IFAPA Instituto de Investigación y Formación Agraria y Pesquera

IHSM La Mayora Instituto de Hortofruticultura Subtropical y Mediterránea

UNIVERSIDAD DE MÁLAGA

CSIC Consejo Superior de Investigaciones Científicas

Breeding Value

Pre-breeding strategies for obtaining new resilient and added value berries

The BreedingValue low-density array for MAS in strawberry

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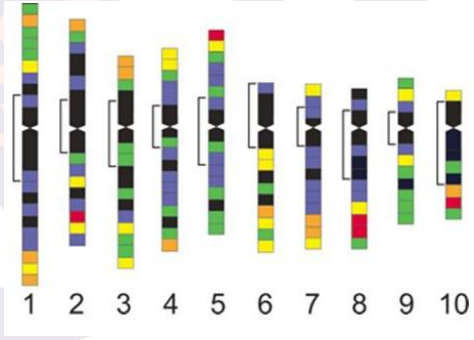
The BreedingValue project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101000747.

BREEDINGVALUE Task 2.2: Investigate the applicability of known molecular markers in Strawberry breeding.

OBJECTIVES

- Development of a 96-SNP Array for Strawberry using markers linked to trait of interest.
- Evaluation of marker polymorphism.
- Evaluation for diversity analyses and pedigree testing.
- Evaluation of Marker/trait associations.



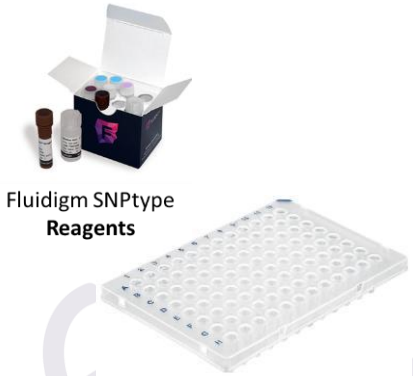
Genotype: Sequence of all the genes in a genome

Selection using DNA markers is faster than selecting based in phenotype

Phenotype: Physical expression of traits

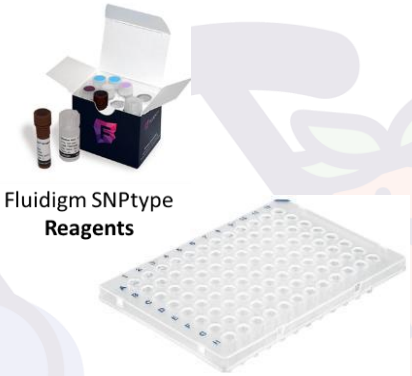
Environment

Selection of markers for the Low-density SNP array for Marker-Assisted Selection (MAS)



FLUIDIGM ARRAY_v1.0

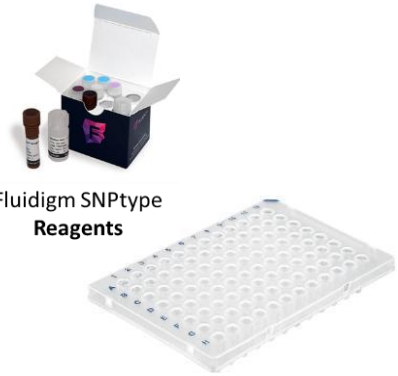
Poor resolution of genotypic classes in ~15% of markers



FLUIDIGM ARRAY_v2.0

PCR conditions modified for 9 assays

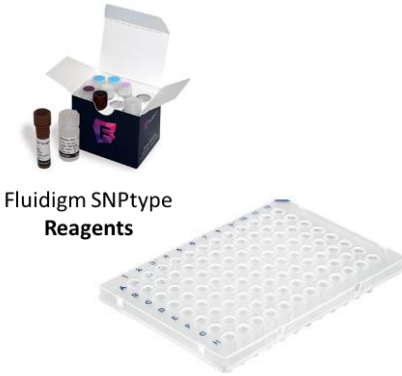
15 probes substituted:
6 redesigned
9 new probes



FLUIDIGM ARRAY_v3.0

Array evaluated in 94 additional strawberry samples

4 probes substituted



FLUIDIGM ARRAY_v4.0

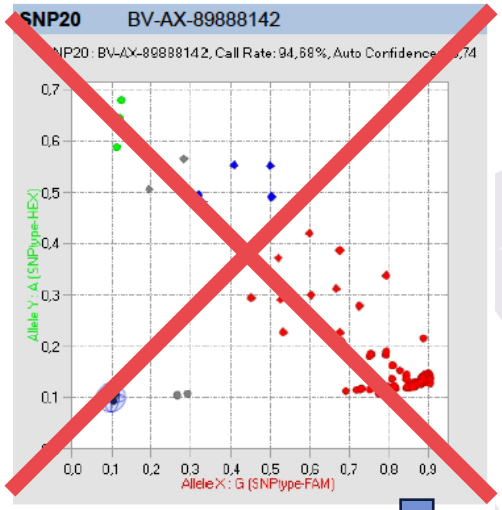
Array evaluated in many additional strawberry samples

10 probes substituted

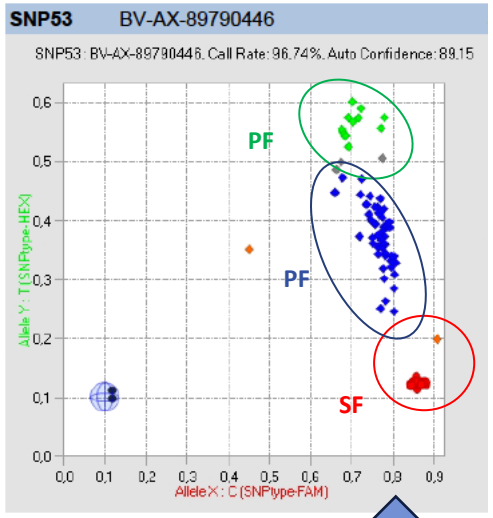
The Fluidigm Array is Flexible

Version 3.0

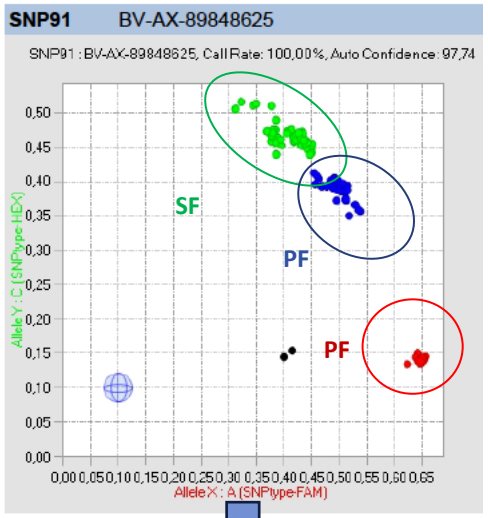
Perpetual Flowering



Perpetual Flowering



Perpetual Flowering

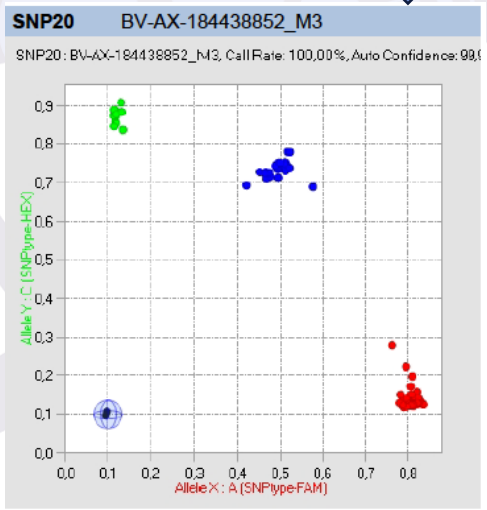


Verma et al., 2017

Same

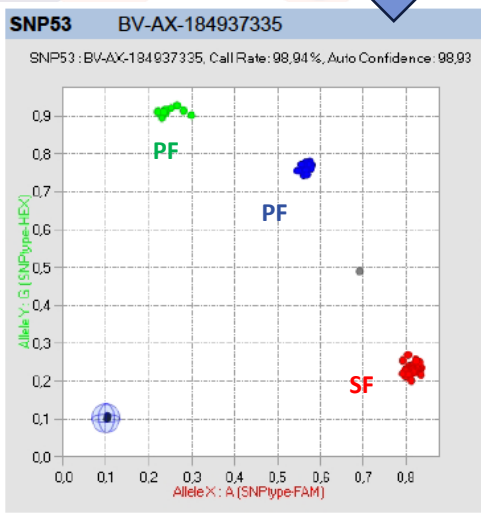
Version 4.0

FaRMp3 4C



Knapp et al., 2024

Perpetual Flowering



Feldmann et al., 2024

Probes can be changed!

Format can be changed!

- IFC 96.96 JUNO
- IFC 48.48 BIOMARK
- 192.24 IFC

Selection of markers for the Low-density SNP array for Marker-Assisted Selection (MAS)

RESISTANCES TO PESTS	SNPs in BV_v3 array	SNPs in BV_v4 array	Number of loci
Rce to Pyhtophtora Cactorum	5	5	4
Rce to Coletotrichum acutatum	1	1	1
Rce to Fusarium oxysporum f. sp.	4	4	3
Rce to Colletotrichum gloesporioides	3	2	1
Resistance to Verticillium dahliae	2	2	2
Resistance to Xanthomonas fragariae	2	1	1
Resistance to Powdery mildew (Podosphaera aphanis)	5	5	5
Resistance to Macrophomina phaseolina	3	4	3
Resistance to Tetranychus urticae	2	2	1
TOTAL	**27**	**26**	**21**
VEGETATIVE TRAITS			
Day neutrality, Everbearing	3	2	1
Runnering	-	2	2
Flowering time	3	4	3
TOTAL	**6**	**8**	**6**
PRODUCTION TRAITS			
Fruit weight (FW)	3	3	3
Total Yield	1	1	1
Class one yield specific	1	1	1
Fruit number	4	4	3
Total Fruit Number & Marketable Number	4	4	4
yield related	2	2	2
TOTAL	**15**	**15**	**14**
FRUIT QUALITY			
Fruit firmness	5	5	4
Vitamin C	4	4	4
SSC, Brix	4	5	5
Sucrose, raffinose, SSC and succinate	3	3	2
pH	1	1	1
pH / acidity perception	1	1	1
Malic acid	1	1	1
Internal fruit color (qualitative)	1	1	1
Fruit color (internal and external; qualitative)	1	1	1
Fruit color (quantitative), pelargonidins and Eriodictyol	1	1	1
Fruit color and pelargonidin-3-glucoside	1	1	1
Fruit brightness	2	1	1
Yellow flesh color and carotenoids	2	1	1
Total anthocyanins	1	2	1
epicatechin glucuronide isomer 1 and 2, kaempferol hexose 1, cyanidin hexose and rutin 2	2	1	1
propelargonidin dimer 2 and kaempferol hexose 2	1	2	2
pelargonidin-3-O-malonylglucoside	1	1	1
ellagic acid deoxyhexoside	1	1	1
Ellagic acid hexose	2	1	1
cinnamoyl glucoside	1	2	2
Galloyl-bis(HHDP)-glucose	3	1	1
Caramel aroma (Mesifurane)	1	3	2
Peach aroma (γ-decalactone)	2	1	1
grape aroma (Methyl anthranilate)	1	2	2
decyl, hexyl, octyl and nonyl acetates, octyl butanoate	2	1	1
Butyl, hexyl, octyl, nonyl, decyl, cinnamyl acetates, butyl and octyl butanoate, butyl and octyl hexanoate	2	2	1
Bostwick consistency	1	1	1
TOTAL	**48**	**47**	**42**
ALL TRAITS			
GRAND TOTAL	**96**	**96**	**83**

Low-density SNP array for Marker-Assisted Selection (MAS)

- The array includes 96 SNPs associated with different traits and can be interrogated with 94 samples (+ 2 internal Controls) at a time.

What we provide:

sgiker
Ikerkuntzarako Zerbitzu Orokorrak
Servicios Generales de Investigación

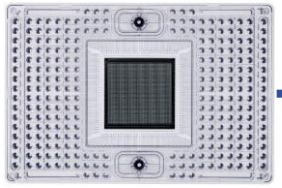
94 DNA **SAMPLES** PLATE

Fluidigm SNPtype **Reagents**

FLUIDIGM WORK **ASSAYS** PLATE

JUNO

1. Load the IFC
2. Pre-amplification: STA/LSP
3. qPCR at end point (ASPs/LSP) fluorescence detection.



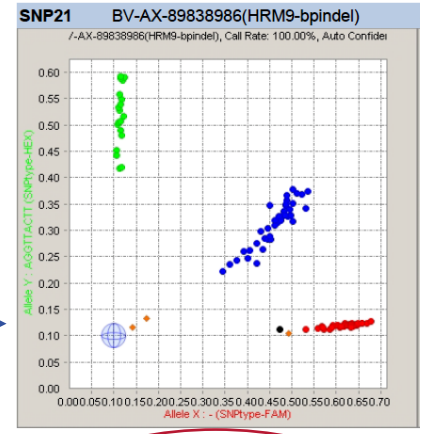
IFC 96.96 JUNO

Each IFC enables 9,216 reactions using 96 samples and assays.

BIOMARK HD

1. Chip Reading and visualization.
2. Analysis of results (SNP Genotyping Analysis Software)

EXAMPLE OF ONE PLOT: RESISTANCE TO ANTHRACNOSIS (*FaRca1*)



96 PLOTS (and excel file), each with genotypes for 94 strawberry samples

What we get:

Evaluation of marker polymorphism and Applicability in Genetic Diversity Analysis and Pedigree Testing

Material and Methods

- **Strawberry samples: 1217 samples (including duplicates)**
  - FNM: 346 samples from breeding program
  - IFAPA: 312 samples including most germplasm diversity
  - INVENIO/INRAE: 188 samples including core-collection and selections
  - JKI: 185 samples from germplasm collection
  - Hansabred: 94 samples including *F. ananassa* and selections introgressed with different species
  - LUKE: 92 samples including *F. chiloensis* × *F. virginiana* lines, cultivars and breeding selections
- **The most recent Fluidigm Array_4.0 used**

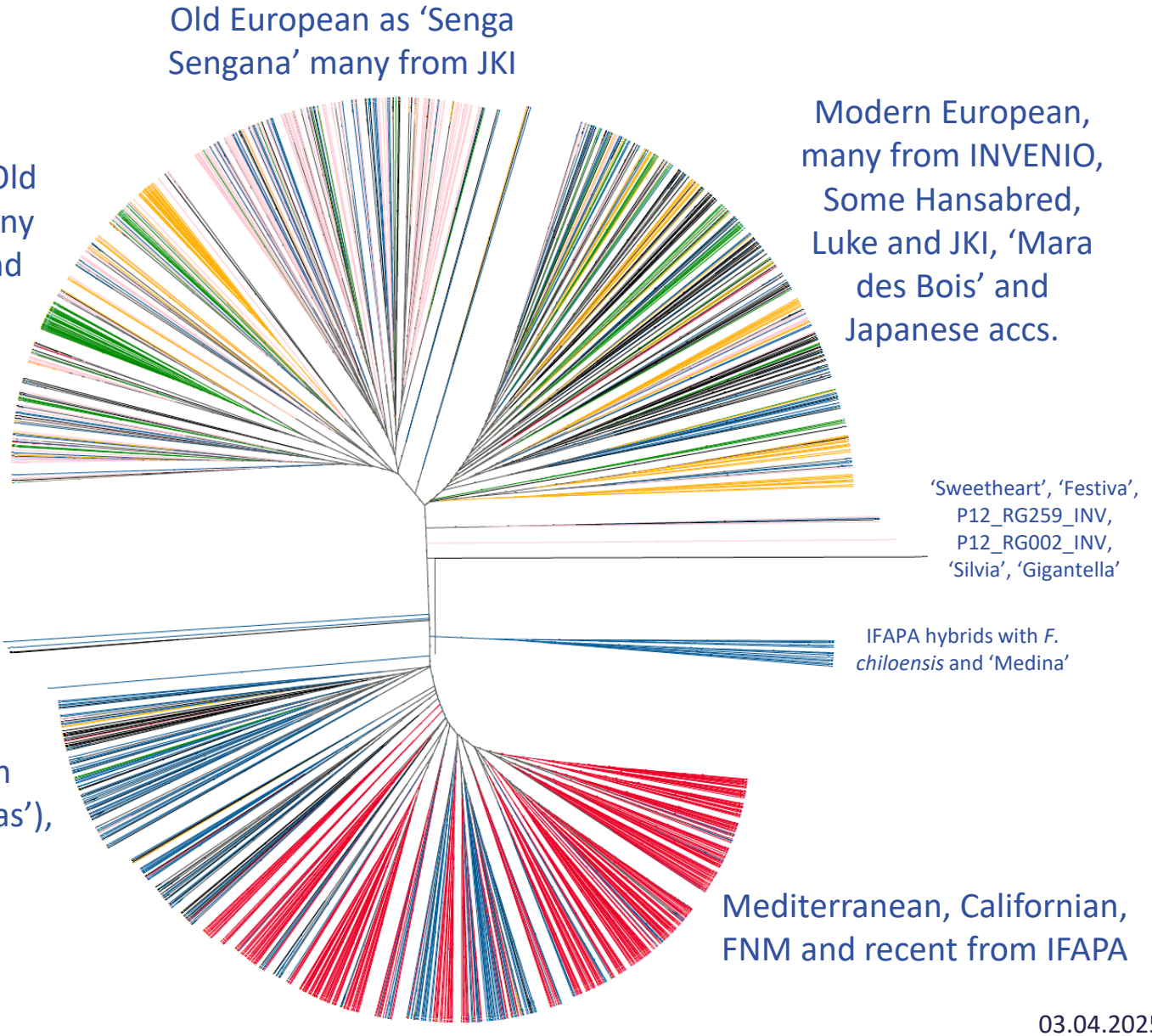
SNP POLYMORPHISM	NUMBER	%
POLYMORPHIC MAF> 0.02	60	62.50
POLYMORPHIC MAF < 0.02	16	16.67
NMH	20	20.83
MONOMORPHIC	0	0
HETEROZYGOSITY > 0.10	85	88.54
HETEROZYGOSITY < 0.10	11	11.46

Evaluation of the Fluidigm array for diversity analyses using 1217 samples

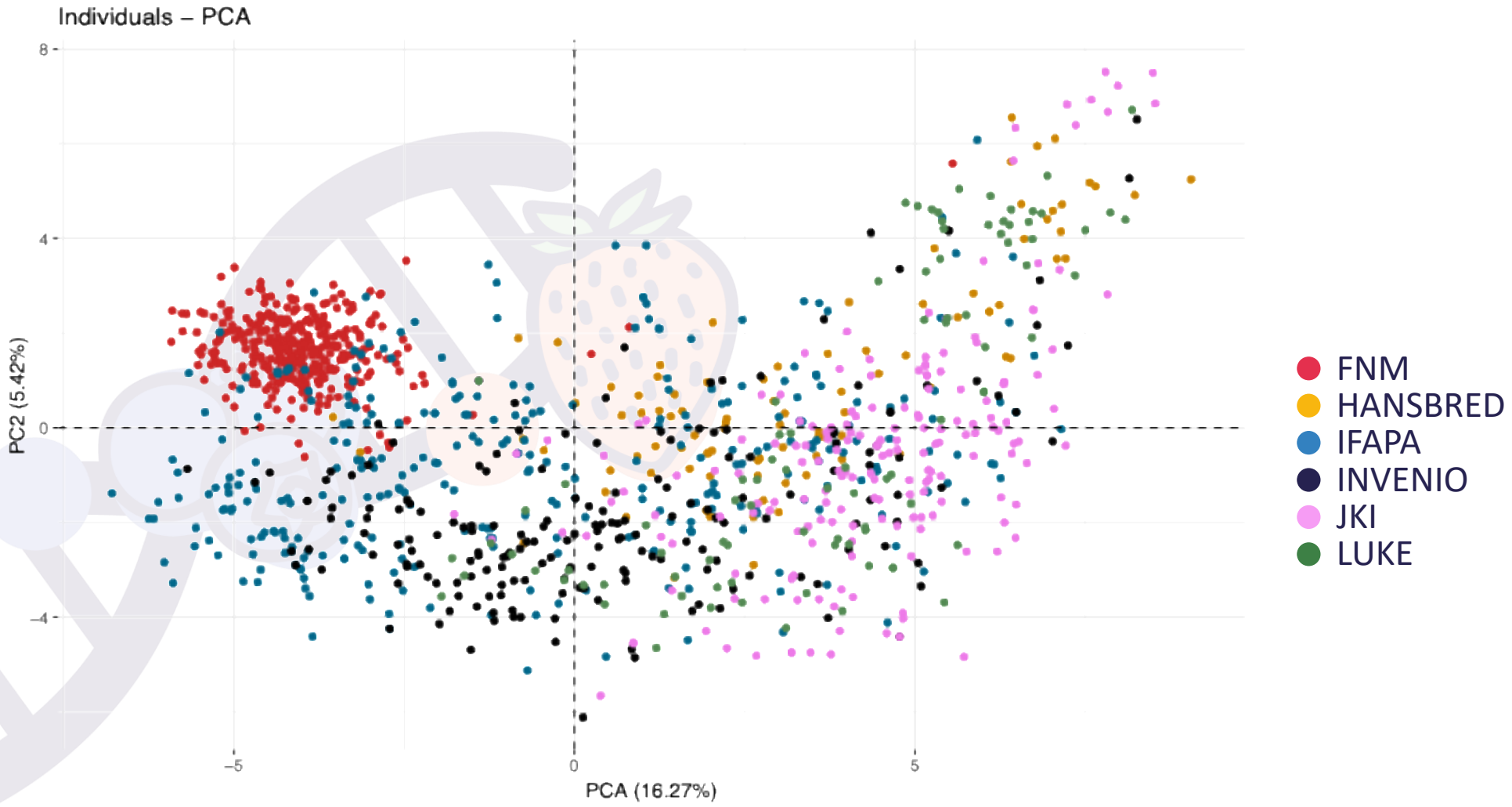
Het_{Exp} Range	0.37 - 0.57
Het_{Exp} Mean	0.4

Duplicated accessions were identified, as well as previously known mistakes

- FNM
- HANSABRED
- IFAPA
- INVENIO
- JKI
- LUKE



Evaluation of the Fluidigm array for diversity analyses using 1217 samples



PCA resulted in Genetic diversity similar to reported analyses

Evaluation of marker / trait Associations

Material and Methods

- **DNA SAMPLES:**

FNM:	346 samples
IFAPA:	138 samples
INVENIO/INRAE:	188 samples
JKI:	185 samples
Hansabred:	94 samples
LUKE:	92 samples

IFAPA TRAITS:
flowering date, runnering, yield, fruit number and weight, fruit firmness, color, acidity, SSC, sucrose, fructose and glucose, vitamin C, carotenoids and volatile organic compounds (VOCs)

INVENIO TRAITS:
Flowering date, everbearing, SSC, acidity, fruit weight, powdery mildew and Phytophthora susceptibility

Hansabred TRAITS:
Yellow flesh, white flesh, …

FNM TRAITS:
Marketable yield
SSC
Rce Powdery Mildew

LUKE TRAITS:
flowering date, runnering, yield-related, fruit colour, firmness, SSC, pH, powdery mildew, crown rot

JKI TRAITS:
Acidity, SSC, Firmness, Vitamin C, external and flesh color, glossiness and Xanthomonas Rce.

- **Marker / Trait association tested by ANOVA**

The Fluidigm Low-density SNP array

Evaluation of Marker/Trait associations

FNM DATA: Validation using 346 accessions

IFAPA DATA: Validation using 128-138 IFAPA accessions (1-3 years)

- **Marker / Trait association tested by ANOVA**

RESISTANCES TO PESTS

SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	Association FNM	OFF TARGETS
BV-AX-89838986(HRM9-bpindel)	Rce to *Coletotrichum acutatum*	Salinas et al., 2019; Salinas et al., 2020.	Fxa 6-3	6B		98.8%	
BV-AX-89797117	Rce to *Colletotrichum gloesporioides*	Anciro et al., 2018	Fxa 6-3	6B			
BV-UFCg1HRM01-T>A	Rce to *Colletotrichum gloesporioides*	Anciro et al., 2018; Bassil et al., 2021; Chandra et al., 2021	Fxa 6-3	6B			
BV-AX-166521396	Rce to *Fusarium oxysporum f. sp.*	Pincot et al., 2018	Fxa 2-3	2D			
BV-AX-184298748	Rce to *Fusarium oxysporum f. sp.*	Pincot et al., 2022	Fxa 6-3	6B			
BV-AX-89879574	Rce to *Fusarium oxysporum f. sp.*	Pincot et al., 2018	Fxa 2-3	2D			
BV-AX-123363542	Rce to *Fusarium oxysporum f. sp.*	Pincot et al., 2022	Fxa 1-4	1A			
BV-AX-184438852	Rce to *Macrophomina phaseolina*	Knapp et al., 2024	Fxa 4-2	4C			
BV-AX-89819829	Rce to *Macrophomina phaseolina*	Nelson et al., 2021	Fxa 2-2	2A			
BV-AX-89781417	Rce to *Macrophomina phaseolina*	Nelson et al., 2021	Fxa 2-2	2A			
BV-AX-89830947	Rce to *Macrophomina phaseolina*	Nelson et al., 2021	Fxa 4-3	4A			
BV-AX-89836603	Rce to Powdery mildew (*Podosphaera aphanis*)	Sargent et al., 2019	Fxa 5-3	5B			
BV-AX-184211493	Rce to Powdery mildew (*Podosphaera aphanis*)	Unpublished	Fxa 6-2	6C		*	7
BV-Affx-88882488(AX-123358917)	Rce to Powdery mildew (*Podosphaera aphanis*)	Cockerton et al., 2018	Fxa 6-4	6D			
BV-AX-184762224	Rce to Powdery mildew (*Podosphaera aphanis*)	Unpublished	Fxa 7-2	7A			
BV-AX-89800832	Rce to Powdery mildew (*Podosphaera aphanis*)	Sargent et al., 2019	Fxa 7-1	7C			
BV-Affx-88880166(AX-89795820)	Rce to *Pyhtophtora cactorum*	Nellist et al., 2019	Fxa 6-2	6C			
BV-RPCHRM5(AX-89845054)	Rce to *Pyhtophtora cactorum*	Mangandi et al., 2017; Noh et al., 2018.	Fxa 7-3	7B		97.7%	
BV-Affx-88882267(AX-89796426)	Rce to *Pyhtophtora cactorum*	Nellist et al., 2019	Fxa 6-4	6D			
BV-RPCKASPH3(AX-89845076)	Rce to *Pyhtophtora cactorum*	Mangandi et al., 2017; Noh et al., 2018.	Fxa 7-3	7B		97.3%	
BV-Affx-88902178(AX-89845399)	Rce to *Pyhtophtora cactorum*	Nellist et al., 2019	Fxa 7-3	7B			
BV-3402043-40-T>G-LG6	Rce to spider mite (*Tetranychus urticae*)	Unpublished	Fxa 6-2	6C			
BV-C1827664-43-A>G-0	Rce to spider mite (*Tetranychus urticae*)	Unpublished	Fxa 6-2	6C			
BV-Affx-88867578(AX-89833535)	Rce to *Verticillium dahliae*	Antanaviciute et al., 2015; Cockerton et al., 2019	Fxa 5-4	5C			
BV-Affx-88836863(AX-89788025)	Rce to *Verticillium dahliae*	Antanaviciute et al., 2015; Cockerton et al., 2019	Fxa 3-2	3B			
BV-Xf1HRM-8073(AX-89798073)	Rce to *Xanthomonas fragariae*	Roach et al., 2016; Oh et al., 2020	Fxa 6-2	6C			

VEGETATIVE TRAITS

Day neutrality, Everbearing

ID_v4	SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	Association FNM	OFF TARGETS
SNP53	BV-AX-184937335	Day neutrality, Everbearing.	Feldmann et al., 2024	Fxa 4-4	4B	Good		
SNP91	BV-AX-89848625	Day neutrality, Everbearing.	Verma et al., 2017	Fxa 4-4	4B	Fair		

Flowering time

ID_v4	SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	Association FNM	OFF TARGETS
SNP07	BV-AX-184201950	Flowering date	Prohaska et al., 2024	Fxa 6-4	6D			
SNP37	BV-1786870-16-G>T	Flowering date	Prohaska et al., 2024	Fxa 6-4	6D			
SNP35	BV-AX-184671198	Flowering time	Prohaska et al., 2024	Fxa 3-4	3A	**		Many
SNP45	BV-AX-184872554	Flowering time	Prohaska et al., 2024	Fxa 6-1	6A			

Runnering

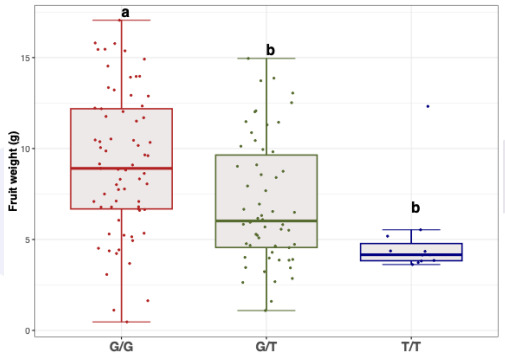
ID_v4	SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	Association FNM	OFF TARGETS
SNP47	BV-AX-184937178	Runner number	Muñoz et al., 2024	Fxa 4-3	4A	*		Many
SNP16	BV-AX-184418817	Runnering date and number	Muñoz et al., 2024	Fxa 5-1	5A	***		Many

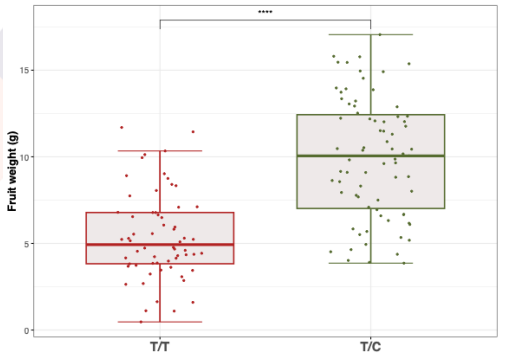
PRODUCTION TRAITS

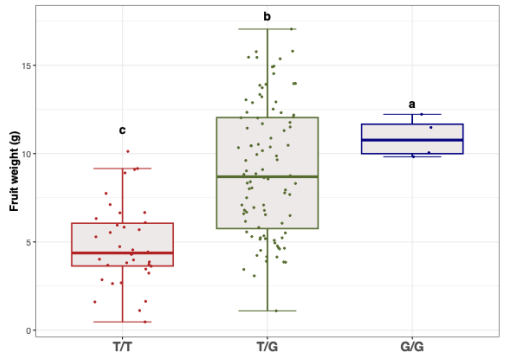
Fruit Weight (FW)

ID_v4	SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	Association FNM	OFF TARGETS	Related Traits
SNP14	BV-30785-64-G>T-LG1-FW	Fruit Weight (FW)	Zorrilla-Fontanesi et al., 2011	Fxa 1-4	1A	***/***/**		Many	FW/Yield/FN
SNP90	BV-AX-184187121	Fruit Weight 3C	Muñoz et al., 2024	Fxa 3-3	3C	***/***/**		Many	FW/Yield/FN
SNP58	BV-AX-184477554	Fruit Weight 3D	Muñoz et al., 2024	Fxa 3-1	3D	***/***/*	*	Many	FW/Yield/FN

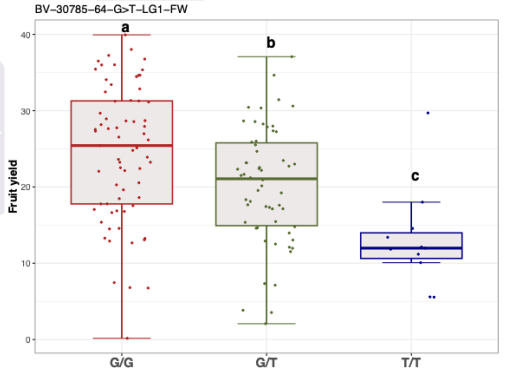
FW

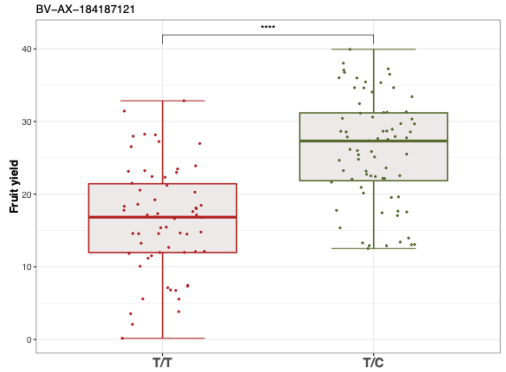


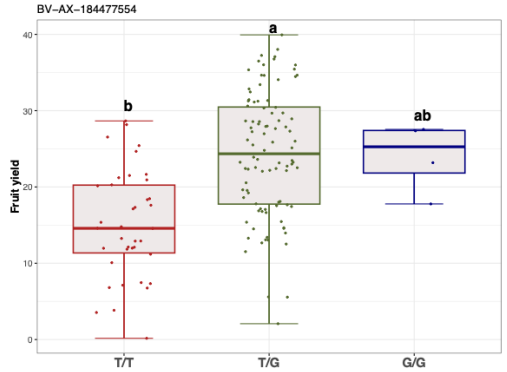




Yield







PRODUCTION TRAITS

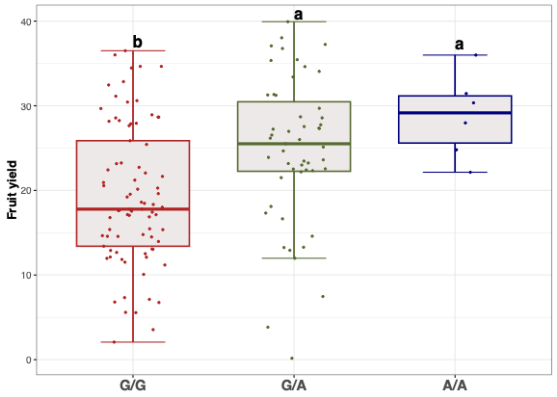
Fruit Number and yield related

ID_v4	SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	Association FNM	OFF TARGETS	Related Traits
SNP38	BV-37396-57-G>A-LG5	Fruit number	Zorrilla-Fontanesi et al., 2011	Fxa 5-3	5B				
SNP50	BV-37540-54-A>C	Fruit number	Zorrilla-Fontanesi et al., 2011	Fxa 5-3	5B	***		Many	
SNP18	BV-AX-166522841	Total Fruit Number	Cockerton et al., 2021	Fxa 4-2	4C				
SNP30	BV-AX-166523870	Total Fruit Number	Cockerton et al., 2021	Fxa 5-4	5C	***/***	**	Many / 7	Yield/FN
SNP28	BV-AX-123361756b	Total & Marketable Fruit Number	Cockerton et al., 2021	Fxa 5-1	5A				
SNP63	BV-AX-123357423	Total & Marketable Fruit Number	Cockerton et al., 2021	Fxa 2-4	2B	***/**	*	Many / 7	Yield/FN
SNP64	BV-AX-123367138	Total & Marketable Fruit Number	Cockerton et al., 2021	Fxa 4-3	4A	***		Many	
SNP80	BV-AX-89826839	Total & Marketable Fruit Number	Cockerton et al., 2021	Fxa 3-2	3B				
SNP77	BV-AX-166520085	Total Fruit Number, class 2 mass	Cockerton et al., 2021	Fxa 6-2	6C	***		Many	
SNP40	BV-AX-123361870	Class one yield specific QTL, Total & Fruit Marketable Number	Cockerton et al., 2021	Fxa 5-2	5D	***/*		Many	Yield/FN
SNP41	BV-AX-166505049	Marketable number, total number & total mass, class 2 mass	Cockerton et al., 2021	Fxa 3-4	3A				
SNP73	BV-25929-8-T>G-LG5	Total Yield	Zorrilla-Fontanesi et al., 2011	Fxa 5-3	5B	***/*		Many	Yield/FN

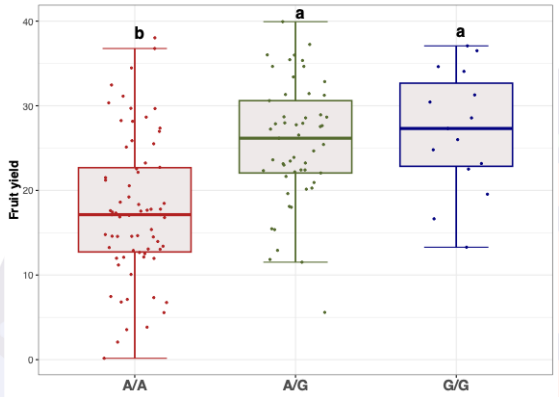
PRODUCTION TRAITS

Fruit Number and yield related

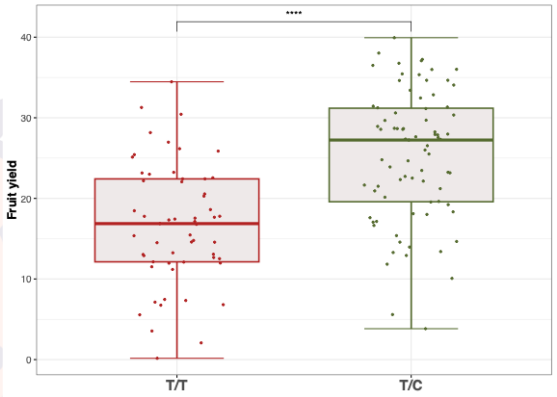
BV-37540-54-A>C



BV-AX-166523870



BV-AX-123357423



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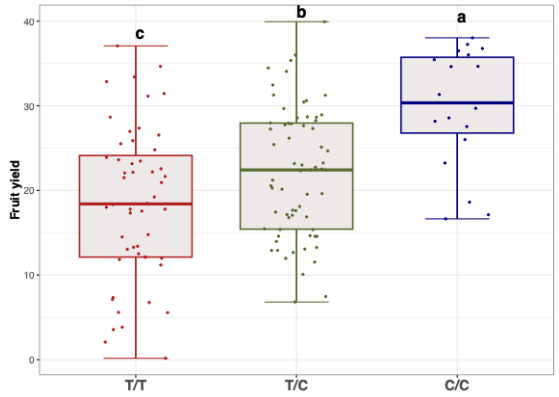
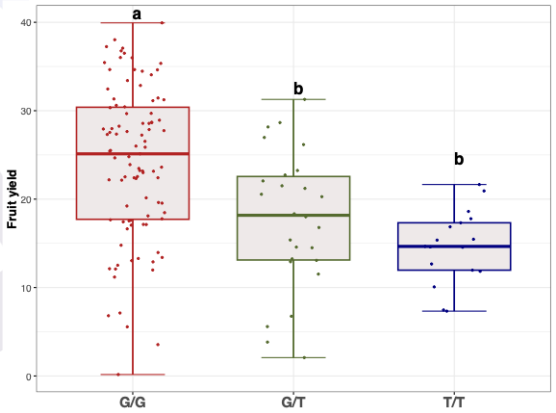


BV-AX-166520085



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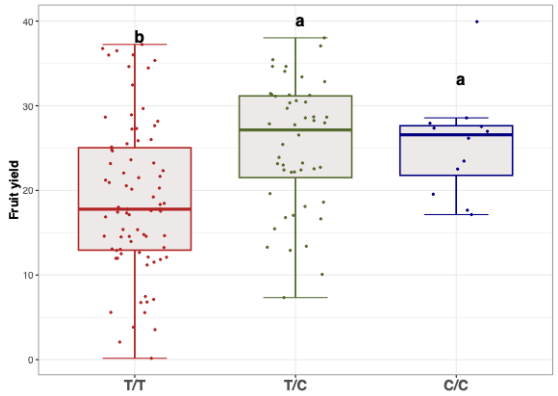
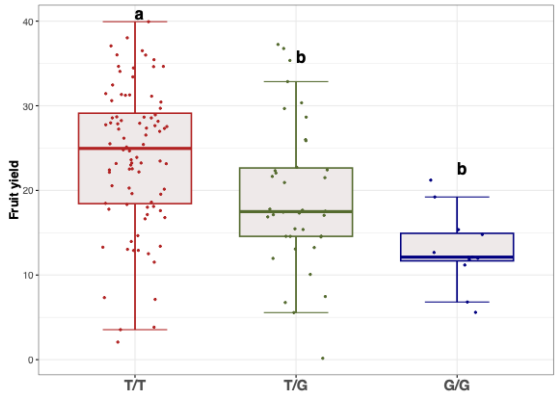


BV-25929-8-T>G-LG5

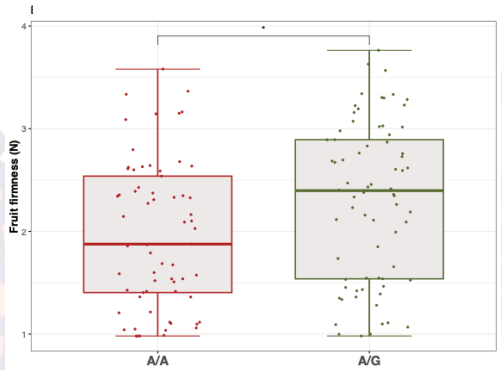


FRUIT QUALITY

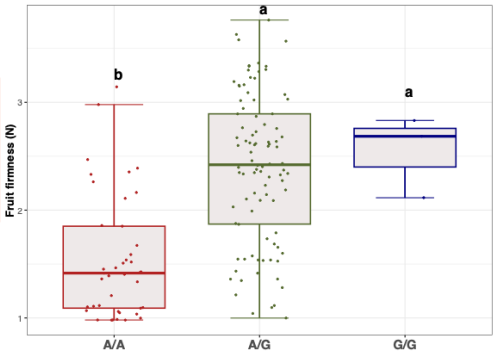
Fruit Firmness

ID_v4	SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	Association FNM	OFF TARGETS	Related Traits
SNP46	BV-F25909-50-T>C-LG7	Fruit firmness	Zorrilla-Fontanesi et al., 2011	Fxa 7-2	7A				
SNP89	BV-AX-89844975	Fruit firmness	Rey-Serra et al., 2021	Fxa 7-1	7C	*		Many	
SNP94	BV-FaEXP2	Fruit firmness	Zorrilla-Fontanesi et a., 2011	Fxa 7-2	7A				
SNP65	BV-AX-166521510	Fruit Firmness 3A	Muñoz et al., 2024	Fxa 3-4	3A	***		Many	
SNP81	BV-AX-184210669	Fruit Firmness 6A	Muñoz et al., 2024	Fxa 6-1	6A	***		Many	

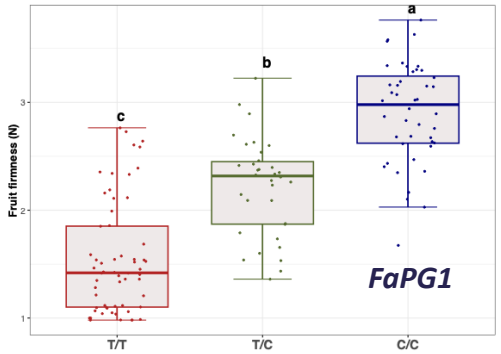
BV-AX-89844975



BV-AX-166521510



BV-AX-184210669



Fruit Acidity

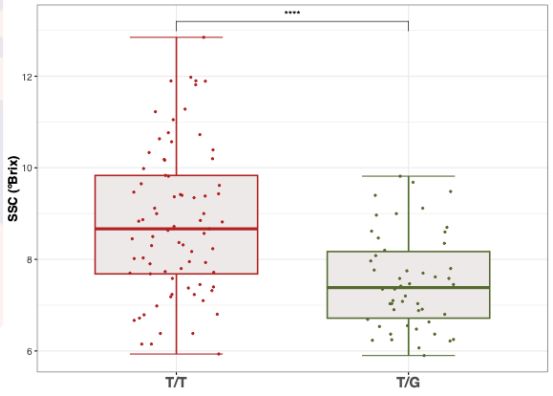
ID_v4	SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	Association FNM	OFF TARGETS	Year
SNP43	BV-AX-184669632	pH	Unpublished	Fxa 1-2	1B	**/*		Many	2021/emmeans
SNP76	BV-AX-123525124	pH / acidity perception	Cockerton et al., 2021	Fxa 5-1	5A				
SNP22	BV-F17638-25-G>A-LG2	Malic acid	Vallarino and Pott et al., 2019	Fxa 2-4	2B	**/*		Many	2021/emmeans

FRUIT QUALITY

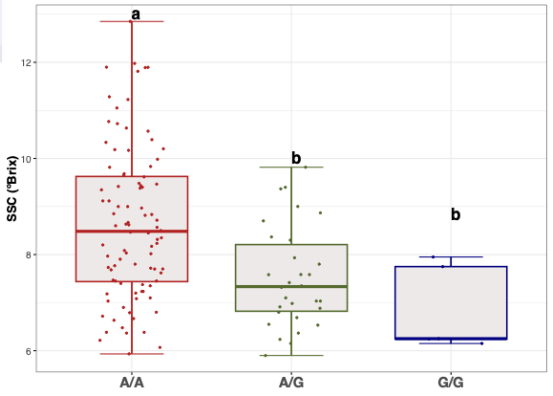
Soluble Solids Content (SSC, Brix)

ID_v4	SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	Association FNM	OFF TARGETS	Related Traits
SNP08	BV-AX-166522371	SSC, Brix	Fan et al., 2024	Fxa 3-2	3B	***/*		Many	4years/Suc
SNP31	BV-AX-184380255	SSC, Brix	Unpublished	Fxa 1-2	1B				
SNP36	BV-UDP-Glu4-epi(5-3)	Sucrose and raffinose	Zorrilla-Fontanesi et al., 2011; Vallarino et al., 2019	Fxa 5-3	5B	*		17	Only for sucrose
SNP60	BV-UDP-Glu4-epi(5-4)	SSC, Brix	Natarajan et al., 2020	Fxa 5-4	5C				
SNP66	BV-AX-184103725	SSC, Brix	Unpublished	Fxa 2-1	2C	***/*		Many	3years/2023
SNP67	BV-AX-184962799	SSC, Brix	Unpublished	Fxa 1-4	1A	***/**		Many	3years/2023
SNP78	BV-AX-184139313_2	SSC, Brix	Unpublished	Fxa 1-1	1D	**		Many	
SNP34	BV-F19519-5-A>T-LG5	SSC, sucrose, raffinose, and succinate	Zorrilla-Fontanesi et al., 2011; Vallarino et al., 2019	Fxa 5-3	5B				

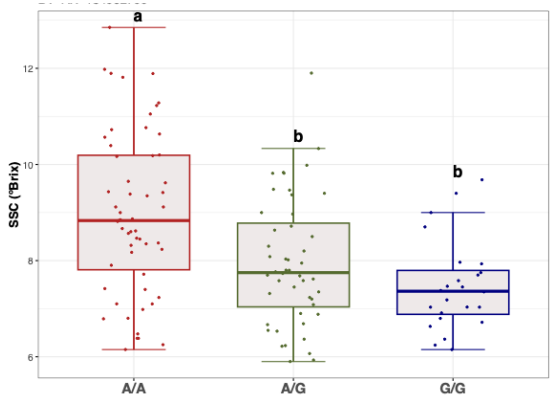
BV-AX-166522371



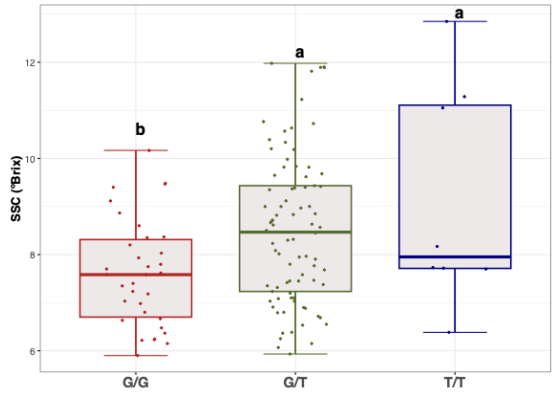
BV-AX-184103725



BV-AX-184962799



BV-AX-184139313_2



FRUIT QUALITY

Fruit color

ID_v4	SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	OFF TARGETS
SNP59	BV-IFC-2	Internal fruit color	Castillejo et al., 2020	Fxa 1-2	1B	***	
SNP72	BV-WS_CID_01	Fruit color (internal and external)	Castillejo et al., 2020	Fxa 1-2	1B	***	
SNP25	BV-AX-184599570	Fruit brightness	Prohaska et al., 2024b	Fxa 3-1	3D		
SNP96	BV-AX-89826853	Fruit color and pelargonidin-3-glucoside	Labadie et al., 2022	Fxa 3-4	3A		
SNP71	BV-M00247-47-C>G-LG5	Fruit color, pelargonidins and Eriodictyol	Zorrilla-Fontanesi et al., 2011; Pott et al., 2020	Fxa 5-1	5A		
SNP82	BV-FaCCD4_YFC-4	Yellow flesh color and carotenoids	Amaya et al., 2025	Fxa 4-4	4B	***	1A, 3D, 4B
SNP32	BV-AX-89795985	Total anthocyanins	not yet published	Fxa 6-1	6A		
SNP57	BV-AX-89899628	Total anthocyanins	not yet published	Fxa 6-1	6A		
SNP52	BV-AX-123362709	pelargonidin-3-O-malonylglucoside	Davik et al., 2020	Fxa 6-2	6C		

Ascorbic Acid (Vitamin C)

ID_v4	SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	OFF TARGETS	Year
SNP13	BV-12939803-5-TG-5Fvb5	Vitamin C	Muñoz et al., 2023	Fxa 5-3	5B			
SNP29	BV-AX-184662098-AsA	Vitamin C	Muñoz et al., 2023	Fxa 3-4	3A	*	Many	Only one
SNP49	BV-1794253-39-CT-3Fvb5	Vitamin C	Muñoz et al., 2023	Fxa 5-1	5A			
SNP83	BV-M33503-35-A>T-LG5	Vitamin C	Vallarino and Pott et al., 2019.	Fxa 5-4	5C			

FRUIT QUALITY

Bostwick consistency

ID_v4	SNP Assay_v4	Trait	Institution	Reference	CAM Chrom.	CAM Position	RR Chrom.	RR Position
SNP54	BV-AX-184047020	Bostwick consistency	INRAE, France	Unpublished	Fxa 6-1	2463405	6A	32521089

Volatile Organic Compounds (VOCs) associated with flavor

ID_v4	SNP Assay_v4	Trait	Reference	CAM Chrom.	RR Chrom.	Association IFAPA	OFF TARGETS
SNP95	BV-NGD001-UFGDKASP	Peach aroma (γ-decalactone)	Sánchez-Sevilla et al., 2015; Chambers et al., 2015; Cruz-Rus et al., 2017; Lee et al., 2017; Noh et al., 2017; Oh et al., 2021	Fxa 3-2	3B	***	10
SNP05	BV-AX-166520175	Caramel aroma (Mesifurane)	Barbey et al., 2021	Fxa 1-2	1B		
SNP86	BV-AX-166502845	Caramel aroma (Mesifurane)	Barbey et al., 2021	Fxa 1-2	1B		
SNP11	BV-FaOMT-SI-NO	Caramel aroma (Mesifurane)	Zorrilla-Fontanesi et al., 2012. Cruz-Rus et al., 2017.	Fxa 7-4	7D	***	3
SNP75	BV-AX-123358624	grape aroma (Methyl anthranilate)	Barbey et al., 2021	Fxa 5-4	5C	**	Many
SNP88	BV-AX-123540849	grape aroma (Methyl anthranilate)	Barbey et al., 2021	Fxa 2-2	2A		
SNP23	BV-FxaC_21g33790-G>A	Butyl, hexyl, octyl, nonyl, decyl, cinnamyl acetates, butyl and octyl butanoate, butyl and octyl hexanoate	Zorrilla-Fontanesi et al., 2012	Fxa 6-1	6A	***	1
SNP62	BV-42001-38-G>A-LG6	Butyl, hexyl, octyl, nonyl, decyl, cinnamyl acetates, butyl and octyl butanoate, butyl and octyl hexanoate	Zorrilla-Fontanesi et al., 2012	Fxa 6-1	6A		
SNP87	BV-AX-123358920	decyl, hexyl, octyl and nonyl acetates, octyl butanoate	Fan et al., 2021	Fxa 6-1	6A	*	Few




FRUIT QUALITY

Specialized Metabolites

ID_v4	SNP Assay_v4	Trait	Institution	Reference	CAM Chrom.	CAM Position	RR Chrom.	RR Position
SNP51	BV-AX-123357156	ellagic acid deoxyhexoside	NIBIO	Davik et al., 2020	Fxa 1-3	7160000	1C	6669958
SNP02	BV-30785-64-G>T-LG1-EA	Ellagic acid hexose	IFAPA, UMA, Spain	unpublished	Fxa 1-4	7751009	1A	8188276
SNP85	BV-29488-18-C>T-LG5	epicatechin glucuronide isomer 1 and 2, kaempferol hexose 1, cyanidin hexose and rutin 2	IFAPA, UMA, Spain	Pott et al., 2020	Fxa 5-3	19661842	5B	8097255
SNP70	BV-F36936-33-A>G-LG4	Galloyl-bis(HHDP)-glucose	IFAPA, UMA, Spain	unpublished	Fxa 4-3	25623347	4A	6331575
SNP04	BV-AX-123361118	cinnamoyl glucoside	NIBIO	Davik et al., 2020	Fxa 3-2	3824400	3B	3472264
SNP42	BV-AX-166524984	cinnamoyl glucoside	NIBIO	Davik et al., 2020	Fxa 6-1	15582900	6A	20012974
SNP10	BV-F13908-63-A>G-LG5	propelargonidin dimer 2 and kaempferol hexose 2	IFAPA, UMA, Spain	Pott et al., 2020	Fxa 5-1	952473	5A	1018042
SNP93	BV-F13400-15-G>A-LG5	propelargonidin dimer 2 and kaempferol hexose 2	IFAPA, UMA, Spain	Pott et al., 2020	Fxa 5-1	19669	5A	268159

Not validated!

The Fluidigm Low-density SNP array

Conclusions:

- The Fluidigm low-density Array can be used for estimating genetic diversity and pedigree relationships
- Many marker/trait associations were confirmed using the Fluidigm array on the tested samples. Most of them are for mayor QTL or mendelian traits, but also several markers for QTL with small effects showed significant differences in important traits such as yield related or SSC.
- ANOVA using the 96 SNPs resulted in many associations: Some might be true associations due to linkage, but other might be FALSE POSITIVES most probably due to population structure and kindship not taken into account.

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