398)	6117
29	G3-1(2)	XM_006347685.2	PREDICTED: Solanum tuberosum mediator of RNA polymerase II transcription subunit 14 (LOC102583398)	6117
30	G3-3	XM_006353600.2	PREDICTED: Solanum tuberosum Golgi SNAP receptor complex member 1-1 (LOC102578556)	1201
31	G3-3(2)	XM_006353600.2	Solanum tuberosum Golgi SNAP receptor complex member 1-1 (LOC102578556)	1201
32	G4-7	XM_006348933.2	Solanum tuberosum putative E3 ubiquitin-protein ligase XBAT31 (LOC102591559)	1956
33	HOS	XM_006366918.2	PREDICTED: Solanum tuberosum myb-related protein 308-like (LOC102602711), mRNA	1117
34	HOS1	BG591826.1	EST499668 P. infestans-challenged leaf Solanum tuberosum cDNA clone BPLI10O18 5' sequence, mRNA sequence	699
35	MBF1C	XM_006351761.2	PREDICTED: Solanum tuberosum multiprotein-bridging factor 1c (LOC102594575), mRNA	730
36	PP2C	XM_006344520.2	PREDICTED: Solanum tuberosum protein phosphatase 2C 37-like (LOC102599348), mRNA	1667
37	RAP-1	HB766367.1	Sequence 115581 from Patent EP2090662	1941

38	RAP-2	BQ506074.2	EST613489 Generation of a set of potato cDNA clones for microarray analyses mixed potato tissues Solanum tuberosum cDNA clone STMGI14 5' end, mRNA sequence	621
39	RAP-3	JZ168070.1	EST268 high tempearture-stressed potato cDNA library Solanum tuberosum cDNA clone TNT268 5' similar to MYC2, mRNA sequence	450
40	RAP-4	BQ508783.2	EST616198 Generation of a set of potato cDNA clones for microarray analyses mixed potato tissues Solanum tuberosum cDNA clone STMGY33 5' end, mRNA sequence	681

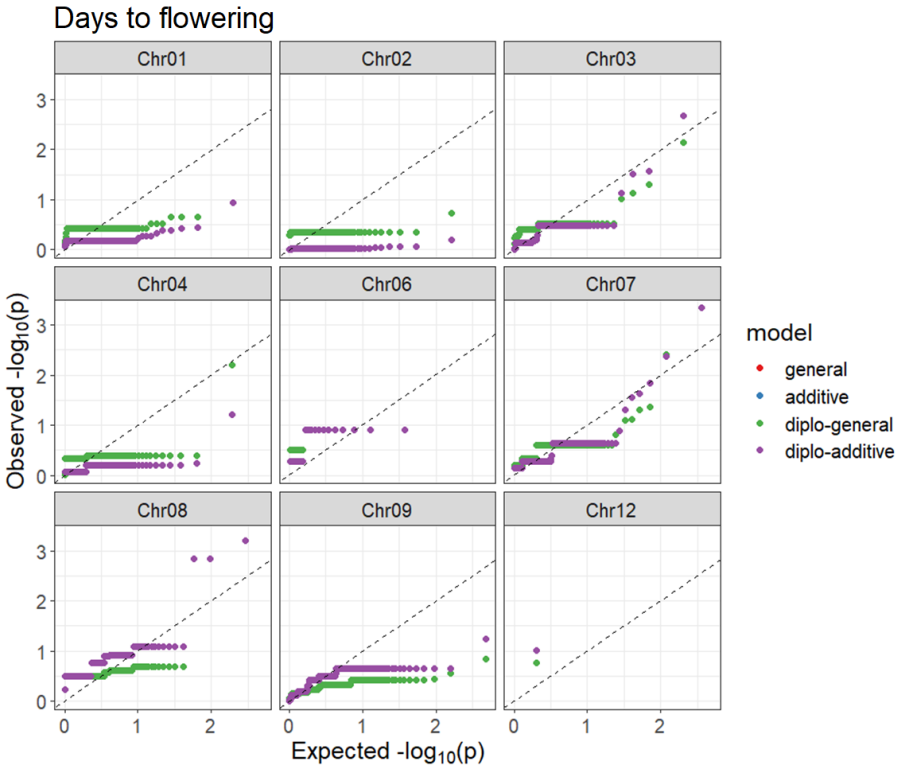
No.	Name	ID	Description	bp
41	RD22	CK277035.1	EST723113 potato abiotic stress cDNA library Solanum tuberosum cDNA clone POADX30 5' end, mRNA sequence	864
42	SAB-1	AC233625	Solanum tuberosum strain Diploid genotype RH89-039-16 chromosome 4 clone RH095M18, *** SEQUENCING IN PROGRESS ***, 15 unordered pieces	149275
43	SAB-3	AC233625	Solanum tuberosum strain Diploid genotype RH89-039-16 chromosome 4 clone RH095M18, *** SEQUENCING IN PROGRESS ***, 15 unordered pieces	149275
44	SAB-4	DR038017.1	51124.2 Late Blight-Challenged Tubers Solanum tuberosum cDNA clone 51124 5', mRNA sequence	698
45	SINAGS	XM_006350559.2	PREDICTED: Solanum tuberosum probable amino-acid acetyltransferase NAGS1, chloroplastic (LOC102598178), transcript variant X2, mRNA	2311
46	P14	Y11688.1	Solanum tuberosum mRNA for 14-3-3 protein, isolate 35G	958
47	ABCG22	XM_006348360.2	PREDICTED: Solanum tuberosum ABC transporter G family member 22-like (LOC102592336), mRNA	2678
48	ABCG40	JF440348.1	Solanum tuberosum cultivar Desiree ABCG subfamily transporter (PDR2) mRNA, complete cds	4689
49	ABH1	XM_006352524.2	PREDICTED: Solanum tuberosum nuclear cap-binding protein subunit 1 (LOC102588913), mRNA	2887
50	ABO	XM_006352524.2	PREDICTED: Solanum tuberosum nuclear cap-binding protein subunit 1 (LOC102588913), mRNA	2887
51	AHK1	XM_006340692.2	PREDICTED: Solanum tuberosum histidine kinase 1 (LOC102596157), transcript variant X1, mRNA	3937
52	Atrbo	AB198716.2	Solanum tuberosum StrbohC mRNA for NADPH oxidase, complete cds	2817
53	AVP1	XM_006359434.2	PREDICTED: Solanum tuberosum pyrophosphate-energized vacuolar membrane proton pump-like (LOC102579479), mRNA	2792
54	CaPK	AB279738.1	Solanum tuberosum StCDPK5 mRNA for calcium-dependent protein kinases, complete cds	2119
55	CHIAI	AF043248.1	Solanum tuberosum class I chitinase (ChtC2) mRNA, complete cds	1097
56	CLC	Y10338.1	Solanum tuberosum mRNA for putative chloride channel, Stclc1	2814
57	CPK21	JF308510.1	Solanum tuberosum calcium-dependent protein kinase 3 (CDPK3) gene, complete cds	11020

58	NCED1	JF308510.2	Solanum tuberosum calcium-dependent protein kinase 3 (CDPK3) gene, complete cds	11020

No.	Name	ID	Description	bp
59	CYP70	XM_006359890.2	PREDICTED: Solanum tuberosum abscisic acid 8'-hydroxylase 1 (LOC102605008), mRNA	1918
60	GORK	XM_006360318.2	PREDICTED: Solanum tuberosum potassium channel SKOR-like (LOC102595275), mRNA	3561
61	HDA6	XM_006347371.2	PREDICTED: Solanum tuberosum histone deacetylase 6-like (LOC102587267), mRNA	1973
62	HDA191	XM_006360267.2	PREDICTED: Solanum tuberosum histone deacetylase 19 (LOC102606021), mRNA	1741
63	KSU	GN102501.1	Sequence 7282 from Patent WO2009037279	1497
64	LHCB6	XM_006351735.2	PREDICTED: Solanum tuberosum chlorophyll a-b binding protein CP24 10B, chloroplastic (LOC102586172), mRNA	963
65	MRP41	XM_006359321.2	PREDICTED: Solanum tuberosum ABC transporter C family member 4 (LOC102595270), transcript variant X1, mRNA	4914
66	PER64	XM_006340884.2	PREDICTED: Solanum tuberosum peroxidase 64-like (LOC102601994), mRNA	1217
67	PER	XM_006356861.2	PREDICTED: Solanum tuberosum peroxidase 3-like (LOC102589695), mRNA	1232
68	PIP1	XM_015305194.1	PREDICTED: Solanum tuberosum aquaporin PIP2-1-like (LOC102587816), mRNA	1012
69	VPP1	XM_006350054.2	PREDICTED: Solanum tuberosum pyrophosphate-energized vacuolar membrane proton pump (LOC102600043), mRNA	2653
70	PRX	XM_006348054.1	PREDICTED: Solanum tuberosum peroxidase 42-like (LOC102596217), mRNA	1294
71	AUXIN	XM_006363449.2	PREDICTED: Solanum tuberosum indole-3-acetic acid-amido synthetase GH3.6 (LOC102584145), mRNA	2141
72	CDL	XM_015306653.1	PREDICTED: Solanum tuberosum serine/threonine-protein kinase CDL1-like (LOC102604142), partial mRNA	900
73	APX	BI978844.1	xG12 Old Blush petal SMART library Rosa chinensis cDNA 5' similar to cytosolic ascorbate peroxidase (APX), mRNA sequence	759
74	BADH	GO500045.1	Mdrtc1028C07.g1 Apple_EST_Mdrtc Malus hybrid rootstock cDNA 5' similar to ref|NP_565094.1| betaine-aldehyde dehydrogenase (BADH) [Arabidopsis thaliana] sp|Q9S795|DHAB_ARATH Betaine-aldehyde dehydrogenase, chloroplast precursor (BADH) pir|H96778 hypothetical protein F9E10.23 [imported] - Arabidopsis thaliana gb|AAD55284	668
75	DGR	GW480279.1	CA00-XX-FB1-111-B10-BG.F Coffea arabica FB1 Coffea arabica cDNA clone CA00-XX-FB1-111-B10-BG, mRNA sequence	592
76	EFF	GW402174.1	WS-Y-08 R74 Withania somnifera cv. WS-Y-08 root tissue Withania somnifera cDNA clone WS-Y-08 3' similar to Lycopersicon esculentum clone 132745F, mRNA sequence, BT013822.1	401
77	FFT1	AJ811697.1	Bellis perennis partial mRNA for 2,1-fructan:2,1-fructan 1-fructosyltransferase precursor (fft gene)	1720

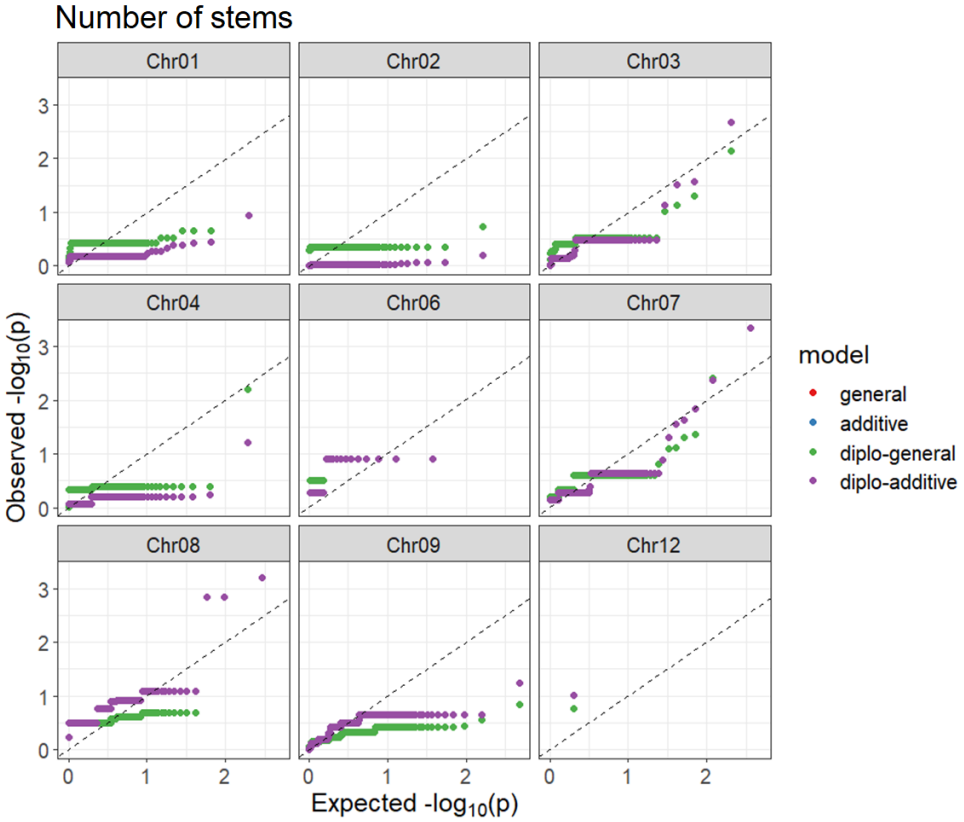
No.	**Name**	**ID**	**Description**	**bp**
78	FFT2	AJ811697.2	Bellis perennis partial mRNA for 2,1-fructan:2,1-fructan 1-fructosyltransferase precursor (fft gene)	1720

79	G6PD	GT981310.1	JGCCJG2054A04.b Jatropha curcas L. germinating seeds (mixed stages) Jatropha curcas cDNA clone JGCCJG2054A04 similar to G6PD6 (GLUCOSE-6-PHOSPHATE DEHYDROGENASE 6); glucose-6-phosphate dehydrogenase, mRNA sequence	482
80	IICB	GR222161.1	IICB_NGFSSH_SSHcDNA_6 Transgenic tobacco leaf SSH cDNA library Nicotiana tabacum cDNA clone NGF6 similar to glucan beta-1,3-glucosidase gene, mRNA sequence	679
81	PCP3	EB691605.1	NecGex_172G07 Ornamental tobacco (LxS8) Stage 6 Floral nectary cDNA library Nicotiana langsdorffii x Nicotiana sanderae cDNA clone NecGEx_Clone 172G07 similar to homologue to PIR|T05707|T05707 phosphate transport protein G7, mitochondrial - ...	571
82	Phos	DV105902.1	chiou00001 Subtractive cDNA library of roots under phosphate starvation Solanum lycopersicum cDNA clone LEPSR01A01, mRNA sequence	472
83	pk5	GT270755.1	WSR443 Withania somnifera cv. WSR08 leaf cDNA library Withania somnifera cDNA clone WSR443 3' similar to Fagus sylvatica mRNA for serine/threonine protein kinase (pk5 gene) AJ606472.1, mRNA sequence	365
84	swpa2	AF109124.2	Ipomoea batatas anionic peroxidase swpa2 (swpa2) mRNA, complete cds	1254
85	YUCCA2-1	XM_019226976.1	PREDICTED: Camelina sativa probable indole-3-pyruvate monooxygenase YUCCA7 (LOC104700128), mRNA	1733
86	KINASE	XM_006363185	Solanum tuberosum protein kinase 2B, chloroplastic (LOC102594187)	1751
87	YUCCA2-2	XM_019226976.2	PREDICTED: Camelina sativa probable indole-3-pyruvate monooxygenase YUCCA7 (LOC104700128), mRNA	1733

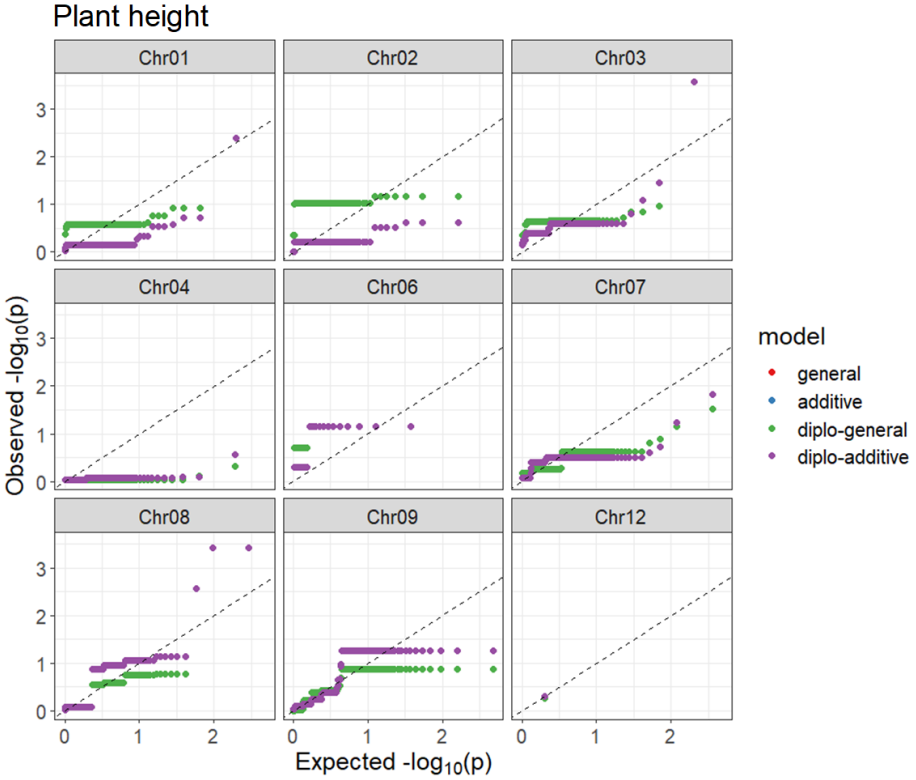


Supplementary Figure 2. Quantile-quantile plot (*QQplot*) for the days to flowering variable. The plot represents the degree of inflation of the p-values with respect to the null hypothesis. The observed -$\log_{10}$(p) vs. expected -$\log_{10}$(p) values for each genetic

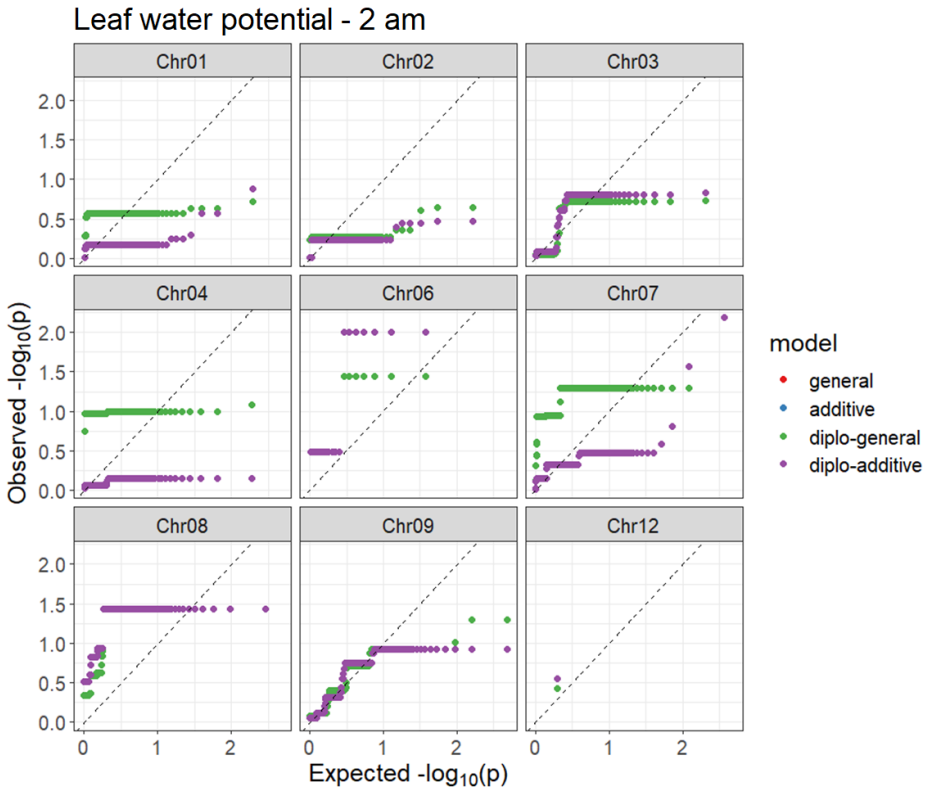
model, including the additive, general, double-general (diplo-general) and double-additive (diplo-additive) models. The line represents the x=y values under a normal distribution.



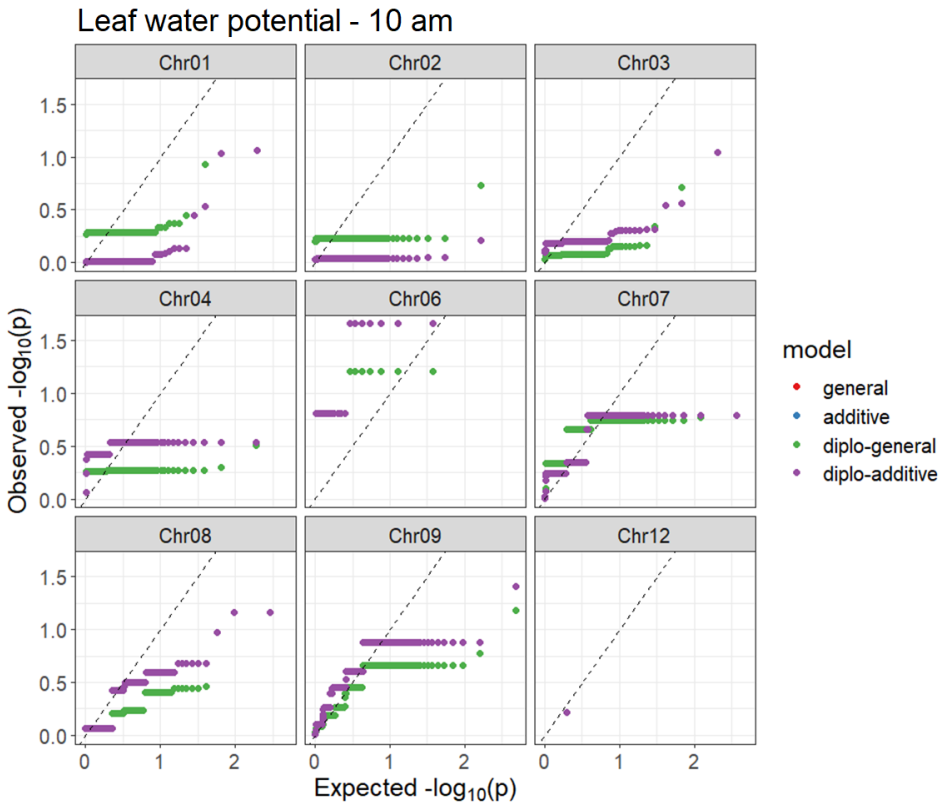
Supplementary Figure 3. Quantile-quantile plot (*QQplot*) for the number of stems variable. The plot represents the degree of inflation of the p-values with respect to the null hypothesis. The observed -$\log_{10}$(p) vs. expected -$\log_{10}$(p) values for each genetic model, including the additive, general, double-general (diplo-general) and double-additive (diplo-additive) models. The line represents the x=y values under a normal distribution.



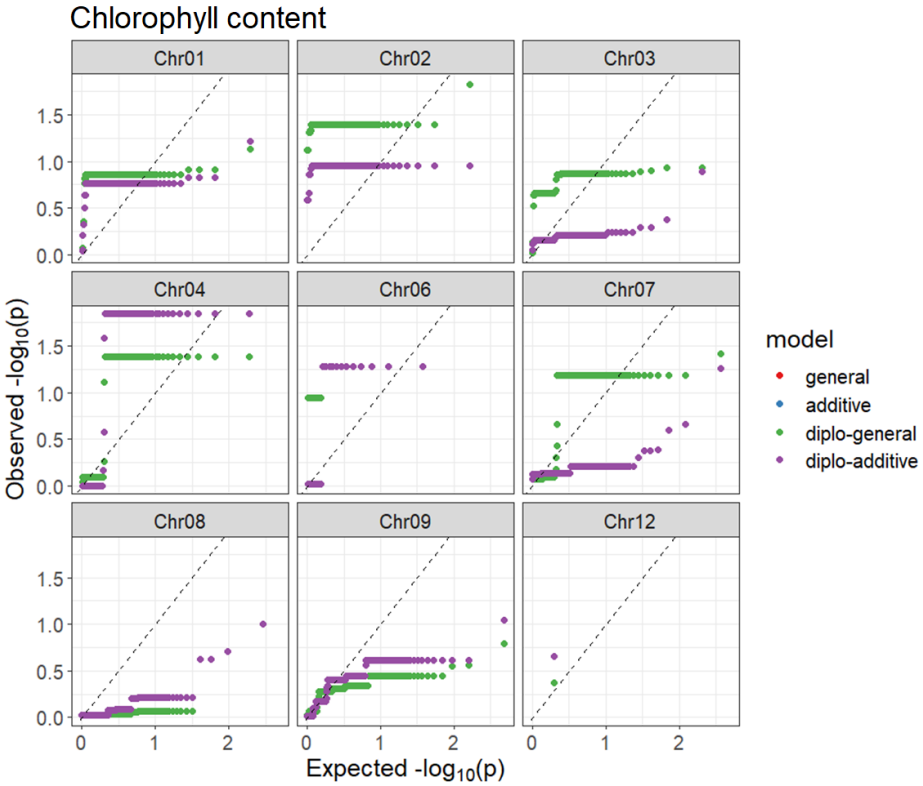
Supplementary Figure 4. Quantile-quantile plot (*QQplot*) for the plant height variable. The plot represents the degree of inflation of the p-values with respect to the null hypothesis. The observed -$\log_{10}$(p) vs. expected -$\log_{10}$(p) values for each genetic model, including the additive, general, double-general (diplo-general) and double-additive (diplo-additive) models. The line represents the x=y values under a normal distribution.



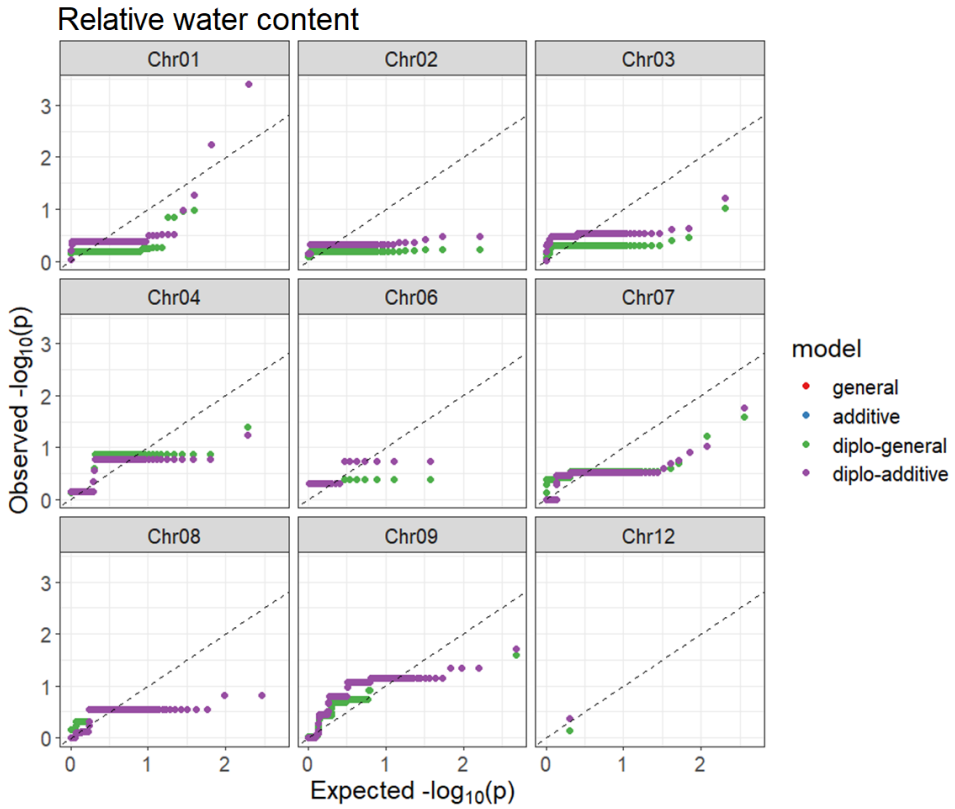
Supplementary Figure 5. Quantile-quantile plot (*QQplot*) for the leaf water potential variable (2 am). The plot represents the degree of inflation of the p-values with respect to the null hypothesis. The observed -$\log_{10}$(p) vs. expected -$\log_{10}$(p) values for each genetic model, including the additive, general, double-general (diplo-general) and double-additive (diplo-additive) models. The line represents the x=y values under a normal distribution.



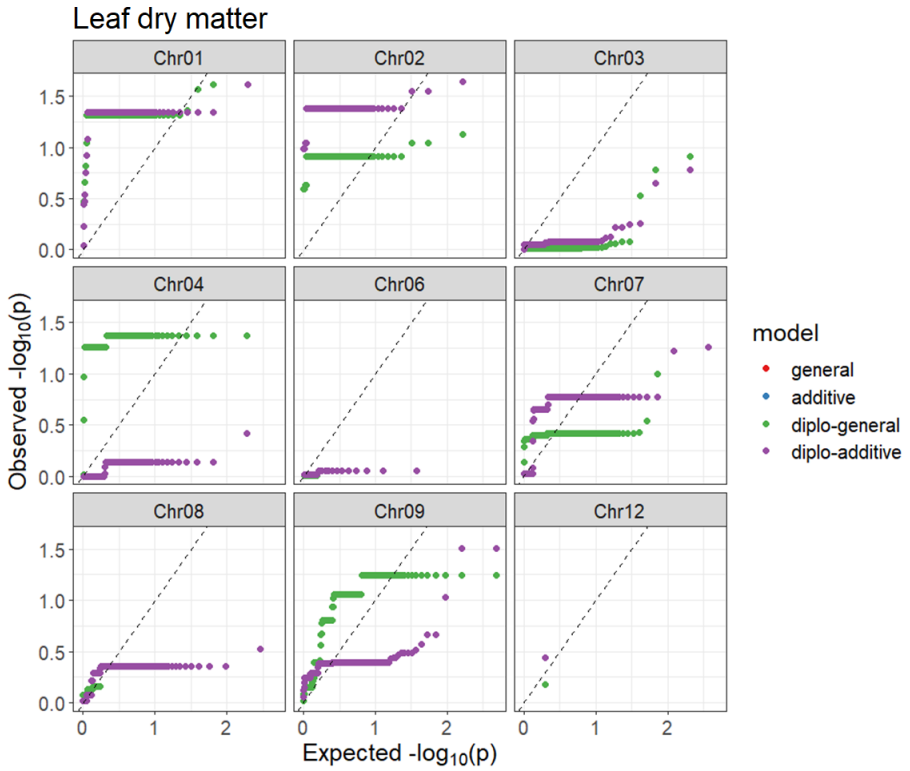
Supplementary Figure 6. Quantile-quantile plot (*QQplot*) for the daytime leaf water potential variable (10 am). The plot represents the degree of inflation of the p-values with respect to the null hypothesis. The observed -$\log_{10}$(p) vs. expected -$\log_{10}$(p) values for each genetic model, including the additive, general, double-general (diplo-general) and double-additive (diplo-additive) models. The line represents the x=y values under a normal distribution.



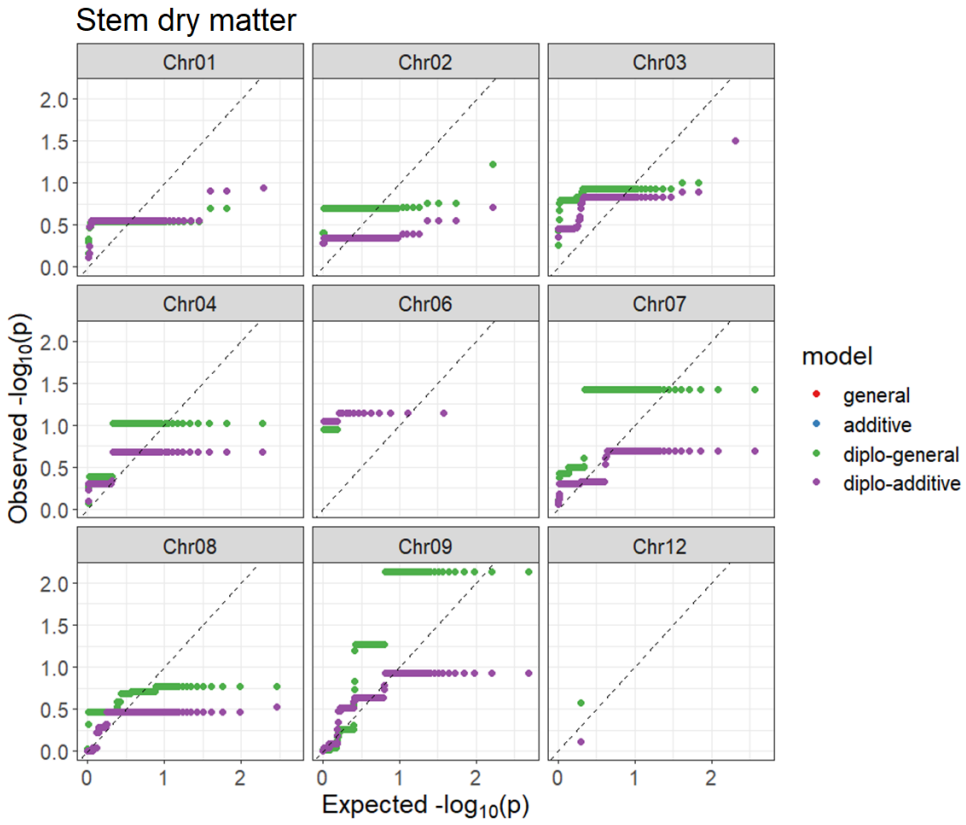
Supplementary Figure 7. Quantile-quantile plot (*QQplot*) for the total chlorophyll content variable. The plot represents the degree of inflation of the p-values with respect to the null hypothesis. The observed -$\log_{10}$(p) vs. expected -$\log_{10}$(p) values for each genetic model, including the additive, general, double-general (diplo-general) and double-additive (diplo-additive) models. The line represents the x=y values under a normal distribution.



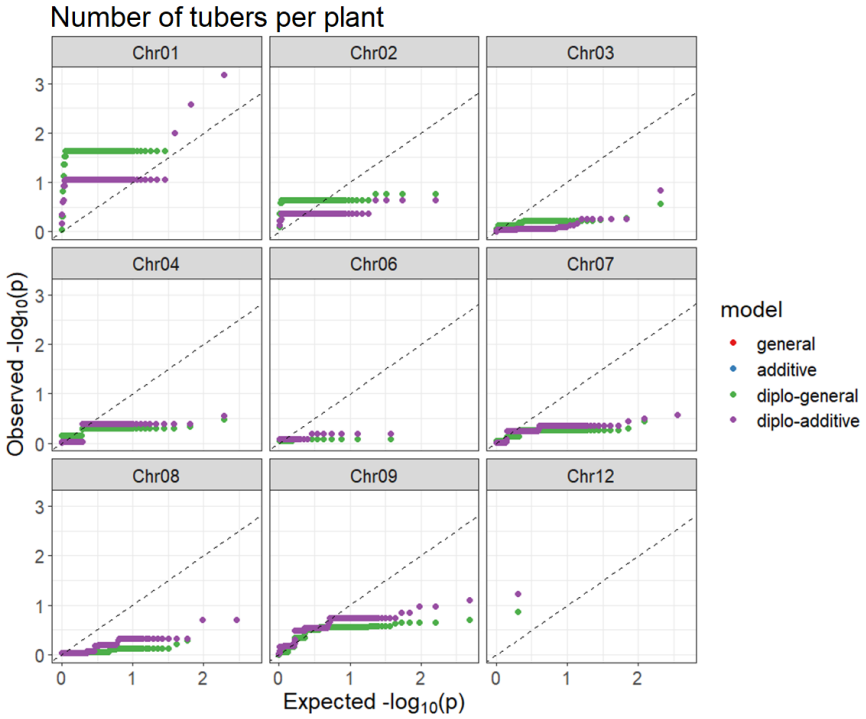
Supplementary Figure 8. Quantile-quantile plot (*QQplot*) for the relative water content variable. The plot represents the degree of inflation of the p-values with respect to the null hypothesis. The observed -$\log_{10}$(p) vs. expected -$\log_{10}$(p) values for each genetic model, including the additive, general, double-general (diplo-general) and double-additive (diplo-additive) models. The line represents the x=y values under a normal distribution.



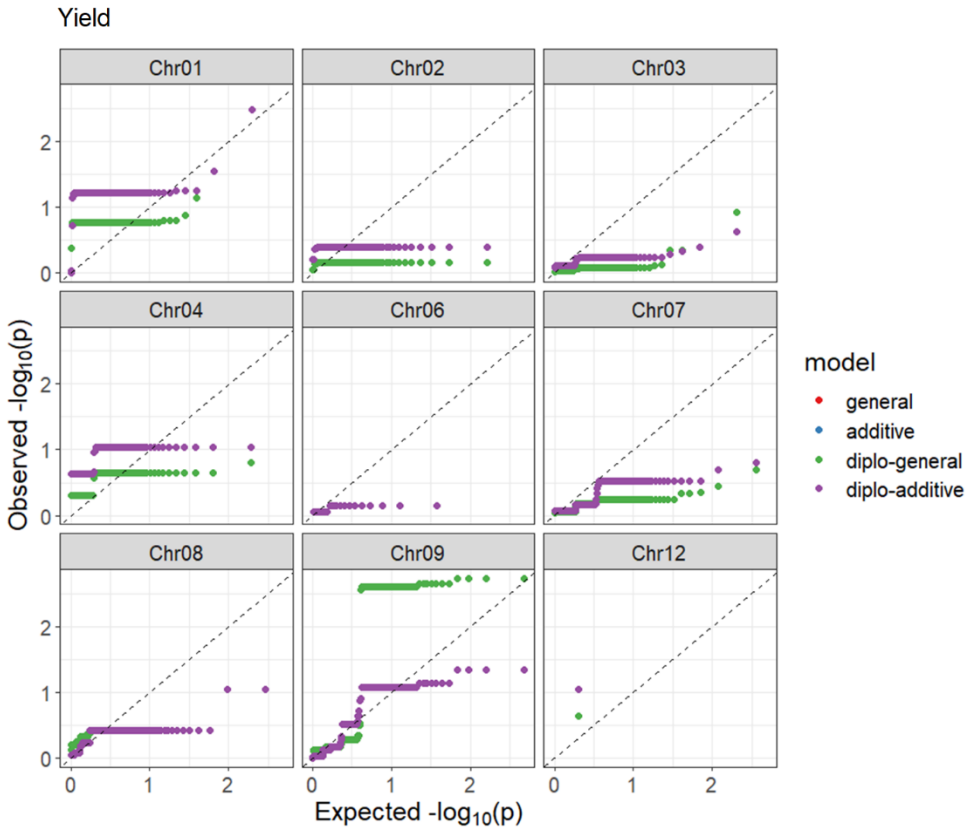
Supplementary Figure 9. Quantile-quantile plot (*QQplot*) for the leaf dry matter accumulation variable. The plot represents the degree of inflation of the p-values with respect to the null hypothesis. The observed -$\log_{10}$(p) vs. expected -$\log_{10}$(p) values for each genetic model, including the additive, general, double-general (diplo-general) and double-additive (diplo-additive) models. The line represents the x=y values under a normal distribution.



Supplementary Figure 10. Quantile-quantile plot (*QQplot*) for the tuber dry matter accumulation variable. The plot represents the degree of inflation of the p-values with respect to the null hypothesis. The observed -$\log_{10}$(p) vs. expected -$\log_{10}$(p) values for each genetic model, including the additive, general, double-general (diplo-general) and double-additive (diplo-additive) models. The line represents the x=y values under a normal distribution.



Supplementary Figure 11. Quantile-quantile plot (*QQplot*) for the variable number of tubers per plant. The plot represents the degree of inflation of the p-values with respect to the null hypothesis. The observed -$\log_{10}$(p) vs. expected -$\log_{10}$(p) values for each genetic model, including the additive, general, double-general (diplo-general) and double-additive (diplo-additive) models. The line represents the x=y values under a normal distribution.



Supplementary Figure 12. Quantile-quantile plot (*QQplot*) for the yield variable. The plot represents the degree of inflation of the p-values with respect to the null hypothesis. The observed -$\log_{10}$(p) vs. expected -$\log_{10}$(p) values for each genetic model, including the additive, general, double-general (diplo-general) and double-additive (diplo-additive) models. The line represents the x=y values under a normal distribution.

REFERENCES

1. Aksenova, N. P., Konstantinova, T. N., Golyanovskaya, S. A., Sergeeva, L. I., & Romanov, G. A. (2012). Hormonal regulation of tuber formation in potato plants. Russian Journal of Plant Physiology, 59(4), 451–466. https://doi.org/10.1134/S1021443712040024
2. Aliche, E. B., Theeuwen, T. P. J. M., Oortwijn, M., Visser, R. G. F., & van der Linden, C. G. (2020). Carbon partitioning mechanisms in POTATO under drought stress. Plant Physiology and Biochemistry, 146, 211–219. https://doi.org/10.1016/j.plaphy.2019.11.019
3. Alter, S., Bader, K. C., Spannagl, M., Wang, Y., Bauer, E., Schön, C.-C., & Mayer, K. F. X. (2015). DroughtDB: an expert-curated compilation of plant drought stress genes and their homologs in nine species. Database, 2015. https://doi.org/10.1093/database/bav046
4. Altschul, S. F., Gish, W., Miller, W., Myers, E. W., & Lipman, D. J. (1990). Basic local alignment search tool. Journal of Molecular Biology, 215(3), 403–410. https://doi.org/10.1016/S0022-2836(05)80360-2
5. Alvarez-Morezuelas, A., Barandalla, L., Ritter, E., & Ruiz de Galarreta, J. I. (2023). Genome-Wide Association Study of Agronomic and Physiological Traits Related to Drought Tolerance in Potato. Plants, 12(4), 734. https://doi.org/10.3390/plants12040734
6. Atchley, W. R., & Fitch, W. M. (1997). A natural classification of the basic helix–loop–helix class of transcription factors. Proceedings of the National Academy of Sciences, 94(10), 5172–5176. https://doi.org/10.1073/pnas.94.10.5172
7. Bayat, H., & Moghadam, A. N. (2019). Drought effects on growth, water status, proline content and antioxidant system in three Salvia nemorosa L. cultivars. Acta Physiologiae Plantarum, 41(9), 149. https://doi.org/10.1007/s11738-019-2942-6
8. Begum, S., Jing, S., Yu, L., Sun, X., Wang, E., Abu Kawochar, M., Qin, J., Liu, J., & Song, B. (2022). Modulation of JA signalling reveals the influence of StJAZ1-like on tuber initiation and tuber bulking in potato. The Plant Journal, 109(4), 952–964. https://doi.org/10.1111/tpj.15606

9. Boter, M., Ruíz-Rivero, O., Abdeen, A., & Prat, S. (2004). Conserved MYC transcription factors play a key role in jasmonate signaling both in tomato and Arabidopsis. Genes & Development, 18(13), 1577–1591. https://doi.org/10.1101/gad.297704
10. Bredeson, J. V., Lyons, J. B., Oniyinde, I. O., Okereke, N. R., Kolade, O., Nnabue, I., Nwadili, C. O., Hřibová, E., Parker, M., Nwogha, J., Shu, S., Carlson, J., Kariba, R., Muthemba, S., Knop, K., Barton, G. J., Sherwood, A. V., Lopez-Montes, A., Asiedu, R., … Rokhsar, D. S. (2022). Chromosome evolution and the genetic basis of agronomically important traits in greater yam. Nature Communications, 13(1), 2001. https://doi.org/10.1038/s41467-022-29114-w
11. D'hoop, B. B., Keizer, P. L. C., Paulo, M. J., Visser, R. G. F., van Eeuwijk, F. A., & van Eck, H. J. (2014). Identification of agronomically important QTL in tetraploid potato cultivars using a marker–trait association analysis. Theoretical and Applied Genetics, 127(3), 731–748. https://doi.org/10.1007/s00122-013-2254-y
12. Deperi, F. (2019). Identificación de factores genéticos asociados a la resistencia a tizón tardío (Phytophthora infestans Mont. De Bary) en papa tetraploide, mediante el uso de mapeo asociativo con SNPs. Escuela de posgrados. Universidad Nacional del Mar de la Plata, Argentina.
13. Devaux, A., Goffart, J.-P., Kromann, P., Andrade-Piedra, J., Polar, V., & Hareau, G. (2021). The Potato of the Future: Opportunities and Challenges in Sustainable Agri-food Systems. Potato Research, 64(4), 681–720. https://doi.org/10.1007/s11540-021-09501-4
14. Ehrenreich, I. M., Hanzawa, Y., Chou, L., Roe, J. L., Kover, P. X., & Purugganan, M. D. (2009). Candidate Gene Association Mapping of Arabidopsis Flowering Time. Genetics, 183(1), 325–335. https://doi.org/10.1534/genetics.109.105189
15. Earl DA, vonHoldt BM. STRUCTURE HARVESTER: a website and program for visualizing STRUCTURE output and implementing the Evanno method. Conserv. Genet. Resour. 2012; 4:359.
16. Evanno G, Regnaut S, Goudet J. (2005). Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. Mol. Ecol. 2005; 14(8):2611±2620. doi: 10.1111/j.1365-294X.2005. 02553.x PMID: 15969739
17. Felcher, K. J., Coombs, J. J., Massa, A. N., Hansey, C. N., Hamilton, J. P., Veilleux, R. E., Buell, C. R., & Douches, D. S. (2012). Integration of Two Diploid Potato Linkage Maps with the Potato Genome Sequence. PLoS ONE, 7(4), e36347. https://doi.org/10.1371/journal.pone.0036347
18. Filiz, E., & Kurt, F. (2021). Expression and Co-expression Analyses of WRKY, MYB, bHLH and bZIP Transcription Factor Genes in Potato (Solanum tuberosum) Under Abiotic Stress Conditions: RNA-seq Data Analysis. Potato Research, 64(4), 721–741. https://doi.org/10.1007/s11540-021-09502-3
19. Gervais, T., Creelman, A., Li, X.-Q., Bizimungu, B., De Koeyer, D., & Dahal, K. (2021). Potato Response to Drought Stress: Physiological and Growth Basis. Frontiers in Plant Science, 12. https://doi.org/10.3389/fpls.2021.698060
20. Heang, D., & Sassa, H. (2012). Antagonistic Actions of HLH/bHLH Proteins Are Involved in Grain Length and Weight in Rice. PLoS ONE, 7(2), e31325. https://doi.org/10.1371/journal.pone.0031325
21. Hudson, K. A., & Hudson, M. E. (2015). A Classification of Basic Helix-Loop-Helix Transcription Factors of Soybean. International Journal of Genomics, 2015, 1–10. https://doi.org/10.1155/2015/603182
22. Karas, B., Amyot, L., Johansen, C., Sato, S., Tabata, S., Kawaguchi, M., & Szczyglowski, K. (2009). Conservation of Lotus and Arabidopsis Basic Helix-Loop-Helix Proteins Reveals New Players in Root Hair Development. Plant Physiology, 151(3), 1175–1185. https://doi.org/10.1104/pp.109.143867
23. Kebede, A., Kang, M. S., & Bekele, E. (2019). Advances in mechanisms of drought tolerance in crops, with emphasis on barley (pp. 265–314). https://doi.org/10.1016/bs.agron.2019.01.008
24. Koda, Y., Kikuta, Y., Tazaki, H., Tsujino, Y., Sakamura, S., & Yoshihara, T. (1991). Potato tuber-inducing activities of jasmonic acid and related compounds. Phytochemistry, 30(5), 1435–1438. https://doi.org/10.1016/0031-9422(91)84180-Z
25. Koizumi, E., Igarashi, T., Tsuyama, M., Ogawa, K., Asano, K., Kobayashi, A., Sanetomo, R., & Hosaka, K. (2021). Association of Genome-Wide SNP Markers with Resistance to Common Scab of Potato. American Journal of Potato Research, 98(2), 149–156. https://doi.org/10.1007/s12230-021-09827-2
26. Lazaro, A., Valverde, F., Piñeiro, M., & Jarillo, J. A. (2012). The Arabidopsis E3 Ubiquitin Ligase HOS1 Negatively Regulates CONSTANS Abundance in the Photoperiodic Control of Flowering. The Plant Cell, 24(3), 982–999. https://doi.org/10.1105/tpc.110.081885
27. Lee, J. H., Kim, J. J., Kim, S. H., Cho, H. J., Kim, J., & Ahn, J. H. (2012). The E3 Ubiquitin Ligase HOS1 Regulates Low Ambient Temperature-Responsive Flowering in Arabidopsis thaliana. Plant and Cell Physiology, 53(10), 1802–1814. https://doi.org/10.1093/pcp/pcs123

28. Linden, K. J., & Callis, J. (2020). The ubiquitin system affects agronomic plant traits. Journal of Biological Chemistry, 295(40), 13940–13955. https://doi.org/10.1074/jbc.REV120.011303
29. Lorenzo, O., Chico, J. M., Saénchez-Serrano, J. J., & Solano, R. (2004). JASMONATE-INSENSITIVE1 Encodes a MYC Transcription Factor Essential to Discriminate between Different Jasmonate-Regulated Defense Responses in Arabidopsis[W]. The Plant Cell, 16(7), 1938–1950. https://doi.org/10.1105/tpc.022319
30. Moskvina, V., & Schmidt, K. M. (2008). On multiple-testing correction in genome-wide association studies. Genetic Epidemiology, 32(6), 567–573. https://doi.org/10.1002/gepi.20331
31. Mosquera, T., Alvarez, M. F., Jiménez-Gómez, J. M., Muktar, M. S., Paulo, M. J., Steinemann, S., Li, J., Draffehn, A., Hofmann, A., Lübeck, J., Strahwald, J., Tacke, E., Hofferbert, H.-R., Walkemeier, B., & Gebhardt, C. (2016). Targeted and Untargeted Approaches Unravel Novel Candidate Genes and Diagnostic SNPs for Quantitative Resistance of the Potato (Solanum tuberosum L.) to Phytophthora infestans Causing the Late Blight Disease. PLOS ONE, 11(6), e0156254. https://doi.org/10.1371/journal.pone.0156254
32. Ospina, N. C. A., Lammerts van Bueren, E. T., Allefs, S., Vos, P. G., van der Linden, G., Maliepaard, C. A., & Struik, P. C. (2021). Association Mapping of Physiological and Morphological Traits Related to Crop Development under Contrasting Nitrogen Inputs in a Diverse Set of Potato Cultivars. Plants, 10(8), 1727. https://doi.org/10.3390/plants10081727
33. Pan, R., Buitrago, S., Peng, Y., Fatouh Abou-Elwafa, S., Wan, K., Liu, Y., Wang, R., Yang, X., & Zhang, W. (2022). Genome-wide identification of cold-tolerance genes and functional analysis of IbbHLH116 gene in sweet potato. Gene, 837, 146690. https://doi.org/10.1016/j.gene.2022.146690
34. Pandey, J., Scheuring, D. C., Koym, J. W., & Vales, M. I. (2022). Genomic regions associated with tuber traits in tetraploid potatoes and identification of superior clones for breeding purposes. Frontiers in Plant Science, 13. https://doi.org/10.3389/fpls.2022.952263
35. Pervaiz, A., Sajid, Z. A., Yousaf, S., & Aftab, F. (2023). Microtuberization Potential of Jasmonic Acid, Kinetin and Putrescine in Potato (Solanum tuberosum L.). American Journal of Potato Research. https://doi.org/10.1007/s12230-023-09905-7
36. PGSC, T. P. G. S. C. (2011). Genome sequence and analysis of the tuber crop potato. Nature, 475(7355), 189–195. https://doi.org/10.1038/nature10158
37. Qu, S., Yang, D., Yu, H., Chen, F., Wang, K., Ding, W., Xu, W., & Wang, Y. (2022). QTL analysis of early flowering of female flowers in zucchini (Cucurbita pepo L.). Journal of Integrative Agriculture. https://doi.org/10.1016/j.jia.2022.09.009
38. Ray, D., & Chatterjee, N. (2020). Effect of non-normality and low count variants on cross-phenotype association tests in GWAS. European Journal of Human Genetics, 28(3), 300–312. https://doi.org/10.1038/s41431-019-0514-2
39. Rosyara, U. R., De Jong, W. S., Douches, D. S., & Endelman, J. B. (2016). Software for Genome-Wide Association Studies in Autopolyploids and Its Application to Potato. The Plant Genome, 9(2). https://doi.org/10.3835/plantgenome2015.08.0073
40. Saidi, A., & Hajibarat, Z. (2020). Application of Next Generation Sequencing, GWAS, RNA seq, WGRS, for genetic improvement of potato (Solanum tuberosum L.) under drought stress. Biocatalysis and Agricultural Biotechnology, 29, 101801. https://doi.org/10.1016/j.bcab.2020.101801
41. Sánchez, M. (2017). Estudio de la variabilidad genética en accesiones de papa (Solanum tuberosum L.) mediante marcadores SSRs. Ciencia y Agricultura, vol. 14, núm. 2, 2017, Julio-, pp. 67-76.
42. Tagliotti, M. E., Deperi, S. I., Bedogni, M. C., & Huarte, M. A. (2021). Genome-wide association analysis of agronomical and physiological traits linked to drought tolerance in a diverse potatoes (Solanum tuberosum) panel. Plant Breeding, 140(4), 654–664. https://doi.org/10.1111/pbr.12938
43. Tiwari, J. K., Vanishree, G., Patil, V. U., Buckseth, T., Dutt, S., Dalamu, & Singh, R. K. (2022). Genomic Designing for Abiotic Stress Tolerant in Potato. In Genomic Designing for Abiotic Stress Resistant Vegetable Crops (pp. 49–75). Springer International Publishing. https://doi.org/10.1007/978-3-031-03964-5_2
44. Tiwari, Jagesh Kumar et al. CRISPR/Cas genome editing in potato: Current status and future perspectives. Frontiers in Genetics, v. 13, p. 82, 2022.
45. Tolessa, E. S. (2019). A review on water and nitrogen use efficiency of potato (Solanum tuberosum L.) in relation to its yield and yield components. Archives of Agriculture and Environmental Science, 4(2), 119–132. https://doi.org/https://doi.org/10.26832/24566632.2019.040201
46. Verhulst, B., & Neale, M. C. (2021). Best Practices for Binary and Ordinal Data Analyses. Behavior Genetics, 51(3), 204–214. https://doi.org/10.1007/s10519-020-10031-x

47. Wang, R., Zhao, P., Kong, N., Lu, R., Pei, Y., Huang, C., Ma, H., & Chen, Q. (2018). Genome-Wide Identification and Characterization of the Potato bHLH Transcription Factor Family. Genes, 9(1), 54. https://doi.org/10.3390/genes9010054
48. Wang, Y., Nishimura, M. T., Zhao, T., & Tang, D. (2011). ATG2, an autophagy-related protein, negatively affects powdery mildew resistance and mildew-induced cell death in Arabidopsis. The Plant Journal, 68(1), 74–87. https://doi.org/10.1111/j.1365-313X.2011.04669.x
49. Waseem, M., Rong, X., & Li, Z. (2019). Dissecting the Role of a Basic Helix-Loop-Helix Transcription Factor, SlbHLH22, Under Salt and Drought Stresses in Transgenic Solanum lycopersicum L. Frontiers in Plant Science, 10. https://doi.org/10.3389/fpls.2019.00734
50. Xu, J., Xu, H., Zhao, H., Liu, H., Xu, L., & Liang, Z. (2022). Genome-wide investigation of bHLH genes and expression analysis under salt and hormonal treatments in Andrographis paniculata. Industrial Crops and Products, 183, 114928. https://doi.org/10.1016/j.indcrop.2022.114928
51. Yang, P., Li, Y., He, C., Yan, J., Zhang, W., Li, X., Xiang, F., Zuo, Z., Li, X., Zhu, Y., Liu, X., & Zhao, X. (2020). Phenotype and TMT-based quantitative proteomics analysis of Brassica napus reveals new insight into chlorophyll synthesis and chloroplast structure. Journal of Proteomics, 214, 103621. https://doi.org/10.1016/j.jprot.2019.103621
52. Yeboah, A., Lu, J., Ting, Y., Karikari, B., Gu, S., Xie, Y., Liu, H., & Yin, X. (2021). Genome-wide association study identifies loci, beneficial alleles, and candidate genes for cadmium tolerance in castor (Ricinus communis L.). Industrial Crops and Products, 171, 113842. https://doi.org/10.1016/j.indcrop.2021.113842
53. Zuo, Z.-F., Lee, H.-Y., & Kang, H.-G. (2023). Basic Helix-Loop-Helix Transcription Factors: Regulators for Plant Growth Development and Abiotic Stress Responses. International Journal of Molecular Sciences, 24(2), 1419. https://doi.org/10.3390/ijms24021419
54. Zuo, Z.-F., Sun, H.-J., Lee, H.-Y., & Kang, H.-G. (2021). Identification of bHLH genes through genome-wide association study and antisense expression of ZjbHLH076/ZjICE1 influence tolerance to low temperature and salinity in Zoysia japonica. Plant Science, 313, 111088. https://doi.org/10.1016/j.plantsci.2021.111088