

**Uso de los marcadores SNP para asociar
resistencia a *Moniliophthora roreri* un
patógeno de importancia del cacao
(*Theobroma cacao*).**

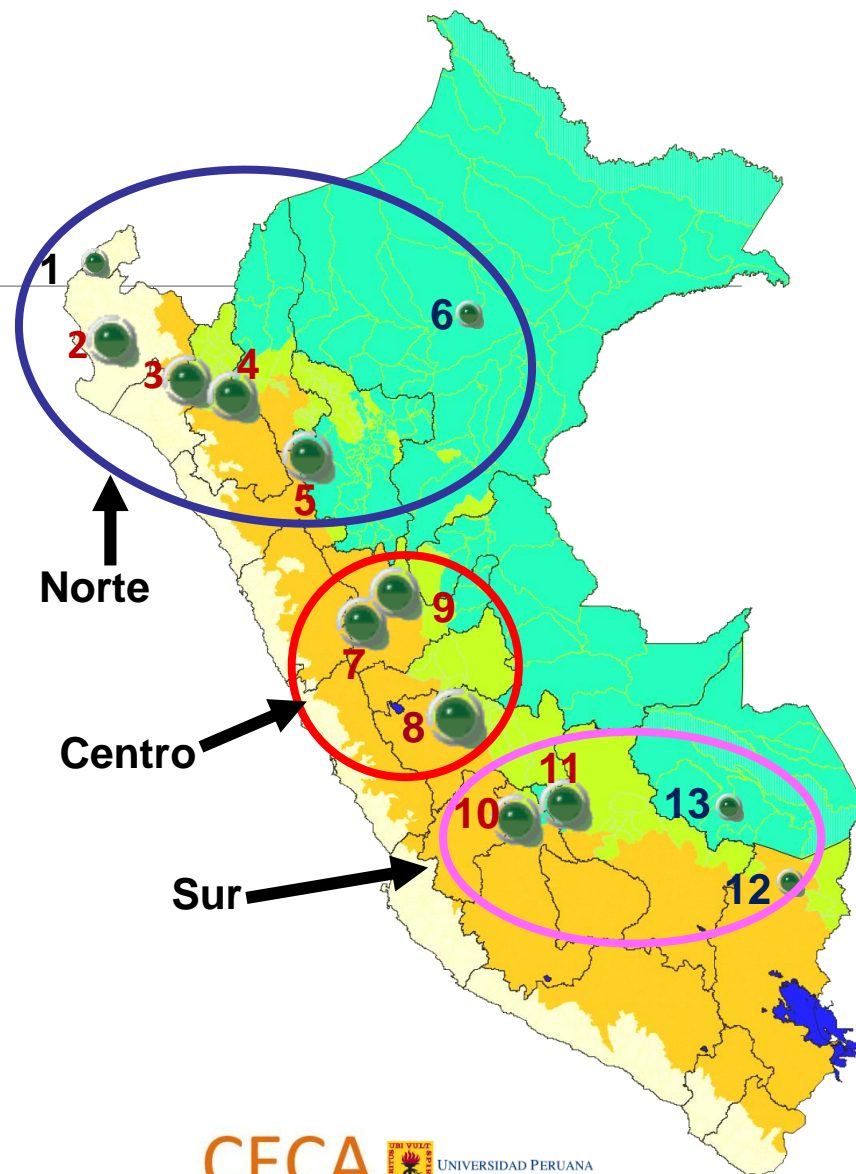
Mg. Boris Gutarra Castillo
Universidad Continental

Cacao en el Perú 2012

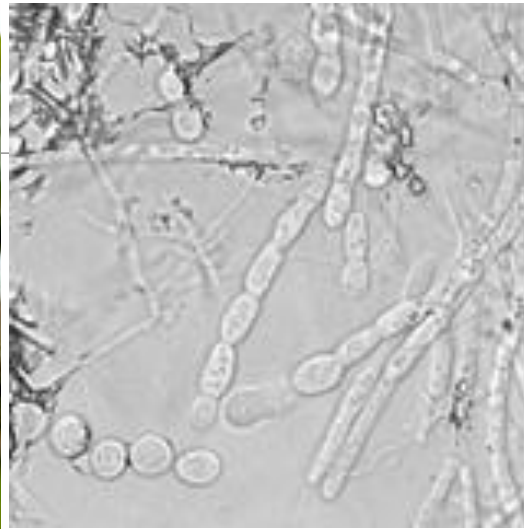
mapa peru cacao.png

Area Total (ha)	91,497
Rendimiento (kg/ha)	683
Produccion (t)	62,492
Unidades Productivas	>60,000
Productores	>30,000
<u>Norte:</u> Tumbes, Piura, Amazonas, Cajamarca, La Libertad, Lambayeque	9,739 ha 11.0%
<u>Centro:</u> San Martín, Huánuco, Jujín, Pasco, Ucayali	47,330 ha 52.0%
<u>Sur:</u> Ayacucho, Cusco, Madre de Dios	34,429 ha 38.0%

Fuente: MINAG: [Series Históricas de Producción Agrícola - Compendio Estadístico \(SISCA\)](#)
Elaboración propia

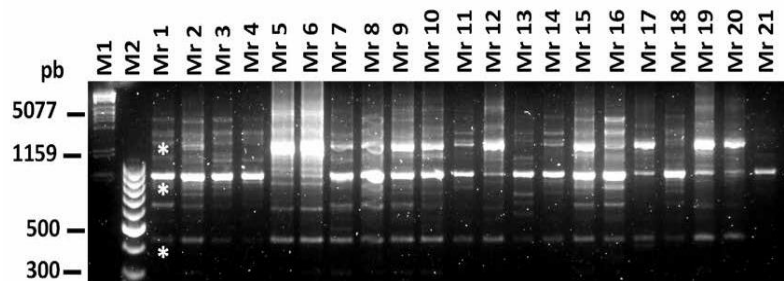


La moniliasis : *Moniliophthora roreri*

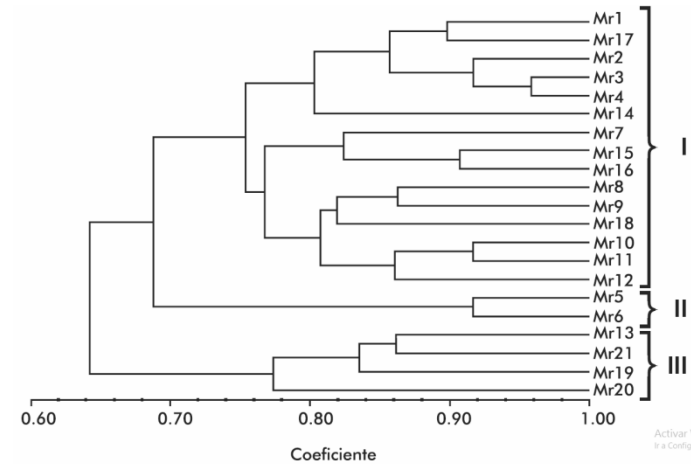


Phillips et al 2007

Clasificación ISI	RANGO
Resistente;	0 - 1,25
Moderadamente Resistente	1,26 - 2,50
Moderadamente susceptible	2,51 - 3,75
Susceptible	3,76 - 5,0



Gutarra et al 2013

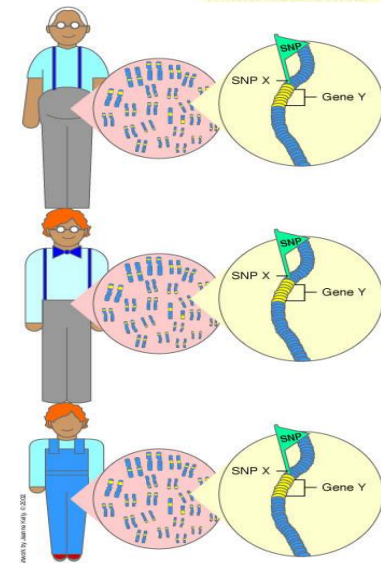
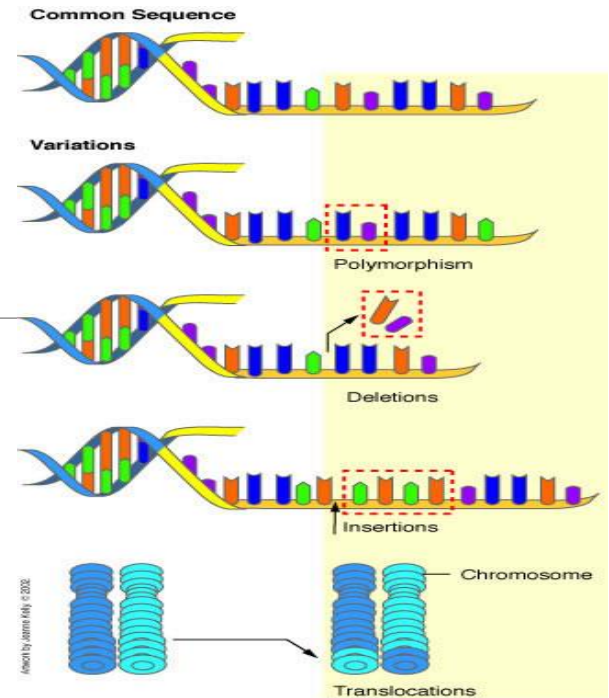


What is SNP ?

- ▶ A **SNP** is defined as a single base change in a DNA sequence that occurs in a significant proportion (more than 1 percent) of a large population.

SNP facts

- ▶ SNPs are found in
 - coding and (mostly) noncoding regions.
- ▶ Occur with a very high frequency
 - about 1 in 1000 bases to 1 in 100 to 300 bases.
- ▶ The abundance of SNPs and the ease with which they can be measured make these genetic variations significant.
- ▶ SNPs close to particular gene acts as a marker for that gene.
- ▶ SNPs in coding regions may alter the protein structure made by that coding region.



Objetivos

Obtener marcadores SNP asociado a resistencia a *Moniliophthora roreri*

Hipótesis

Se espera encontrar algunos marcadores SNP estén asociados a resistencia a *Moniliophthora roreri*

Aspectos metodológicos: población y muestra

Clones resistentes y clones sensibles

Clon	Genotipo	N.R	Clon	Genotipo	N.R
SCA6	A A	R	ICT1281	A A	S
EET 400	A T	R	ICT1414	A A	S
ICT2821	A A	R	ICT2161	A T	S
ICT1043	A A	R	ICT2163	A T	S
ICT1561	A A	R	ICT2171	A T	S
ICT1251	A A	R	ICT2173	A T	S
ICT1506	A A	R	ICT2174	A A	S
ICT1187	A A	R	ICT2492	A T	S
ICT1182	A A	R	ICT2501	A T	S
ICT2824	A T	R	ICT2504	A A	S
ICT2653	A T	R	ICT2702	T T	S
ICT2142	A A	R	ICT2703	A A	S
ICT1112	A A	R	ICT2705	A T	S
ICT2823	A A	R	ICT2825	A T	S
ICS1	T T	S			
ICT1104	A T	S			
ICT1189	A T	S			

UNIVERSIDAD NACIONAL DE SAN MARTÍN- TARAPOTO
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TESIS:
NIVELES DE RESISTENCIA DE GENOTIPOS PROMISORIOS DE
CACAO (*Theobroma cacao* L.). COLECCIÓN ICT PARA
Monilophthora roreri EN LA REGIÓN SAN MARTÍN.

PRESENTADO POR EL BACHILLER:
GINA PAOLA SÁNCHEZ TORRES

PARA OBTENER EL TÍTULO PROFESIONAL DE
INGENIERO AGRÓNOMO
TARAPOTO - PERÚ

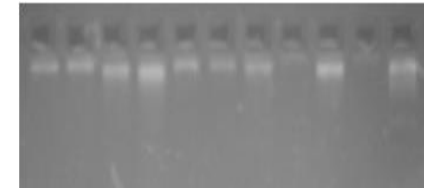
2011



31 clones que forman parte de la colección del Instituto de cultivos tropicales (ICT-Tarapoto)

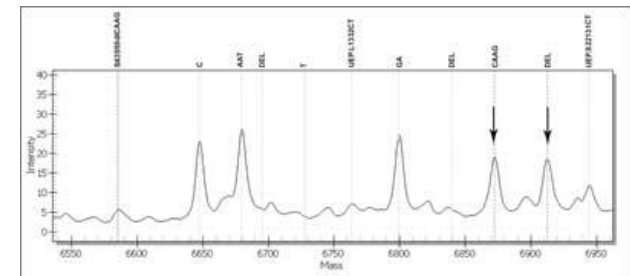
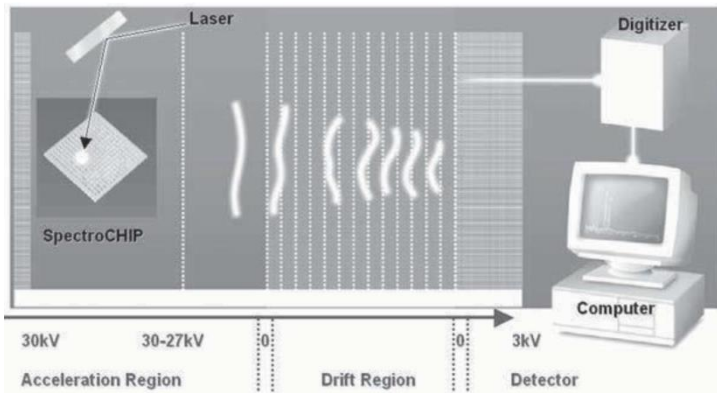
14 clones son resistentes y 17 sensibles a *M.roreri*

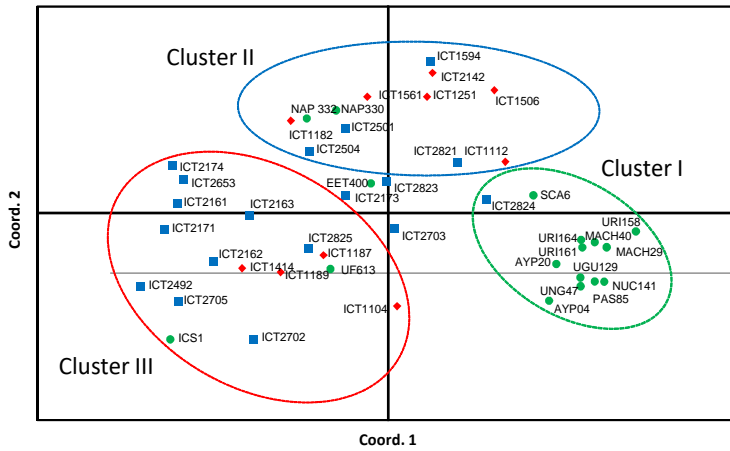
Extracción de ADN de *Theobroma cacao*



Genotipificación:

Sistema: sistema de espectrofotometría de masas MALDI-TOF (Sequenom .Inc)

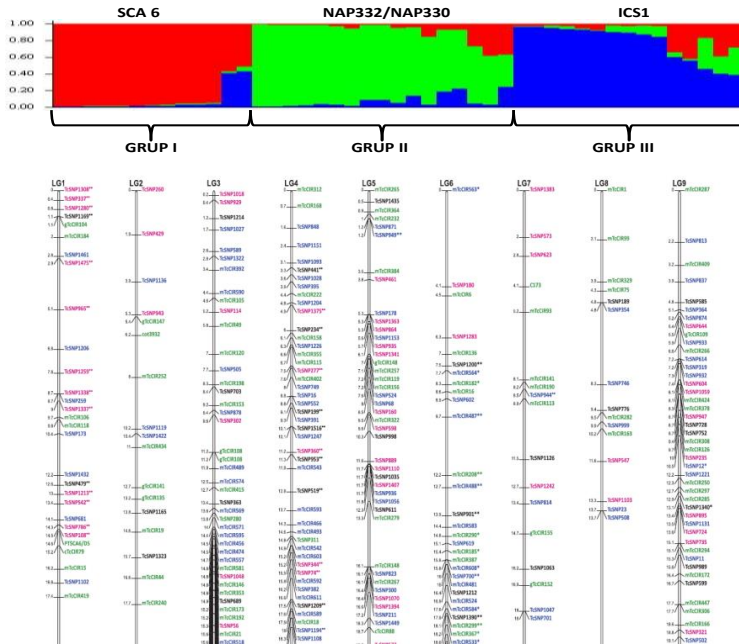




Búsqueda de nuevos SNPs



Distribución de los clones ICT en base a SNP (Gutarra & Gallegos 2012)



Mapa de alta densidad del Cacaco (Allegre et al 2012)

Nº	SNP	Cromosoma	Alelos
1	TcSNP568	1	A/G
2	TcSNP233	4	T/C
3	TcSNP960	4	A/T
4	TcSNP1387	6	A/G
5*	TcSNPWRKYp	7	C/G
6*	TcSNPWRKY3	7	C/T
7*	TcSNPWRKY7b2	7	A/T
8*	TcSNP1912	9	C/T
9	TcSNP1064	9	A/G
10	TcSNP617	2	A/G
11	TcSNP800	8	T/C
12	TcSNP1498	8	A/G
13	TcSNP1285	8	A/G
14	TcSNP491	2	C/G
15	TcSNP316	2	A/T
16	TcSNP494	2	A/G
17	TcSNP1042	8	C/G
18	TcSNP1124	8	T/C
19	TcSNP190	8	T/G
20	TcSNP942	8	A/G
21	TcSNP1431	1	T/G
22	TcSNP1018	3	C/G

Aspectos metodológicos: análisis de datos

Fisher's exact

This test is similar than the **Chi-square test** but in the case of to have a small sample size, it is better to use Fisher's exact test than Chi-squared.

$$p = \frac{(a+b)!(c+d)!(a+c)!(b+d)!}{n!a!b!c!d!}$$

Programa: R

Linkage Disequilibrium (LD)

$$D' = \frac{|D|}{D_{\max}}$$

where $D_{\max} = \min(p_A p_b, p_a p_B)$ if $D > 0$;

$D_{\max} = \min(p_A p_B, p_a p_b)$ if $D < 0$

$$r^2 = \frac{D^2}{p_A p_a p_B p_b}$$

Programa: Haploview

Resultados

	SNP	P
1	TcSNP568	0.06066
2	TcSNP233	0.7823
3	TcSNP960	0.5111
4	TcSNP1387	0.005046
5	TcSNPWRKYp	0.8124
6	TcSNPWRKY3	0.3076
7	TcWRKY7b2	0.04627
8	TcNPR1_1912	0.5916
9	TcSNP1064	0.1977
10	TcSNP617	0.02007
11	TcSNP800	1
12	TcSNP1498	0.2045
13	TcSNP1285	0.06214
14	TcSNP491	0.4812
15	TcSNP316	0.3311
16	TcSNP494	0.4754
17	TcSNP1042	0.1168
18	TcSNP1124	0.09426
19	TcSNP190	0.07113
20	TcSNP942	0.2148
21	TcSNP1431	0.01103
22	TcSNP1018	1



Predicción

Welcome to the Cacao Genome Project

Cacao production is important! Not only is it the basic ingredient in the world's favorite confection, chocolate, but it provides a livelihood for over 6.5 million farmers in Africa, South America and Asia and ranks as one of the top ten agriculture commodities in the world. Historically, cocoa production has been plagued by serious losses due to pests and diseases. The release of the cacao genome sequence will provide researchers with access to the latest genomic tools, enabling more efficient research and accelerating the breeding process, thereby expediting the release of superior cacao cultivars. The sequenced genotype, Matina 1-6, is representative of the genetic background most commonly found in the cacao producing countries, enabling results to be applied immediately and broadly to current commercial cultivars. Matina 1-6 is highly homozygous which greatly reduces the complexity of the sequence assembly process. While the sequence provided is a preliminary release, it already covers 92% of the genome, with approximately 35,000 genes. We will continue to refine the assembly and annotation, working toward a complete finished sequence. Updates will be made available via this website, to keep informed check back regularly or join CGSD mailing list. If you have any questions, feedback or problems, please contact us through the contact link on the navigation bar, we promise to respond in a timely manner.



- ### News
- "The genome sequence of the most widely cultivated cacao type, and its use to identify candidate genes regulating pod color"
 - CacaoCyc version 1.0 is available on CSDA
 - The Cacao Genome Sequence is released 3 years ahead of schedule!
 - Cacao Genome Database Computer Demo at PAG 2012
 - Public and scientific interest in the chocolate genome continues to grow!
 - Cacao Genome Sequencing and Breeders Workshop at PAG 2012



Description	Max score	Total Query score	Coverage	E value	Ident	Accession
PF02127. Theobroma cacao ADP47F carrier protein 1, mitochondrial [LOC18184341_cDNA]	188	188	100%	1e-43	100%	XM_018184344
Theobroma cacao genome assembly (chromosome 1)	188	188	100%	1e-43	100%	TT284382.1
PF02127. Theobroma cacao ADP47F carrier protein 1, mitochondrial [LOC18184341_cDNA]	188	188	87%	2e-20	90%	XM_017728485
PF02127. Theobroma cacao ADP47F carrier protein 1, mitochondrial [LOC18184341_cDNA]	188	188	87%	2e-20	90%	XM_017728484
PF02127. Theobroma cacao ADP47F carrier protein 1, mitochondrial [LOC18184341_cDNA]	182	182	87%	1e-18	89%	XM_018835588

- Se encontró asociación al QTL respectivo 4 marcadores SNP asociados a genes de resistencia:

TcSNP617: catalyze key steps in the pathway of lignin monomer biosynthesis.

TcWRKY7b2: increased expression of over 2000 plant defense genes.

TcSNP617: biosynthesis of aromatic amino acids (phenylalanine, tyrosine, and tryptophan) precursors for the synthesis of defense and repair compounds.

TcSNP1431: proteins from a solute carrier family which transfer molecules across the membranes of the mitochondria.

Discusión

Debido a la poca cantidad de marcadores SNP utilizados así como la cantidad de genotipos es posible que la asociación este limitada a la colección ICT y su linaje: Por tener carácter cuantitativo y poligenico la resistencia solo se habría logrado asociar un locus que están dentro de haplotipos. Por esta razón es necesario utilizar técnica de SNP array, SNPLine o RADseq para lograr una asociación genómica (GWAS) con miles de SNP

RESEARCH ARTICLE

Open Access



Identification of candidate genes involved in Witches' broom disease resistance in a segregating mapping population of *Theobroma cacao* L. in Brazil

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Abstract

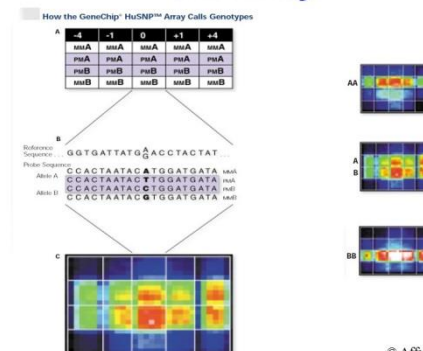
Background: Witches' broom disease (WBD) caused by the fungus *Moniliophthora perniciosa* is responsible for considerable economic losses for cacao producers. One of the ways to combat WBD is to plant resistant cultivars. Resistance may be governed by a few genetic factors, mainly found in wild germplasm.

Results: We developed a dense genetic linkage map with a length of 852.8 cM that contains 3,526 SNPs and is based on the MP01 mapping population, which counts 459 trees from a cross between the resistant 'TSH 1188' and the tolerant 'CCN 51' at the Mars Center for Cocoa Science in Barro Preto, Bahia, Brazil. Seven quantitative trait loci (QTL) that are associated with WBD were identified on five different chromosomes using a multi-trait QTL analysis for outbreeders. Phasing of the haplotypes at the major QTL region on chromosome IX on a diversity panel of genotypes clearly indicates that the major resistance locus comes from a well-known source of WBD resistance, the clone 'SCAVINA 6'. Various potential candidate genes identified within all QTL may be involved in different steps leading to disease resistance. Preliminary expression data indicate that at least three of these candidate genes may play a role during the first 12 h after infection, with clear differences between 'CCN 51' and 'TSH 1188'.

Activar
Ir a Confir



SNP Array



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Conclusiones

Se ha logrado asociar 4 SNP relacionados a resistencia a patógenos los que podrían utilizarse en programas de selección asistida por marcadores moleculares (MAS) en poblaciones que tengan ascendencia con la de la colección ICT.

Referencias bibliográficas

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GRACIAS

