

Global multi-environment resistance QTL for foliar late blight resistance in tetraploid potato with tropical adaptation

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Abstract (single paragraph of max 250 words):

The identification of environmentally stable and globally predictable resistance to potato late blight is challenged by the crop's clonal and polyploid nature and the pathogen's rapid evolution. Genome-wide analysis (GWA) of multi-environment trials can add precision to breeding for complex traits. A diversity panel of tetraploid potato germplasm bred for multiple resistance and quality traits was genotyped by genotyping by sequencing (GBS) and phenotyped for late blight resistance in a trait observation network spanning three continents addressed by the International Potato Center's (CIP's) breeding program. The aims of this study were to (i) identify QTL underlying resistance in and across environments and (ii) develop prediction models to support the global deployment and use of promising resistance sources in local breeding and variety development programs. Health-indexed *in vitro* plants of 380 clones and varieties were distributed from CIP headquarters in Peru to China and Ethiopia and tuber seed was produced centrally in each country. Phenotypes were recorded as rAUDPC following field exposure to local isolates of *Phytophthora infestans*. Stringent filtering for individual read depth >60 resulted in 3,239 tetraploid SNPs. Meanwhile, 55,748 diploid SNPs were identified using diploidized data and individual read depth >17. The kinship matrix was utilized to obtain BLUP and identify best performing germplasm in each and all environments. Genotypes with high levels of resistance in all environments were identified from the B3,

- 45 LBHT and B3-LTVR populations. GWA identified stable QTL for late blight resistance in
- 46 chromosome 9 and environment specific QTL in chromosomes 3, 5, 6 and 10.

Introduction

Potato genetic resources comprise a polyploid series consisting of a tremendously diverse germplasm of wild relatives and cultivated landraces (Spooner 2014; Ovchinnikova et al., 2011). However, most commercially cultivated potato varieties are tetraploid ($2n=4x=48$) with the genome consisting mostly of *Solanum tuberosum* Group *tuberosum* with some introgressions from a few wild species and cultivated landraces (Bradshaw et al., 2006; reviewed by Bethke et al., 2017; reviewed by Gaiero et al., 2018). Tetraploid potato is a highly heterozygous, outcrossing autopolyploid, crop which complicates genetic analysis. Most of the early genetic mapping studies utilized bi-parental populations at the simpler, diploid level ($2n=2x=24$) and several disease resistance loci were identified in the genome of potato this way (reviewed by Gebhardt and Valkonen 2001). However, this approach does not permit the assessment of large gene pools or multi-allelic interactions that influence traits in polyploids. Significant progress has recently been made in the development of algorithms and software for genotype calling, linkage and QTL analysis in polyploid species. SNP arrays have been developed for potato: 8K SolCAP (Hamilton et al., 2011) and the 20K SolSTW arrays (Vos et al., 2015). These were developed using North American and European potato germplasm, respectively, and are not consequently the best options for genotyping CIP germplasm since it contains more introgressions from the native South American gene pool. According to our previous experience, less than 50% of the SNPs on the 8K SolCAP array were informative in a test sample of CIP germplasm (Lindqvist-Kreuze et al., 2014). Genotyping by sequencing (GBS) has been applied to tetraploid potato (Uitdewilligen et al., 2013, Sverrisdottir et al., 2017); and variant calling from short read sequencing data considering allele dosage is now possible using several different tools, such as GATK, Freebayes, or SAMtools to name a few (Clevenger et al., 2015). However, reliable dosage calling in the heterozygous individuals depends on the read depth in the SNP loci. It was recently demonstrated in autopolyploid

blueberry, that a read depth of 61 was adequate to reliably call the allele dosage, while only 17 reads were needed to reliably classify simplex tetraploids as heterozygous with 95% accuracy (Matias et al., 2019). The identification of QTL in autopolyploids is facilitated by new tools, such as called GWASpoly that considers allele dosage effects (Rosyara et al., 2016). Together, these advances make genomic analysis of tetraploid potato more informative and applicable to evolutionary and breeding studies.

The goal of CIP potato breeding program is to develop resilient, high yielding, healthy and early maturing varieties for small-holder farming systems in the developing world. We are targeting farming systems that usually function with minimum input of pesticides and therefore a high level of disease resistance is an indispensable trait. To this end, CIP's potato breeding program has developed breeding populations selected for high levels of resistance to late blight caused by the oomycete *Phytophthora infestans*, and resistance to Potato Virus Y (PVY), Potato Virus X (PVX) and Potato Leaf Roll Virus (PLRV). Previous studies have identified genomic regions in CIP's breeding germplasm explaining resistance to late blight focusing on phenotypic data collected from field trials in Peru or using local pathogen strains in greenhouse conditions (Li et al., 2010; Lindqvist-Kreuze et al., 2014, Jiang Rui et al., 2018). Performance information has been sporadically published about CIP's bred materials in the target regions where they have been distributed to (Muhinyuza et al., 2015; Hirut et al., 2017b) but to our knowledge no genetic analysis has been published identifying resistance QTL for resistance in CIP germplasm tested in environments outside Peru.

The overall goal of this research was to collect data on the foliar late blight resistance of CIP's advanced tetraploid potato clones in Ethiopia and China to inform breeding decisions. To systematically evaluate CIP's breeding materials in diverse target environments we established

a trait observation network (TON) of collaborators and assembled a diversity panel that consists of representative advanced clones (including elite materials) from each of CIP's breeding populations. This so-called TON panel was then distributed from Peru to China and Ethiopia, where it was included in a series of trait evaluation experiments by national research and CIP institutions. The specific aims of this study were to (i) identify QTL underlying resistance in and across environments and (ii) develop prediction models to support the global deployment and use of promising resistance sources in local breeding and variety development programs.

We report the genotyping, estimation of linkage disequilibrium and population structure of the TON panel and identification of QTL for late blight resistance via genome wide association (GWA). In addition, we present a case for genomics assisted breeding for foliar late blight resistance and show how the use of genomics and pedigree information can be used to select best bet clones for breeding and variety development in diverse target environments.

Materials and methods

Germplasm

The TON panel used in this study consisted of 380 genotypes representing seven CIP breeding populations as well as a group of varieties with variable origins (Table 1.). 'Population A' was developed at 1980-1990 with emphasis on late blight resistance. Sources of late blight resistance were improved materials with *S. demissum*-derived resistance from breeding programs around the world, native Andean cultivars *S. tuberosum* groups *andigena*, *phureja* and *stenotomun*, wild species *S. acaule* and *S. bulbocastanum*. 'Population B3' genotypes were derived from 'Population A' with emphasis on increasing frequencies and levels of quantitative resistance to late blight. The 'B1 population' is derived from *S. tuberosum* group *andigena*.

The ‘LTVR population’ is characterized mainly for its resistance to the most important virus diseases (PVY, PVX and PLRV), short crop duration, and adaptation to warm environments. The ‘LB-HT’ population combines late blight resistance from the ‘B3 population’ and heat tolerance from North American and European bred varieties and the LTVR population. ‘B3-LTVR’ population contains hybrid genotypes originating from crosses between ‘B3’ and ‘LTVR populations’. The ‘pre-Bred’ population has genotypes that have LB resistance introduced from wild *Solanum* species into the tetraploid background of ‘B3’ or ‘LTVR’. The varieties group consists of a group of potato varieties or key breeding lines: ‘Desiree’, ‘Atlantic’, ‘Spunta’, ‘Granola’, ‘Yungay’, ‘Tomas Condemayta’, ‘DTO-33’, ‘Kufri Yoti’, and ‘Chucmarina’. CIP numbers and the parentage of the 380 genotypes are given in the Supplementary Table S1.

Environments

The field sites in Ethiopia and China are important potato production areas, while in the field site in Peru, potato is not the main crop (Table 2). The late blight pathogen populations have been described in each location. In Peru and Ethiopia only the A1 mating type has been identified and different clonal lineages are present that frequently contain virulence to most of the known *S. demissum* *R* genes (Lindqvist-Kreuze et al.; 2019 Mihretu et al., 2019). In contrast in Southern China A2 mating type has been found dominating (Chen et al., 2017).

Field trials

Standard protocols at CIP were utilized for planning and conducting the field trials (Bonierbale, 2007). The statistical designs in each trial are shown Table 2. Uniform tuber seed was produced centrally in each country following the introduction of *in vitro* plants or mini-tubers from CIP facilities in Peru or Kenya.

Late blight resistance was evaluated under endemic disease pressure. The disease level in the plots was recorded as ‘percent leaf area infected’ at 7-day intervals until susceptible controls reached 100% infection. These values were used to calculate the area under the disease progress curve (AUDPC) and relative AUDPC (rAUDPC). The data was collected and processed using the HiDAP field book system (<https://research.cip.cgiar.org/gtdms/hidap/>).

Statistical analysis of phenotypic data

From the weekly observations of the disease incidence in the plots, the AUDPC was calculated and the estimated means (BLUEs) were transformed to the relative AUDPC (rAUDPC) to facilitate the comparisons among the different locations. The best linear unbiased predictor (BLUP) and best linear unbiased estimator (BLUE) and values as well as ranked predictors were calculated using ASREML package.

Genotyping, variant calling and filtering for association analysis

In total 380 potato clones were genotyped. Library construction and genotyping by sequencing (GBS) was outsourced to the Genomics Facility at Cornell University in 2015. The DNA was digested with EcoT221 restriction enzyme and the libraries were 48x multiplexed for sequencing. The diploid calling was by the service provider using the Tassel pipeline (Bradbury et al., 2007). The resulting Variant Call Format (VCF) file was processed with Bcftools (<https://samtools.github.io/bcftools/>) to filter the variants for minimum read depth (RD) of 17, minimum genotype quality (GQ) of 30, and minor allele frequency (MAF) 0.03. The SNPs that didn’t pass these criteria were changed to missing call, and finally only the SNP sites that contained less than 30% missing data were selected.

For polyploid calling the raw FASTQ files were processed with Stacks (Catchen et al., 2013) to remove the barcodes and with TrimGalore <https://github.com/FelixKrueger/TrimGalore> to trim the ends of reads. The reads were aligned to the reference genome version *S. tuberosum_448_v4.03* (Sharma et al., 2013) using BWA (Li and Durbin, 2009) and the resulting SAM files were converted to BAM files using Samtools (Li et al., 2009). The variants were called using GATK HaplotypeCaller option (Poplin et al., 2017), disabling the duplicate read filter (this is recommended for GBS data) and joint genotyping using the -ERC GVCF mode. From the VCF files SNP calls were filtered using Bcftools for minimum RD of 61, minimum GQ of 30 and MAF of 0.03. The samples that didn't pass these criteria were changed to missing call, and finally only the SNP sites that contained less than 30% missing data were included in the analysis.

Analysis of the Population sub-structuring

Population sub-structuring with tetraploid data was done using PolyRAD (Clark et al., 2019). Only variants in pairwise LD under 0.1 were previously filtered (LD pruning). The diploid dataset was analyzed using SNPrelate (Zheng et al., 2012) using the subset of bi-allelic SNPs, filtering for LD (0.2) MAF (<0.03) and missingness (<0.3).

GWA

Marker trait associations were modelled for all the trials independently and using the diploid and the tetraploid marker sets with the GWASpoly package (Rosyara et al., 2016). For the tetraploid dataset *general*, *additive*, *simplex dominant* (1-dom) and *duplex dominant* (2-dom) models were used while for the diploid dataset *diplo-general*, *diplo-additive*, and the *simplex dominant* (1-dom) model were used. The parameters used for the GWAS modelling function *GWASpoly* in R were the following: no fixed effects, 4 principal components were included

as covariates, a minimum MAF of 0.03 and a maximum genotype frequency (after applying dominance relations) equal to 0.95 were set, and P3D approximation was used. To detect statistical significance, the Bonferroni correction method was used, ensuring the genome-wide type I error is not greater than 0.05. Manhattan plots were generated to display significant SNP in the different genetic models. In addition, Q-Q plots were used to evaluate the goodness of fit of the genetic model and the quality of the phenotypic data.

The genomic positions of the resulting SNPs associated with plausible QTL for pathogen resistance in relation to other loci, known genes and QTL, were determined using the *S. tuberosum* Group Phureja DM1-3 516R44 (v4.03) pseudomolecule browser (<http://solanaceae.plantbiology.msu.edu/>) available from the Potato Genome Sequence Consortium (PGSC). To obtain an approximate of the physical location of markers for pathogen resistance present in literature, the position in cM was obtained from the GABI Primary Database (<https://www.gabipd.org/projects/Pomamo/>) and then “translated” to an approximate physical position in Mbp using information provided in Sharma et al. (2013) that integrates the potato genome and physical and genetic maps.

Results and discussion

Population structure

The tetraploid dataset included a total of 305,345 SNPs after the GATK variant calling, while the diploidized dataset after Tassel pipeline had a SNP count of 312,727. After filtering these datasets, the Principal component analyses based on 23,804 diploid SNPs and 182,435 tetraploid variants identified no strong population sub-structuring (Figure 1 for the tetraploid data, Figure S1 for the diploid data) in the diversity panel which makes it an ideal genotype set for GWA. Only population ‘B1’ separates from the rest most likely because the genetic

background of the ‘B1’ population is *S. tuberosum* group *andigena*, while the rest are mostly group tuberosum type. ‘LB-HT’ shares alleles with the ‘B3’ population, as expected since these clones are hybrids with B3 clones in their pedigrees. ‘B3’ and ‘LTVR’ population clones are also mostly separated with a few exceptions of clones that may have been mislabelled. ‘B3-LTVR’, which is a hybrid between the two populations and this can be clearly seen in the PCA plot as well. Not surprisingly, Population ‘A’ is intermingled within population ‘B3’ since the ancestors of the ‘B3’ clones were selected clones of the ‘A’ population.

SNP sets for GWA

After applying filtering parameters tailored for GWA to all variants in both datasets, the numbers of SNPs were reduced to 3,239 tetraploid SNPs and 55,748 diploid SNPs. The DP thresholds were based on the Matias et al. 2019. The study points out, that assuming the GBS method entails 0.5% allelic error, a minimal RD of 17 is necessary in order to classify simplex tetraploid calls as heterozygous with a 95% accuracy. To annotate the allele dosage with the same accuracy, a much higher RD of 61 is needed. Both sets of filtered SNPs were used in the GWAS to identify trait-linked QTL.

Linkage disequilibrium

LD decay was estimated using the tetraploid marker set. A spline was fitted on the 90th percentile of the squared correlation coefficient between the alleles in a pair of markers (r^2) and the physical distance between these pairs of markers on the “short” distance of up to 10 Mb (Figure 2A) and “long” distance up to 80 Mb (Figure 2B) over all chromosomes. The intersection of a defined significance threshold of $r^2=0.1$ and the fitted spline allowed us to estimate the short distance vs long distance LD decay. On the short distance the threshold is reached at 2Mb, while on the long distance it is reached at 5.5Mb. Considering the short

distance LD-decay estimate of $r^2_{1/2\max, 90}$, which was suggested as the most consistent estimator for LD decay in potato by Vos et al (2017), we obtain the $r^2_{1/2\max, 90}$ value of 0.55 Mb. This $r^2_{1/2\max, 90}$ value is equivalent in Vos (2017) data for recent European potato varieties (0.6 Mb) and a bit lower than the study of Sharma et al., (2018) where the $r^2_{1/2\max, 90}$ value was 0.91 Mb. The average r^2 for the short distance in our dataset was 0.091, which is a bit lower than the average r^2 (0.19-0.22) reported for the European varieties (Vos et al., 2017), indicating that there were probably more founder haplotypes in our diversity panel than in the European pooled varieties. The LD decay estimated was moderate, and comparable to the LD decay found in the European potato germplasm. Estimates based on the average r^2 of the markers along the short distance suggest that high diversity is retained in the germplasm and that tens of thousands of markers would be needed to cover the entire tetraploid genome.

Late blight resistance

There was a high number of genotypes with rAUDPC values comparable to the resistant control genotype which is released as a variety called Chucmarina in Peru and as Belete in Ethiopia (Figure 3). Notably, most of the genotypes tested in China were more resistant than the local variety C-88 that has been popular because of its good late blight resistance.

In this research project over 300 advanced tetraploid clones from CIP were shared with partners, but due to various reasons not all were evaluated in all environments. The genotypes can only be internationally distributed as *in vitro* plants with a health certificate. After receiving the plants, there needs to be at least two rounds of multiplication involving either cuttings or tubers to obtain seed for the replicated trials. However, the performance of all genotyped clones and their pedigree parents could be estimated in all environments by incorporating the marker data into the mixed model.

The GGE biplot for predicted values (BLUP) and marker-based kinship matrix shows the performance of the genotypes and some of their parents in all environments (Figure 4). The most resistant genotypes belong to the B3 and B3-HT populations, while only a few from the LTVR and B3-LTVR had high level of resistance. From this figure, we evidenced that some genotypes' resistance to late blight is environment specific, nevertheless several genotypes show stable resistance across environments. The correlations among environments were high (Figure 5). Particularly the environment in Peru is highly correlated with all the other environments suggesting that resistant clones selected in Peru will also likely have good resistance in these other environments.

CIP's breeding strategy defined in the 1990s focused on improving the quantitative resistance in the B3 population by phenotypic recurrent selection under endemic pressure from the "new population" of *P. infestans* in the Peruvian Andes supplemented by progeny tests to identify parents with good general combining ability and eliminate those resulting in segregation for hypersensitive response against the test isolates. The pathogen population in this area is dominated by the A1 mating type and EC-1 clonal lineage, which is highly aggressive and complex in its virulence (Lindqvist-Kreuze et al., 2019). Despite the differences in the pathogen populations in terms of the mating type and clonal lineages among the countries it seems that phenotypic selection for late blight resistance in Peru was largely successful and results transferable across the three environments tested here.

QTL for late blight resistance

Several SNPs were significantly associated with late blight resistance in the field trials with a total of 16 markers tagging possible QTL (Table 3 and Table 4). In the tetraploid data, 6 markers for late blight resistance were found in chromosomes III, V and IX while in the diploid dataset 14 markers on chromosomes 0, III, V, VI, IX and X were associated with the resistance

phenotype. Populations of *P. infestans* are diverse and there is a trend of increasing diversity in potato-growing regions worldwide (Cooke and Lees, 2004). Taking this into account, the markers on chromosome IX could indicate a QTL for broad resistance not specific to regional late blight strains, because the QTL was observed in data from trials in Peru and China, while markers associated to the QTL on chromosomes III, V and VI were found with the data of unique locations (Table 3 and Table 4). For example, the neighbouring SNPs on chromosome III, could be indicating a QTL particularly responsible for resistance against late blight strains specific to Holeta, Ethiopia. On the other hand, the marker on chromosome X found in the diploidized data could indicate a QTL for broad resistance having been observed in data collected in Peru as well as in China (Table 4). The highest number of SNPs associated to late blight resistance in the GWAS were mapped between 59 and 61.2 Mbp of chromosome IX. All mapped to the end of the long arm of the chromosome, in a region that had been previously associated with late blight resistance in Peru (Lindqvist-Kreuze et al., 2014). Additionally, the markers were within or surrounding the segment between 59.3 and 61.0 Mbp that forms a large cluster of putative resistance genes. For example, the locus PGSC0003DMG400020587 encodes a homolog of *Rpi-vnt1* (Mosquera et al., 2015), a major gene for resistance to *P. infestans* that has been previously cloned and characterized in the wild potato species *Solanum venturii* (Pel et al., 2009; Foster et al., 2009).

A few markers for late blight resistance QTL have been found in the past on chromosome III. These include a QTL tagged between the markers GP25 and CP6 (Gebhardt and Valkonen, 2001) which can be located near the locus PGSC0003DMB000000154 or approx. between 42 and 51 Mbp in the Phureja DM1-3 genome. The SNPs 3_45458723, 3_45458753 and 3_45458754 found in the diploidized dataset mapped close to this QTL. These markers are located within a WREBP-2 protein / transcription factor IIIA gene (PGSC0003DMG400009082). Marker 3_3319097 is localized within a gene encoding an iron

binding oxidoreductase (PGSC0003DMG40002252) that is located near an already known QTL tagged with marker T6135 (Gebhardt and Valkonen, 2001).

Several late blight resistance genes originated from *Solanum demissum* have been already mapped to specific positions of the genome. These include *R1* in chromosome V, *R3*, *R6* and *R7*, all in the distal segment of chromosome XI and *R2* in chromosome IV. Also, the genes *Rber* and *Rblc* belonging to other *Solanum* groups have been mapped to chromosomes X and VIII, respectively. These genes show resistance to contemporary races of *Phytophthora infestans*. Additionally, promising QTL are thought to be located on chromosomes III, IV, V and VI (Gebhardt and Valkonen 2001). The region with *R1* and QTL for late blight resistance in chromosome V is flanked by the markers GP21 and GP179 approximately between 2 and 5 Mbp in the DM genome containing the SNPs 5_4260524 and 5_5572873 in the tetraploid and the diploid dataset, respectively (Table 3, 4). The SNP 10_51544544 found on chromosome X in the diploid dataset and associated with resistance in three experiments (Table 4), is mapped to a region associated with late blight resistance conferred by gene *Rber*, tagged by the marker TG63 is located approximately at 52 Mbp in the DM genome.

The SNP 6_45694949 found on chromosome VI, mapped to a physical position around 45.7 Mbp, is located near a gene and two flanking QTL that have been recently associated with quantitative late blight resistance. Álvarez et al. (2017) used association mapping to identify SNPs in genes from a set of candidate genes in *Solanum tuberosum* group Phureja associated with quantitative resistance. The gene expresses a stem 28 kDa glycoprotein and is located around 49.1 Mbp between the Ib6a and Pin6b – lb6b QTL. The favourable allele of this SNP has a significant effect on the resistance in Yunnan in 2015 and 2016, and an additive effect since the individuals homozygous for the favourable allele are more resistant than the heterozygous individuals (Figure 6).

The 48x plex multiplexing of the samples during the sequencing and stringent filtering for minimum read depth in all samples yielded relatively few SNP (less than 4K), which is too few considering the level of LD decay-based estimate of 10s of thousands SNP to fully cover the genome. Therefore, the GWA was done using both tetraploid and the diploidized data. Three of the markers (9_58779951, 9_59967523, 9_60067335) map in chromosome 9 in the same chromosome region previously found associated with late blight resistance in Peru (Lindqvist-Kreuze et al., 2014; Li et al., 2010). These markers are physically separated in the DM genome by 1.3Mb, which fits the estimate for the LD decay in these potato genotypes. In the QTL dPI09c reported by Li et al. (2010) the R8 gene originating from *Solanum demissum* was recently identified (Jian Rui et al., 2018). The QTL dPI09c interval in potato DM1-3 516 R44 (Potato Genome Sequencing Consortium, 2011) begins at 60615044 bp, hence over 600Mbp away from the nearest GBS marker (9_60067335) we identified in the current research. In our tetraploid GBS marker set there are no SNP mapping in the dPI09c interval possibly because of the low sequencing depth and the complexity of the region that consists of several RXLR type resistance genes (Jiang Rui et al., 2018). In the diploid marker set, however, four markers (9_60067335, 9_61106174, 9_61108928, 9_61261167) that map in the QTL dPI09c interval were identified.

Conclusions

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Contributions:

Designed experiments and obtained funding: HLK, MB

Implemented field trials and collected data: MG, XL, JQ, GW, BH, ZP, QS, KN, IS

Analyzed and interpreted data: HLK, DG, BDB, PU

Bioinformatics: HLK, JDV, DG

Statistics: BDB

Wrote the manuscript: HLK, PU, MB

Literature

- Álvarez M.F., M. Angarita, M.C. Delgado, C. García, J. Jiménez-Gomez, C. Gebhardt and T. Mosquera. “Identification of Novel Associations of Candidate Genes with Resistance to Late Blight in *Solanum tuberosum* Group Phureja.” *Front. Plant Sci.* (2017) 8:1040.
- Bethke, P.C., Halterman, D.A. and Jansky, S., 2017. Are we getting better at using wild potato species in light of new tools?. *Crop Science*, 57(3), pp.1241-1258.
- Bonierbale, M. *Procedures for standard evaluation trials of advanced potato clones. An international cooperator's guide*. International Potato Center, 2007.
- Bradbury, P.J., Zhang, Z., Kroon, D.E., Casstevens, T.M., Ramdoss, Y., Buckler, E.S. (2007) TASSEL: Software for association mapping of complex traits in diverse samples. *Bioinformatics* 23:2633-2635.
- Bradshaw, J.E., Bryan, G.J. and Ramsay, G., 2006. Genetic resources (including wild and cultivated *Solanum* species) and progress in their utilisation in potato breeding. *Potato Research*, 49(1), pp.49-65.
- Catchen, J., Hohenlohe, P., Bassham, S., Amores, A., and Cresko, W. Stacks: an analysis tool set for population genomics. *Molecular Ecology*. 2013.
- Chao, J.F., Chao Huo, Zhechao Pan, Zhijian Zhao, and Qijun Sui. “Study and application of potato late blight monitoring and contro in southwest China”,in *Chinese potato essays 2017*, Harbin Cartographic Publishing House 456-461(in Chinese)

Clark, Lindsay V., Alexander E. Lipka, and Erik J. Sacks. "polyRAD: Genotype calling with uncertainty from sequencing data in polyploids and diploids." *G3: Genes, Genomes, Genetics* 9, no. 3 (2019): 663-673.

Clevenger, Josh, Carolina Chavarro, Stephanie A. Pearl, Peggy Ozias-Akins, and Scott A. Jackson. "Single nucleotide polymorphism identification in polyploids: a review, example, and recommendations." *Molecular plant* 8, no. 6 (2015): 831-846.

Cooke D.E.L. and A.K. Lees. "Markers, old and new, for examining *Phytophthora infestans* diversity." *Plant Pathology*. (2004) 53:6.

Foster SJ, Park TH, Pel M, Brigneti G, Śliwka J, Jagger L, van der Vossen E, Jones JD. Rpi-vnt1. 1, a Tm-22 homolog from *Solanum venturii*, confers resistance to potato late blight. *Molecular plant-microbe interactions*. 2009 May;22(5):589-600.

Gaiero, P., Speranza, P. and de Jong, H., 2018. Introgressive Hybridization in Potato Revealed by Novel Cytogenetic and Genomic Technologies. *American Journal of Potato Research*, 95(6), pp.607-621.

Gebhardt, C. and Valkonen, J.P., 2001. Organization of genes controlling disease resistance in the potato genome. *Annual review of phytopathology*, 39(1), pp.79-102.

Hamilton, John P., Candice N. Hansey, Brett R. Whitty, Kevin Stoffel, Alicia N. Massa, Allen Van Deynze, Walter S. De Jong, David S. Douches, and C. Robin Buell. "Single nucleotide

polymorphism discovery in elite North American potato germplasm." *BMC genomics* 12, no. 1 (2011): 302.

Hirut, Betaw G., Hussein A. Shimelis, Rob Melis, Mengistu Fentahun, and Walter De Jong. "Yield, Yield-related Traits and Response of Potato Clones to Late Blight Disease, in North-Western Highlands of Ethiopia." *Journal of Phytopathology* 165, no. 1 (2017): 1-14.

Hirut, B., Shimelis, H., Fentahun, M., Bonierbale, M., Gastelo, M. and Asfaw, A., 2017. Combining ability of highland tropic adapted potato for tuber yield and yield components under drought. *PloS one*, 12(7), p.e0181541.

Jiang Rui, Jingcai Li, Zhendong Tian, Juan Du, Miles Armstrong, Katie Baker, Joanne Tze-Yin Lim et al. "Potato late blight field resistance from QTL dPI09c is conferred by the NB-LRR gene R8." *Journal of experimental botany* 69, no. 7 (2018): 1545-1555.

Koenker, Roger, Stephen Portnoy, Pin Tian Ng, Achim Zeileis, Philip Grosjean, and Brian D. Ripley. "Package 'quantreg'." *Cran R-project. org* (2018).

Li H. and Durbin R. (2009) Fast and accurate short read alignment with Burrows-Wheeler Transform. *Bioinformatics*, 25:1754-60. [PMID: 19451168]

Li, Heng, Bob Handsaker, Alec Wysoker, Tim Fennell, Jue Ruan, Nils Homer, Gabor Marth, Goncalo Abecasis, and Richard Durbin. "The sequence alignment/map format and SAMtools." *Bioinformatics* 25, no. 16 (2009): 2078-2079.

Li, Jingcai, Hannele Lindqvist-Kreuze, Zhendong Tian, Jun Liu, Botao Song, Juan Landeo, Leticia Portal et al. "Conditional QTL underlying resistance to late blight in a diploid potato population." *Theoretical and applied genetics* 124, no. 7 (2012): 1339-1350.

Lindqvist-Kreuze, Hannele, Manuel Gastelo, Willmer Perez, Gregory A. Forbes, David de Koeper, and Merideth Bonierbale. "Phenotypic stability and genome-wide association study of late blight resistance in potato genotypes adapted to the tropical highlands." *Phytopathology* 104, no. 6 (2014): 624-633.

Lindqvist-Kreuze H, Gamboa S, Izarra M, Pérez W, Correa MY, Astete A, Särkinen T, Cueva M, Gonzáles P. Population structure and host range of the potato late blight pathogen *Phytophthora infestans* in Peru spanning two decades. *Plant Pathology*. 2020 Feb;69(2):334-46.

Matias, F., Meireles, K., Nagamatsu, S., Barrios, S., do Valle, C., Carazzolle, M., ... & Endelman, J. (2019). Expected Genotype Quality and Diploidized Marker Data from Genotyping-by-Sequencing of *Urochloa* spp. Tetraploids. bioRxiv, 525618.

Mihretu E, Mohammad W, Kassa B, Lindqvist-Kreuze H. Virulence Spectrum of *Phytophthora infestans* and Spatial Distribution of Physiological Races in Northwestern Ethiopia. *Ethiopian Journal of Agricultural Sciences*. 2020;30(1):69-85.

Muhinyuza, J.B., Shimelis, H., Melis, R. et al. Yield Response and Late Blight Reaction of Potato Genotypes in Rwanda. *Am. J. Potato Res.* 92, 10–22 (2015). <https://doi.org/10.1007/s12230-014-9406-8>

484 Ovchinnikova A, Krylova E, Gavrilenko T, Smekalova T, Zhuk M, Knapp S, Spooner DM.
 485 2011. Taxonomy of cultivated potatoes (Solanum section Petota: Solanaceae) Bot. J. Linn.
 486 Soc. 165: 107-155
 487
 488 Pel MA, Foster SJ, Park TH, Rietman H, van Arkel G, Jones JD, Van Eck HJ, Jacobsen E,
 489 Visser RG, Van der Vossen EA. Mapping and cloning of late blight resistance genes from
 490 Solanum venturii using an interspecific candidate gene approach. Molecular plant-microbe
 491 interactions. 2009 May;22(5):601-15.
 492
 493 Poplin, Ryan, Valentin Ruano-Rubio, Mark A. DePristo, Tim J. Fennell, Mauricio O. Carneiro,
 494 Geraldine A. Van der Auwera, David E. Kling et al. "Scaling accurate genetic variant discovery
 495 to tens of thousands of samples." *BioRxiv* (2017): 201178.
 496 Potato Genome Consortium. "Genome sequence and analysis of the tuber crop potato." *Nature*.
 497 (2011) 475: 189–195.
 498
 499 Rosyara, Umesh R., Walter S. De Jong, David S. Douches, and Jeffrey B. Endelman. "Software
 500 for genome-wide association studies in autopolyploids and its application to potato." *The plant*
 501 *genome* 9, no. 2 (2016).
 502
 503 Sharma, Sanjeev Kumar, Katrin MacKenzie, Karen McLean, Finlay Dale, Steve Daniels, and
 504 Glenn J. Bryan. "Linkage Disequilibrium and Evaluation of Genome-Wide Association
 505 Mapping Models in Tetraploid Potato." *G3: Genes, Genomes, Genetics* 8, no. 10 (2018): 3185-
 506 3202.
 507

508 Spooner DM, Ghislain M, Simon R, Jansky SH, Gavrilenko T. 2014. Systematics, diversity,
509 genetics, and evolution of wild and cultivated potatoes. *Botanical Review* 80: 283–383.
510

511 Sverrisdóttir, Elsa, Stephen Byrne, Ea Høegh Riis Sundmark, Heidi Øllegaard Johnsen, Hanne
512 Grethe Kirk, Torben Asp, Luc Janss, and Kåre L. Nielsen. "Genomic prediction of starch
513 content and chipping quality in tetraploid potato using genotyping-by-
514 sequencing." *Theoretical and Applied Genetics* 130, no. 10 (2017): 2091-2108.
515

516 Uitdewilligen, Jan GAML, Anne-Marie A. Wolters, B. Bjorn, Theo JA Borm, Richard GF
517 Visser, and Herman J. van Eck. "A next-generation sequencing method for genotyping-by-
518 sequencing of highly heterozygous autotetraploid potato." *PloS one* 8, no. 5 (2013): e62355.
519

520 Vos, Peter G., Jan GAML Uitdewilligen, Roeland E. Voorrips, Richard GF Visser, and Herman
521 J. van Eck. "Development and analysis of a 20K SNP array for potato (*Solanum tuberosum*):
522 an insight into the breeding history." *Theoretical and Applied Genetics* 128, no. 12 (2015):
523 2387-2401.
524

525 Vos, Peter G., M. João Paulo, Roeland E. Voorrips, Richard GF Visser, Herman J. van Eck,
526 and Fred A. van Eeuwijk. "Evaluation of LD decay and various LD-decay estimators in
527 simulated and SNP-array data of tetraploid potato." *Theoretical and Applied Genetics* 130, no.
528 1 (2017): 123-135.
529

530 Zheng X, Levine D, Shen J, Gogarten S, Laurie C, Weir B (2012). "A High-performance
531 Computing Toolset for Relatedness and Principal Component Analysis of SNP Data."
532 *Bioinformatics*, 28(24), 3326-3328. doi: 10.1093/bioinformatics/bts606.

Table 1. Contribution of CIP breeding populations to the TON diversity panel

Breeding population	Genotypes evaluated	main breeding objective
A	13	late blight resistance
B1	11	late blight resistance
B3	100	late blight resistance
B3-HT	37	late blight resistance, heat tolerance
B3-LTVR	25	Hybrid population combining late blight resistance, heat tolerance, virus resistance
LTVR	186	virus resistance, heat tolerance, drought tolerance, salinity tolerance
PREBRED	2	late blight resistance
VARIETY	6	varied
Grand Total	380	

Table 2. Description of the phenotypic evaluations involving the TON panel clones.

Country	Location	Year	number of genotypes evaluated (statistical design)	checks
Peru	Pasco, Oxapampa 10.5853°S, 75.4053°W	2014	241 (RCBD)	Chucmarina Unica Tomas Desiree
China	Yunnan, Kunming 24.8801°N, 102.8329°E	2015	306 (RCBD)	Chucmarina C-88
		2016	336 (RCBD)	Unica Desiree
Ethiopia	Oromia, Holetta 9.0633°N, 38.4902°E	2017	60 (RCBD)	Belete Gudene Unica
		2016	128 (Augmented)	Gudene Belete Unica Tomas

Table 3. Markers tagging QTL for late blight resistance with the general, additive and 1-dominance models of GWASpoly and tetraploid SNP.

Marker (chromosome followed by position)	Ref	Alt	Trait	Model	Threshold	Score	Effect
0_36073482	G	A	Oxa2014	general	4.71	8.64	NA
				additive	4.78	8.64	-0.18
				1-dom-alt	4.71	8.64	-0.18
			Yun2015	general	4.74	20.21	NA
				additive	4.78	20.21	-0.29
				1-dom-alt	4.71	20.21	-0.29
			Yun2016	general	4.75	19.37	NA
				additive	4.78	19.37	-0.25
				1-dom-alt	4.71	19.37	-0.25
3_3319097	A	T	Hol2017	general	4.49	5.83	NA
				1-dom-alt	4.71	6.05	0.43
5_4260524	A	T	Yun2016	1-dom-alt	4.71	5.01	0.14
9_58779951	G	A	Oxa2014	additive	4.78	4.84	-0.11
				1-dom-alt	4.71	5.52	-0.13
			Yun2015	general	4.74	4.94	NA
				1-dom-alt	4.71	5.92	-0.14
			Yun2016	1-dom-alt	4.71	4.77	-0.11
9_59967523	A	T	Yun2015	general	4.74	7.3	NA
				additive	4.78	6.94	-0.14
				1-dom-alt	4.71	8.18	-0.17
			Yun2016	general	4.75	7.76	NA
				additive	4.78	8.29	-0.13
				1-dom-alt	4.71	8.38	-0.15
9_60067335	A	G	Hol2016	1-dom-alt	4.71	4.81	-0.22
				1-dom-alt	4.71	4.81	-0.22
			Oxa2014	general	4.71	11.11	NA
				additive	4.78	11.64	-0.2
				1-dom-alt	4.71	12.1	-0.21
			Yun2015	general	4.74	19.39	NA
				additive	4.78	18.99	-0.26
				1-dom-alt	4.71	20.31	-0.28
			Yun2016	general	4.75	16.53	NA
				additive	4.78	17.26	-0.22
				1-dom-alt	4.71	17.59	-0.23

Table 4. Markers tagging QTL for late blight resistance with the diplo-general, diplo-additive and 1-dominance models of GWASpoly and diploid SNP.

Marker (chromosome followed by position)	Ref	Alt	Trait	Model	Threshold	Score	Effect
0_36073482	G	A	Oxa2014	diplo-general	6	10.27	NA
				diplo-additive	6.03	10.27	-0.2

			Yun2015	1-dom-alt	6.02	10.27	-0.2
				diplo-general	6.01	25.69	NA
				diplo-additive	6.03	25.69	-0.31
			Yun2016	1-dom-alt	6.02	25.69	-0.31
				diplo-general	6.02	25.69	NA
				diplo-additive	6.03	25.69	-0.27
3_45458723	A	C	Hol2017	1-dom-alt	6.02	25.69	-0.27
				diplo-general	5.94	25.69	NA
				diplo-additive	6.02	25.69	0.47
3_45458753	G	A	Hol2017	1-dom-alt	6.02	25.69	0.47
				diplo-additive	6.02	25.69	0.47
3_45458754	A	T	Hol2017	1-diplo-alt	6.02	25.69	0.47
				diplo-additive	6.02	25.69	0.47
5_5572873	G	A	Oxa2014	1-diplo-alt	6.02	25.69	0.17
				diplo-additive	6.03	25.69	0.17
6_45694949	G	A	Yun2015	1-dom-alt	6.02	25.69	-0.15
				diplo-general	6.01	25.69	NA
			Yun2016	1-dom-alt	6.02	25.69	-0.13
				diplo-general	6.02	25.69	NA
9_58779951	G	A	Oxa2014	1-dom-alt	6.02	25.69	-0.17
				diplo-additive	6.03	25.69	-0.17
				diplo-general	6	25.69	NA
			Yun2015	1-dom-alt	6.02	25.69	-0.18
				diplo-additive	6.03	25.69	-0.18
				diplo-general	6.01	25.69	NA
			Yun2016	1-dom-alt	6.02	25.69	-0.15
				diplo-additive	6.03	25.69	-0.15
				diplo-general	6.02	25.69	NA
9_59967523	A	T	Oxa2014	1-dom-alt	6.02	25.69	-0.14
				diplo-additive	6.03	25.69	-0.14
				diplo-general	6	25.69	NA
			Yun2015	1-dom-alt	6.02	25.69	-0.21
				diplo-additive	6.03	25.69	-0.21
				diplo-general	6.01	25.69	NA
			Yun2016	1-dom-alt	6.02	25.69	-0.19
				diplo-additive	6.03	25.69	-0.19
				diplo-general	6.02	25.69	NA
9_59997331	T	C	Oxa2014	1-dom-alt	6.02	25.69	-0.21
				diplo-additive	6.03	25.69	-0.21
				diplo-general	6	25.69	NA
			Yun2015	1-dom-alt	6.02	25.69	-0.25
				diplo-additive	6.03	25.69	-0.25
				diplo-general	6.01	25.69	NA
			Yun2016	1-dom-alt	6.02	25.69	-0.21
				diplo-additive	6.03	25.69	-0.21
				diplo-general	6.02	25.69	NA
9_60067335	A	G	Oxa2014	1-dom-alt	6.02	25.69	-0.23
				diplo-additive	6.03	25.69	-0.23
				diplo-general	6	25.69	NA
			Yun2015	1-dom-alt	6.02	25.69	-0.32
				diplo-additive	6.03	25.69	-0.32
				diplo-general	6.01	25.69	NA
			Yun2016	diplo-general	6.02	25.69	NA

				diplo-additive	6.03	25.69	-0.26
				1-dom-alt	6.02	25.69	-0.26
9_61106174	C	T	Yun2015	diplo-general	6.01	25.69	NA
				diplo-additive	6.03	25.69	-0.14
				1-dom-alt	6.02	25.69	-0.14
			Yun2016	diplo-general	6.02	25.69	NA
				diplo-additive	6.03	25.69	-0.14
				1-dom-alt	6.02	25.69	-0.14
9_61108928	T	G	Oxa2014	diplo-additive	6.03	25.69	-0.12
				1-dom-alt	6.02	25.69	-0.12
			Yun2015	diplo-general	6.01	25.69	NA
				diplo-additive	6.03	25.69	-0.17
				1-dom-alt	6.02	25.69	-0.17
			Yun2016	diplo-general	6.02	25.69	NA
				diplo-additive	6.03	25.69	-0.14
				1-dom-alt	6.02	25.69	-0.14
9_61261167	A	C	Oxa2014	diplo-general	6	25.69	NA
				diplo-additive	6.03	25.69	-0.21
				1-dom-alt	6.02	25.69	-0.23
			Yun2015	diplo-general	6.01	25.69	NA
				diplo-additive	6.03	25.69	-0.3
				1-dom-alt	6.02	25.69	-0.32
			Yun2016	diplo-general	6.02	25.69	NA
				diplo-additive	6.03	25.69	-0.25
				1-dom-alt	6.02	25.69	-0.26
10_51544544	A	G	Oxa2014	diplo-general	6	25.69	NA
				diplo-additive	6.03	25.69	-0.22
				1-dom-alt	6.02	25.69	-0.22
			Yun2015	diplo-general	6.01	25.69	NA
				diplo-additive	6.03	25.69	-0.3
				1-dom-alt	6.02	25.69	-0.3
			Yun2016	diplo-general	6.02	25.69	NA
				diplo-additive	6.03	25.69	-0.27
				1-dom-alt	6.02	25.69	-0.27

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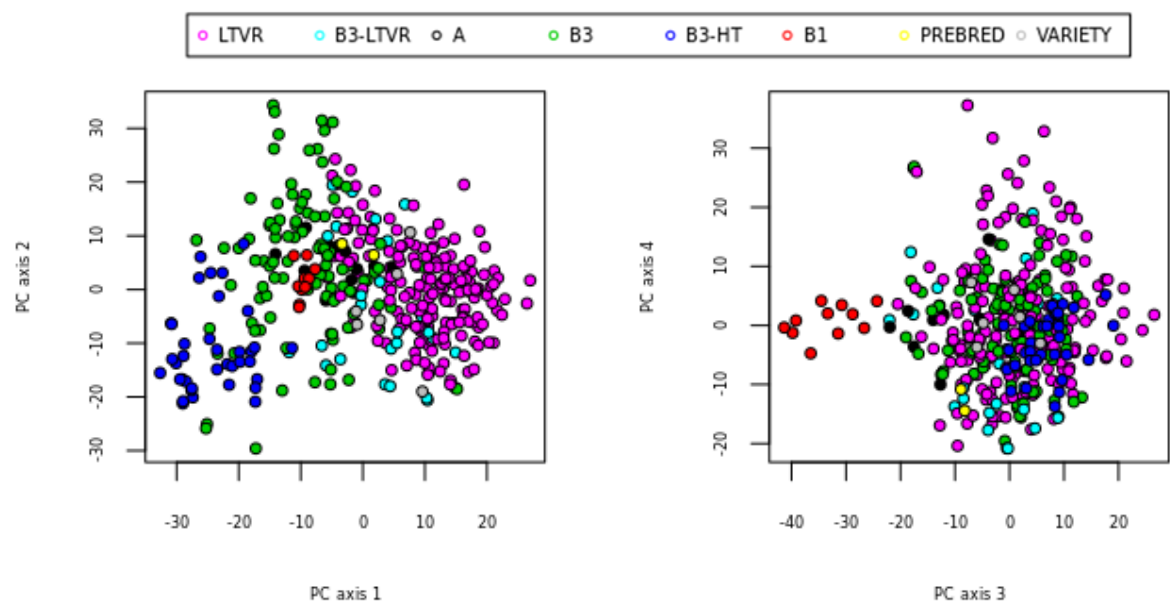
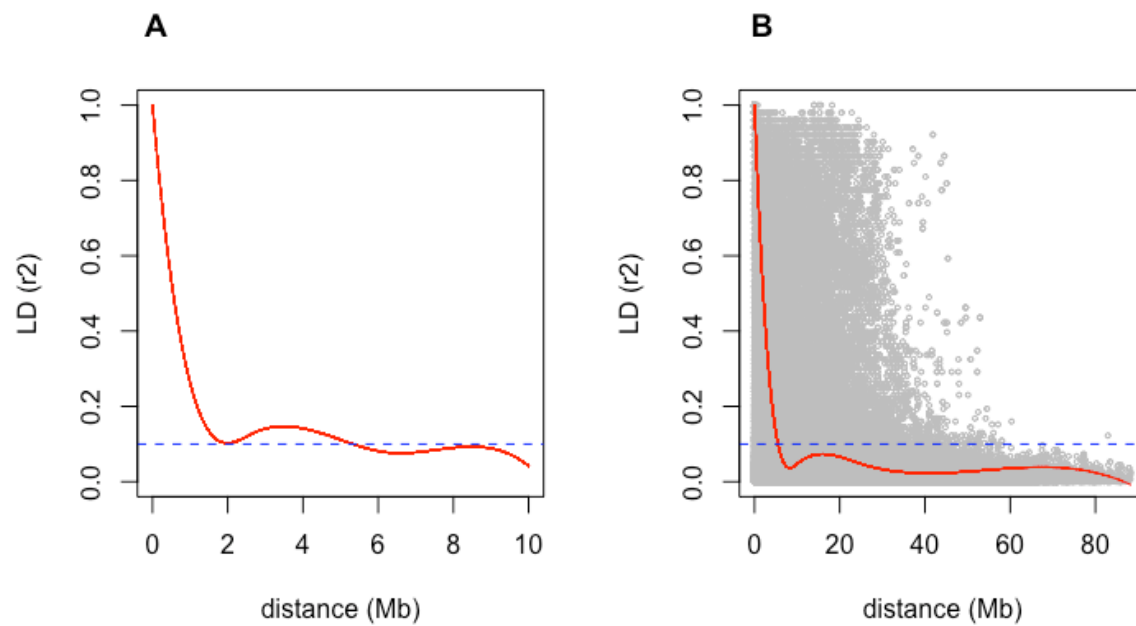


Figure 1. Population sub-structuring based on polyRAD estimation of genotype probabilities



554 **Figure 2.** Linkage disequilibrium (LD) estimated in the TON panel based on Pearson
555 correlation coefficient (r^2) plotted against the physical map distance (Mb) between pairs of
556 SNP.

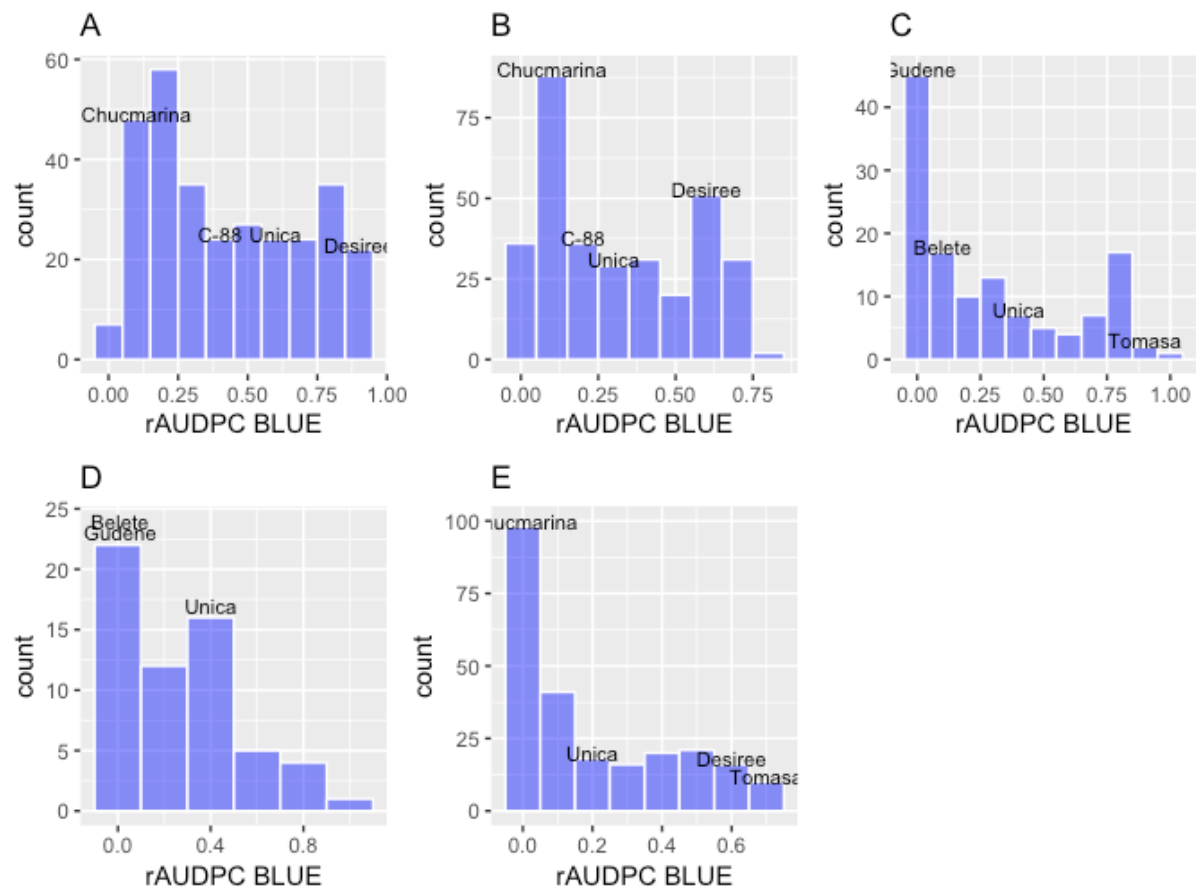


Figure 3. Histograms of rAUDPC values in Kunming, China in 2015(A) and in 2016 (B), Holeta, Ethiopia in 2016 (C) and 2017 (D), and Oxapampa, Peru at 2014 (E). The control genotypes (checks) in each trial are indicated in the plots based on their rAUDPC value.

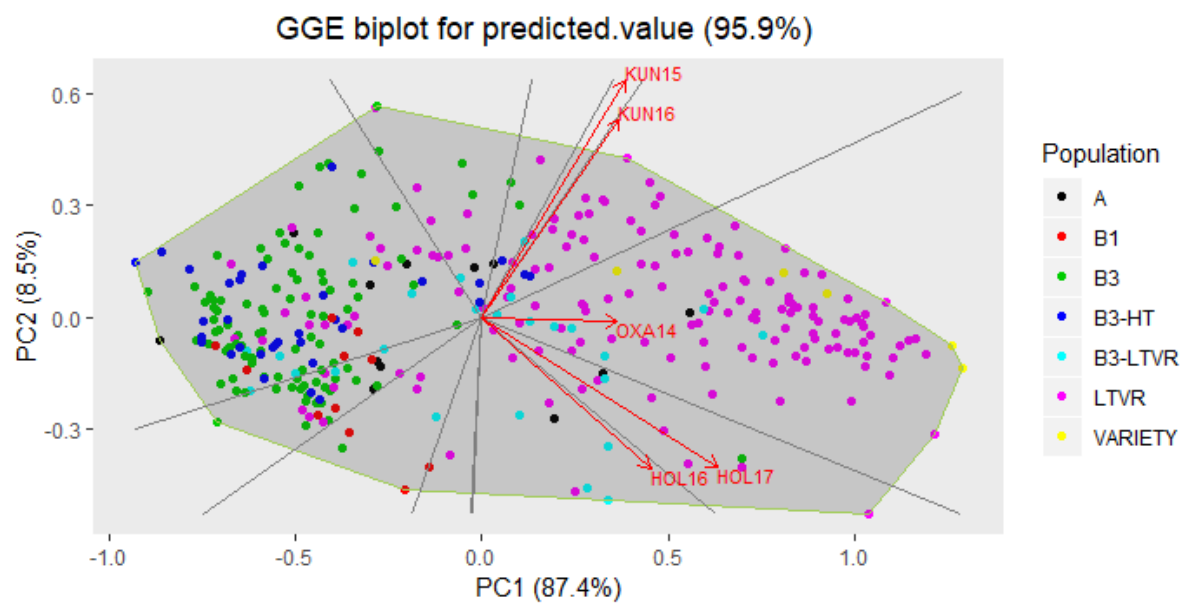


Figure 4. GGE biplot for predicted performance (based on BLUP and genetic kinship matrix) of the test genotypes and their parents in Kunming 2015 (KUN15) and 2016 (KUN16), Oxapampa 2014 (OXA14), Holetta 2016 (HOL16) and 2017 (HOL17).

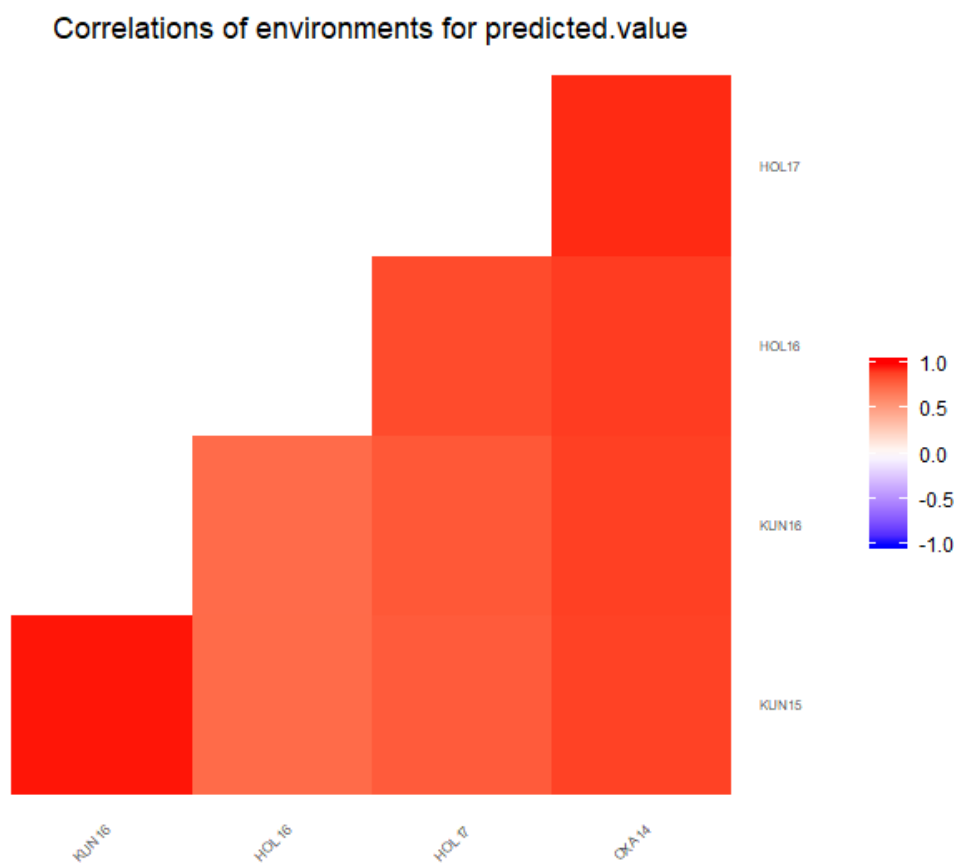


Figure 5. Heat-plot on the correlations among the different trials.

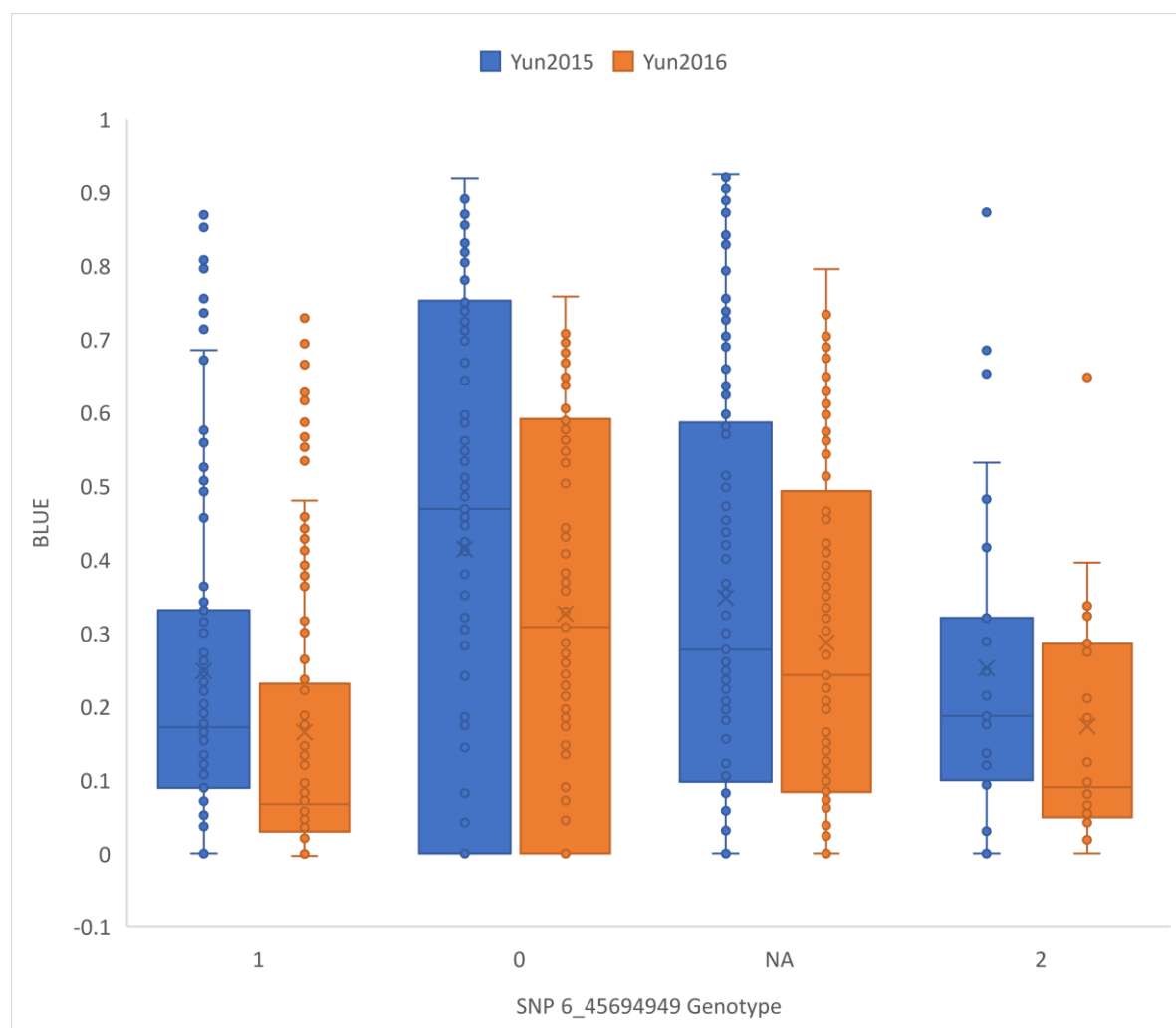


Figure 6. Boxplot for BLUE value distribution in the different genotype classes for the SNP 6_45694949 found in the diploidized dataset. “0” and “2” stand for homozygous for the reference and alternative alleles respectively. “1” indicates heterozygote and “NA”, missing genotype. Lower values indicate a higher resistance.

Table S1. Population denominations, and parentage of the potato genotypes evaluated in this study.

population	CIP code	female parent	male parent
A	CIP384866.5	376724.1=(85LB70.5)	BULK PRECOZ
A	CIP381379.12	378356.895	PRECOZ BULK
A	CIP381381.9	378493.915	PRECOZ BULK
A	CIP381381.13	378493.915	PRECOZ BULK
A	CIP381403.16	378507.833	BULK
A	CIP381178.14	378943.565	PHY BULK
A	CIP384321.3	380479.15	BULK 3
A	CIP391691.96	381381.9	LB-CUZ.1
A	CIP387224.11	382121.25	676008=(I-1039)
A	CIP374080.5	801013=(MEX 72 =I-1058)	700764=(Casa Blanca EE-2010)
A	CIP380011.12	GRETA	SEEDLINGS 79 BULK
A	CIP380496.6	INDIA-1058 B	XY BULK
A	CIP377744.1	M-1266-14 MEX	374035.1
B1	CIP399053.15	395230.1	395322.11
B1	CIP399067.22	395257.2	395271.6
B1	CIP399075.32	395266.2=(B1C4046.2)	395282.3=(B1C4062.3)
B1	CIP399075.7	395266.2=(B1C4046.2)	395282.3=(B1C4062.3)
B1	CIP399078.11	395266.3=(B1C4046.3)	395260.8=(B1C4040.8)
B1	CIP399048.24	395272.2	395257.6
B1	CIP399079.22	395274.1	395257.6
B1	CIP399085.17	395296.2=(B1C4076.2)	395256.1=(B1C4036.1)
B1	CIP399085.30	395296.2=(B1C4076.2)	395256.1=(B1C4036.1)
B1	CIP399083.4	395296.2=(B1C4076.2)	395247.1=(B1C4027.1)
B1	CIP399085.23	395296.2=(B1C4076.2)	395256.1=(B1C4036.1)

B3	CIP389746.2	381379.9	386614.16=(XY.16)
B3	CIP393220.54	381400.22	387170.9
B3	CIP387164.4	382171.1	575049=(CEW-69-1)
B3	CIP391046.14	386209.1	387338.3
B3	CIP391047.34	386209.1	387338.3
B3	CIP393228.67	386209.1	387170.9
B3	CIP391002.6	386209.1	386206.4
B3	CIP393227.66	386209.1	381400.22
B3	CIP391583.25	386209.15	387170.9
B3	CIP392617.54	387002.11	387170.9
B3	CIP393248.55	387002.11	386614.16=(XY.16)
B3	CIP393242.50	387002.11	381400.22
B3	CIP391580.30	387002.2	387214.9
B3	CIP393079.4	387004.13	390357.4
B3	CIP393079.24	387004.13	390357.4
B3	CIP391004.18	387004.4	386206.4
B3	CIP393284.39	387015.12	387170.9
B3	CIP393073.179	387015.13	389746.2
B3	CIP393073.197	387015.13	389746.2
B3	CIP393280.82	387015.3	386316.14=(XY.14)
B3	CIP393280.64	387015.3	386316.14=(XY.14)
B3	CIP393280.57	387015.3	386316.14=(XY.14)
B3	CIP391011.17	387041.12	386206.4
B3	CIP391585.179	387132.2	387170.9
B3	CIP391585.5	387132.2	387170.9
B3	CIP392633.64	387132.2	387334.5
B3	CIP392634.49	387136.14	387170.9
B3	CIP392634.52	387136.14	387170.9
B3	CIP392637.10	387143.22	387170.9

B3	CIP392637.27	387143.22	387170.9
B3	CIP392639.34	387143.22	387334.5
B3	CIP393339.242	387164.4	SANI IMILLA
B3	CIP393371.157	387170.16	389746.2
B3	CIP393371.58	387170.16	389746.2
B3	CIP393371.164	387170.16	389746.2
B3	CIP393371.159	387170.16	389746.2
B3	CIP391058.175	387170.16	387338.3
B3	CIP393349.68	387170.6	387338.3
B3	CIP392650.12	387181.7	387170.9
B3	CIP393382.44	387205.5	387338.3
B3	CIP393385.47	387231.7	387170.9
B3	CIP393385.39	387231.7	387170.9
B3	CIP393399.7	387303.71	387338.3
B3	CIP393075.54	387315.27	389746.2
B3	CIP393083.2	387315.27	390357.4
B3	CIP393084.31	387326.27	390357.4
B3	CIP392657.171	387341.1	387170.9
B3	CIP392657.8	387341.1	387170.9
B3	CIP393077.159	387348.2	389746.2
B3	CIP391065.81	387348.2	387338.3
B3	CIP393077.54	387348.2	389746.2
B3	CIP393085.5	387348.2	390357.4
B3	CIP391065.69	387348.2	387338.3
B3	CIP396008.104	391002.15	393382.64
B3	CIP396004.263	391002.6	393382.64
B3	CIP396004.225	391002.6	393382.64
B3	CIP396004.337	391002.6	393382.64
B3	CIP396012.266	391004.1	393280.58

B3	CIP396009.240	391004.4	393280.58
B3	CIP396009.258	391004.4	393280.58
B3	CIP395037.107	391004.4	391679.12
B3	CIP396018.241	391046.14	393280.58
B3	CIP396023.109	391047.34	393280.57
B3	CIP396244.12	391580.3	392633.1
B3	CIP395077.12	391586.109	393053.6
B3	CIP395109.29	391589.26	393079.4
B3	CIP395109.34	391589.26	393079.4
B3	CIP395112.19	391686.15	393079.4
B3	CIP395112.32	391686.15	393079.4
B3	CIP395112.6	391686.15	393079.4
B3	CIP395112.36	391686.15	393079.4
B3	CIP395111.13	391686.5	393079.4
B3	CIP396027.205	392633.23	393382.64
B3	CIP396026.101	392633.4	393280.64
B3	CIP396026.103	392633.4	393280.64
B3	CIP395084.9	392633.6	393053.6
B3	CIP396031.119	392633.64	393382.64
B3	CIP396031.108	392633.64	393382.64
B3	CIP396241.4	392634.52	392626.9
B3	CIP396033.102	392639.53	393382.64
B3	CIP395169.17	392652.8	391679.12
B3	CIP396034.268	393042.5	393280.64
B3	CIP396034.103	393042.5	393280.64
B3	CIP395123.6	393046.7	393079.4
B3	CIP396036.201	393077.51	393382.64
B3	CIP396038.101	393077.54	393280.64
B3	CIP396037.215	393077.54	393382.64

B3	CIP396038.107	393077.54	393280.64
B3	CIP396038.105	393077.54	393280.64
B3	CIP395015.6	393083.2	391679.12
B3	CIP395017.14	393085.13	392639.8
B3	CIP395017.229	393085.13	392639.8
B3	CIP395017.242	393085.13	392639.8
B3	CIP395017.227	393085.13	392639.8
B3	CIP395011.2	393085.5	392639.8
B3	CIP395096.2	393085.5	393053.6
B3	CIP396240.2	393371.58	391679.12
B3	CIP396240.23	393371.58	391679.12
B3	CIP396043.226	393401.55	393280.57
B3	CIP396046.105	TX.Y.4	393280.64
B3-HT	CIP398180.612	392657.171	392633.64
B3-HT	CIP398180.289	392657.171	392633.64
B3-HT	CIP398180.292	392657.171	392633.64
B3-HT	CIP398180.253	392657.171	392633.64
B3-HT	CIP398180.144	392657.171	392633.64
B3-HT	CIP398193.650	393077.54	392633.64
B3-HT	CIP398192.213	393077.54	392633.54
B3-HT	CIP398190.735	393077.54	392639.2
B3-HT	CIP398190.112	393077.54	392639.2
B3-HT	CIP398192.41	393077.54	392633.54
B3-HT	CIP398192.592	393077.54	392633.54
B3-HT	CIP398190.571	393077.54	392639.2
B3-HT	CIP398190.615	393077.54	392639.2
B3-HT	CIP398190.404	393077.54	392639.2
B3-HT	CIP398190.530	393077.54	392639.2
B3-HT	CIP398193.553	393077.54	392633.64

B3-HT	CIP398193.158	393077.54	392633.64
B3-HT	CIP398190.605	393077.54	392639.2
B3-HT	CIP398192.553	393077.54	392633.54
B3-HT	CIP398190.200	393077.54	392639.2
B3-HT	CIP398190.523	393077.54	392639.2
B3-HT	CIP398201.510	393242.5	392633.64
B3-HT	CIP398203.509	393280.82	392633.64
B3-HT	CIP398098.65	393371.58	392639.31
B3-HT	CIP398208.58	393371.58	392633.64
B3-HT	CIP398208.33	393371.58	392633.64
B3-HT	CIP398098.205	393371.58	392639.31
B3-HT	CIP398208.219	393371.58	392633.64
B3-HT	CIP398208.670	393371.58	392633.64
B3-HT	CIP398098.231	393371.58	392639.31
B3-HT	CIP398098.203	393371.58	392639.31
B3-HT	CIP398098.570	393371.58	392639.31
B3-HT	CIP398208.704	393371.58	392633.64
B3-HT	CIP398098.119	393371.58	392639.31
B3-HT	CIP398208.29	393371.58	392633.64
B3-HT	CIP398208.505	393371.58	392633.64
B3-HT	CIP398208.620	393371.58	392633.64
B3-LTVR	CIP301056.54	385205.5	393613.2=(TXY.2)
B3-LTVR	CIP301037.85	387205.5	702853=(LOP-868)
B3-LTVR	CIP301045.74	387205.5	391207.2=(LR93.050)
B3-LTVR	CIP301024.14	388615.22=(C91.640)	387170.9
B3-LTVR	CIP301024.95	388615.22=(C91.640)	387170.9
B3-LTVR	CIP301026.23	389746.2	BOGNA
B3-LTVR	CIP301041.26	389746.2	LOP-886
B3-LTVR	CIP301055.53	389746.2	393617.1=(TXY.11)

B3-LTVR	CIP301023.15	391180.6=(C90.266)	387170.9
B3-LTVR	CIP301044.36	392025.7=(LR93.221)	LOP-886
B3-LTVR	CIP396063.1	392633.1	TXY.12
B3-LTVR	CIP396063.16	392633.1	TXY.12
B3-LTVR	CIP396180.22	392633.6	393615.6=(TXY.6)
B3-LTVR	CIP396268.9	392639.34	393613.2=(TXY.2)
B3-LTVR	CIP396272.18	392639.34	TXY.12
B3-LTVR	CIP396268.1	392639.34	393613.2=(TXY.2)
B3-LTVR	CIP396272.21	392639.34	TXY.12
B3-LTVR	CIP396272.12	392639.34	TXY.12
B3-LTVR	CIP396272.2	392639.34	TXY.12
B3-LTVR	CIP396272.37	392639.34	TXY.12
B3-LTVR	CIP396273.48	393220.54	TXY.12
B3-LTVR	CIP396269.16	393371.58	393613.2=(TXY.2)
B3-LTVR	CIP396269.14	393371.58	393613.2=(TXY.2)
B3-LTVR	CIP301029.18	C97.255	C95.397
B3-LTVR	CIP301040.63	UNICA	702853=(LOP-868)
LTVR	CIP394899.5	28.68	C90.205
LTVR	CIP394898.13	28.68	BWH-87.344R
LTVR	CIP385558.2	32) 2	NT 91.002
LTVR	CIP394901.2	34.73	393617.1=(TXY.11)
LTVR	CIP394900.1	34.73	BWH-87.344R
LTVR	CIP392285.72	36.14	382157.3
LTVR	CIP379706.27	377257.1=(LT-1)	PVX + PVY BULK
LTVR	CIP388676.1	378015.18	PVY-BK
LTVR	CIP385561.124	38) 8	ML 91.007
LTVR	CIP391180.6	385305.1=(XY.9)	378017.2=(LT-7)
LTVR	CIP388972.22	386316.1=(XY.20)	377964.5

LTVR	CIP397079.6	386768.10=(MARIA TAMBEÃ'A)	392820.1=(C93.154)
LTVR	CIP397079.26	386768.10=(MARIA TAMBEÃ'A)	392820.1=(C93.154)
LTVR	CIP392797.22	387521.3	APHRODITE
LTVR	CIP303381.30	388611.22=(C91.612)	676008=(I-1039)
LTVR	CIP395434.1	388611.22=(C91.612)	N93.067
LTVR	CIP394600.52	388611.22=(C91.612)	388972.22=(C89.315)
LTVR	CIP395192.1	388611.22=(C91.612)	C92.044
LTVR	CIP395195.7	388611.22=(C91.612)	C92.167
LTVR	CIP397044.25	388611.22=(C91.612)	391180.6=(C90.266)
LTVR	CIP395193.6	388611.22=(C91.612)	C92.030
LTVR	CIP303381.106	388611.22=(C91.612)	676008=(I-1039)
LTVR	CIP397197.9	388615.22=(C91.640)	388972.22
LTVR	CIP304345.102	388615.22=(C91.640)	676008=(I-1039)
LTVR	CIP395432.51	388615.22=(C91.640)	C92.030
LTVR	CIP397039.53	388615.22=(C91.640)	388972.22=(C89.315)
LTVR	CIP395436.8	388615.22=(C91.640)	388615.22=(C91.640)
LTVR	CIP397039.51	388615.22=(C91.640)	388972.22=(C89.315)
LTVR	CIP392759.1	388676.1=(Y84.027)	PENTLAND CROWN
LTVR	CIP397006.18	389468.3=(92.119)	88.052
LTVR	CIP397067.2	390663.8=(C91.628)	392820.1=(C93.154)
LTVR	CIP300101.11	390674.33=(95.303)	387170.9
LTVR	CIP397065.2	391180.6=(C90.266)	392820.1=(C93.154)
LTVR	CIP397065.28	391180.6=(C90.266)	392820.1=(C93.154)
LTVR	CIP399101.1	391213. 1	388972.22
LTVR	CIP300066.11	391382.18=(95.108)	392820.1=(C93.154)
LTVR	CIP300065.4	391382.18=(95.108)	387170.9
LTVR	CIP397098.12	391533.1=(LR93.060)	391207.2=(LR93.050)

LTVR	CIP397012.20	391846.5=(LR93.309)	88.052
LTVR	CIP397012.22	391846.5=(LR93.309)	88.052
LTVR	CIP397078.12	391846.5=(LR93.309)	392820.1=(C93.154)
LTVR	CIP393617.1	391896.15=(DXY.15)	DXY.33
LTVR	CIP393613.2	391896.15=(DXY.15)	391894.7=(DXY.7)
LTVR	CIP396311.1	391925.2	C92.030
LTVR	CIP397036.7	392011.1=(LR93.160)	392745.7=(92.187)
LTVR	CIP397077.16	392025.7=(LR93.221)	392820.1=(C93.154)
LTVR	CIP397014.2	392739.4=(92.062)	88.108
LTVR	CIP397060.19	392739.4=(92.062)	392820.1=(C93.154)
LTVR	CIP397196.8	392797.22	388611.22=(C91.612)
LTVR	CIP397196.3	392797.22	388611.22=(C91.612)
LTVR	CIP397069.11	392797.22=(C92.140)	392820.1=(C93.154)
LTVR	CIP397069.5	392797.22=(C92.140)	392820.1=(C93.154)
LTVR	CIP304347.6	392820.1=(C93.154)	676008=(I-1039)
LTVR	CIP397099.4	392822.3=(LR93.073)	391207.2=(LR93.050)
LTVR	CIP397099.6	392822.3=(LR93.073)	391207.2=(LR93.050)
LTVR	CIP397073.15	392823.4=(LR93.120)	392820.1=(C93.154)
LTVR	CIP397073.7	392823.4=(LR93.120)	392820.1=(C93.154)
LTVR	CIP397100.9	392823.4=(LR93.120)	391207.2=(LR93.050)
LTVR	CIP397073.16	392823.4=(LR93.120)	392820.1=(C93.154)
LTVR	CIP304366.46	392823.4=(LR93.120)	676008=(I-1039)
LTVR	CIP397035.26	392823.4=(LR93.120)	92.187
LTVR	CIP300048.12	392973.48=(95.048)	392820.1=(C93.154)
LTVR	CIP300046.22	392973.48=(95.048)	393613.2=(TXY.2)
LTVR	CIP300099.22	393533.2=(95.302)	392820.1=(C93.154)
LTVR	CIP300063.9	393536.13=(95.103)	392820.1=(C93.154)
LTVR	CIP300063.4	393536.13=(95.103)	392820.1=(C93.154)
LTVR	CIP396285.1	393617.1=(TXY.11)	104.12 LB

LTVR	CIP395448.1	393617.1=(TXY.11)	BWH-87.344R
LTVR	CIP385499.11	65-ZA-5	377964.5
LTVR	CIP391919.3	69.4 (1043) BW	-
LTVR	CIP392780.1	703364=(SEDAFIN)	YY.3
LTVR	CIP389468.3	720087=(SERRANA)	388216.1=(YY.5)
LTVR	CIP390478.9	720087=(SERRANA)	386287.1=(XY.4)
LTVR	CIP390663.8	720087=(SERRANA)	386316.14=(XY.14)
LTVR	CIP388611.22	720091=(MEX-32)	385305.1=(XY.9)
LTVR	CIP394904.20	720118.1=(37-35A)	C90.205
LTVR	CIP302498.70	720139=(YAGANA-INIA)	391180.6=(C90.266)
LTVR	CIP302499.30	720139=(YAGANA-INIA)	392820.1=(C93.154)
LTVR	CIP394611.112	780280=(PW-88-6203)	676008=(I-1039)
LTVR	CIP304383.41	800824=(RED PONTIAC)	92.187
LTVR	CIP304383.80	800824=(RED PONTIAC)	92.187
LTVR	CIP391724.1	800959=(GRANOLA)	386316.1=(XY.20)
LTVR	CIP391207.2	800959=(GRANOLA)	385305.1=(XY.9)
LTVR	CIP392739.4	86001	386614.16=(XY.16)
LTVR	CIP392740.4	87055	386614.16=(XY.16)
LTVR	CIP397054.3	87059	392820.1=(C93.154)
LTVR	CIP397055.2	88052	392820.1=(C93.154)
LTVR	CIP392745.7	88078	386316.1=(XY.20)
LTVR	CIP397029.21	92.118	92.187
LTVR	CIP397016.7	92.119	88.108
LTVR	CIP397030.31	93.003	92.187
LTVR	CIP300054.29	95.059	392820.1=(C93.154)
LTVR	CIP300056.33	95.071	387170.9
LTVR	CIP300055.32	95.071	393613.2=(TXY.2)
LTVR	CIP300072.1	95.139	392820.1=(C93.154)
LTVR	CIP300137.31	95.187	387170.9

LTVR	CIP300093.14	95.206	392820.1=(C93.154)
LTVR	CIP388615.22	B-71-240.2	386614.16=(XY.16)
LTVR	CIP392781.1	B71-74-49.12	385280.1=(XY.13)
LTVR	CIP394034.65	B79.638.1	676008=(I-1039)
LTVR	CIP394034.7	B79.638.1	676008=(I-1039)
LTVR	CIP394881.8	B84-606.5	386287.1=(XY.4)
LTVR	CIP393536.13	BEROLINA	386287.1=(XY.4)
LTVR	CIP394895.7	BWH-87.230R	C90.205
LTVR	CIP391930.1	BWH-87.338	SELF
LTVR	CIP395438.1	BWH-87.344R	393617.1=(TXY.11)
LTVR	CIP395445.16	BWH-87.415	391894.7=(DXY.7)
LTVR	CIP394906.6	BWH-87.420	C90.205
LTVR	CIP395446.1	BWH-87.446R	393613.2=(TXY.2)
LTVR	CIP395186.6	C91.902	C92.032
LTVR	CIP395197.5	C91.921	BK-RKN-3
LTVR	CIP398014.2	C91.923	N93.107
LTVR	CIP395194.9	C93.059	C93.030
LTVR	CIP304350.78	CHIEFTAIN	392820.1=(C93.154)
LTVR	CIP304350.95	CHIEFTAIN	392820.1=(C93.154)
LTVR	CIP304350.100	CHIEFTAIN	392820.1=(C93.154)
LTVR	CIP304349.8	CHIEFTAIN	92.187
LTVR	CIP304350.18	CHIEFTAIN	392820.1=(C93.154)
LTVR	CIP304351.109	CHIEFTAIN	676008=(I-1039)
LTVR	CIP304351.31	CHIEFTAIN	676008=(I-1039)
LTVR	CIP304350.118	CHIEFTAIN	392820.1=(C93.154)
LTVR	CIP393615.6	DXY.33	391896.15=(DXY.15)
LTVR	CIP395196.4	ES-92.005	BK-RKN-1
LTVR	CIP391533.1	G-7445	385280.1=(XY.13)
LTVR	CIP394579.36	KONDOR	393615.6=(TXY.6)

LTVR	CIP392973.48	KRASA	385280.1=(XY.13)
LTVR	CIP392025.7	LINEA 21	386614.16=(XY.16)
LTVR	CIP392032.2	LOTOS	385280.1=(XY.13)
LTVR	CIP392822.3	MARIELA	YY.1
LTVR	CIP302428.20	MARIELA	392745.7=(92.187)
LTVR	CIP391382.18	MARIELA	386287.1=(XY.4)
LTVR	CIP304369.22	MARIELA	676008=(I-1039)
LTVR	CIP300135.14	MARIVA	392820.1=(C93.154)
LTVR	CIP300135.3	MARIVA	392820.1=(C93.154)
LTVR	CIP304371.67	MONALISA	92.187
LTVR	CIP392820.1	MONALISA	388216.1=(YY.5)
LTVR	CIP304371.20	MONALISA	92.187
LTVR	CIP304371.58	MONALISA	92.187
LTVR	CIP392821.1	PW-31	385280.1=(XY.13)
LTVR	CIP390637.1	PW-31	385305.1=(XY.9)
LTVR	CIP393708.31	PW-31	391895.10=(DXY.10)
LTVR	CIP304387.39	REINHORT	92.187
LTVR	CIP304387.92	REINHORT	92.187
LTVR	CIP304387.17	REINHORT	92.187
LTVR	CIP304394.56	SHEPODY	391207.2=(LR93.050)
LTVR	CIP304399.5	SNOWDEN	92.187
LTVR	CIP304399.15	SNOWDEN	92.187
LTVR	CIP391931.1	SR-17.50	SELF
LTVR	CIP302476.108	TITIA	392745.7=(92.187)
LTVR	CIP394613.139	TXY.4	676008=(I-1039)
LTVR	CIP394613.32	TXY.4	676008=(I-1039)
LTVR	CIP394614.117	TXY.8	676008=(I-1039)
LTVR	CIP394638.3	TXY.8	TITIA
LTVR	CIP396287.5	TXY.8	387170.9

LTVR	CIP304405.47	WA.018	676008=(I-1039)
LTVR	CIP304405.42	WA.018	676008=(I-1039)
LTVR	CIP304406.31	WA.077	676008=(I-1039)
LTVR	CIP394223.9	XY.13	C-282LM87B
LTVR	CIP394223.19	XY.13	C-282LM87B
LTVR	CIP302476.19	TITIA	392745.7=(92.187)
LTVR	CIP304330.34	391382.18=(95.108)	676008=(I-1039)
LTVR	CIP304345.47	388615.22=(C91.640)	676008=(I-1039)
LTVR	CIP304349.110	CHIEFTAIN	92.187
LTVR	CIP304349.4	CHIEFTAIN	92.187
LTVR	CIP304351.15	CHIEFTAIN	676008=(I-1039)
LTVR	CIP304351.9	CHIEFTAIN	676008=(I-1039)
LTVR	CIP309003.11	388611.22	304387.17
LTVR	CIP309017.101	395438.1	801088
LTVR	CIP309024.1	397036.7	392820.1
LTVR	CIP309026.72	397036.7	801088
LTVR	CIP309028.32	397036.7	801152
LTVR	CIP309062.106	303381.106	302499.24
LTVR	CIP309064.42	303381.30	392797.22
LTVR	CIP309064.76	303381.30	392797.22
LTVR	CIP309074.123	304330.34	392745.7
LTVR	CIP309078.56	304330.34	304356.32
LTVR	CIP309088.120	304347.6	302499.24
LTVR	CIP309093.50	304349.25	392820.1
LTVR	CIP309103.85	304349.8	801152
LTVR	CIP309128.87	304368.46	304356.32
LTVR	CIP309129.11	304368.46	304371.19
LTVR	CIP309131.16	304387.31	392820.1
LTVR	CIP309137.95	800258	396311.1

LTVR	CIP380389.1	BL-1.2	MURILLO III-80
LTVR	CIP720043	NARANJA	(KATAHDIN x MANTARO)
LTVR	CIP720088	MPI 61.375/23	B 25.65=(Atleet x Huinkul MAG)
PREBRED	CIP694474.16	4x-84.1	2x-5.26
PREBRED	CIP694474.33	4x-84.1	2x-5.26
VARIETY-Tomasa	CIP720072	(B 606.37 X KATAHDIN)	(RENACIMIENTO x YANA IMILLA)
VARIETY-Kufri Jyoti	CIP800258	3069D (4)	2814A (1)
VARIETY-Atlantic	CIP800827	800823=(WAUSEON)	B-5141.6
VARIETY-Spunta	CIP800923	BEA	USDA X 96.56
VARIETY-Desiree	CIP800048	URGENTA	DEPESCHE
VARIETY-DTO-33	CIP800174	WISC 639	W5295.7

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