

***Ustilago maydis* COMO UN MODELO DE DIFERENCIACIÓN  
CELULAR Y PATOGÉNESIS**



**José Ruiz Herrera**

## ALGUNAS DEFINICIONES DE DIFERENCIACIÓN

“...*growth* is an increase in cellular mass; *development* refers to the sequence of structural and functional changes occurring during the life cycle of an organism; *differentiation* connotes the acquisition of a determined function or structure during development. *Morphogenesis* encompasses those aspects of development related to morphological changes”.

*Nickerson and Bartnicki-García (1964)*

“...*Development* (can be) divided into three separate aspects...

- 1 ... *Differentiation* involves the structural and functional specialization of individual cells from one of a number of common basic stem cells which are usually competent to differentiate in several different ways....
- 2 ... *Pattern formation*, is concerned with the spatial organization of differentiation: that is how cells of different types develop in the correct spatial, temporal and proportional relationships to each other...
- 3 ... “*Morphogenesis*, refers to the development of the shape and form of the organism and its individual parts...”

*Garrod and Ahworth (1973)*

“... *growth*, *morphogenesis* and *development* (are) fundamentally problems of biological order... the term *morphogenesis* will refer to the processes that generate the forms of cells in the course of their growth, division or development”

*Harold (1990)*

“..*biochemical differentiation* is a process of cellular specialization in which a substance is accumulated to a unique extent over a specified period of time”

*Wright (1978)*

“... *differentiation* constitutes the series of events that carefully organized in time and space give rise to cell specialization, without alterations in their genetic characteristics”

*Ruiz-Herrera (1984)*

# ALGUNAS VENTAJAS DE LOS HONGOS COMO MODELOS DE ESTUDIO

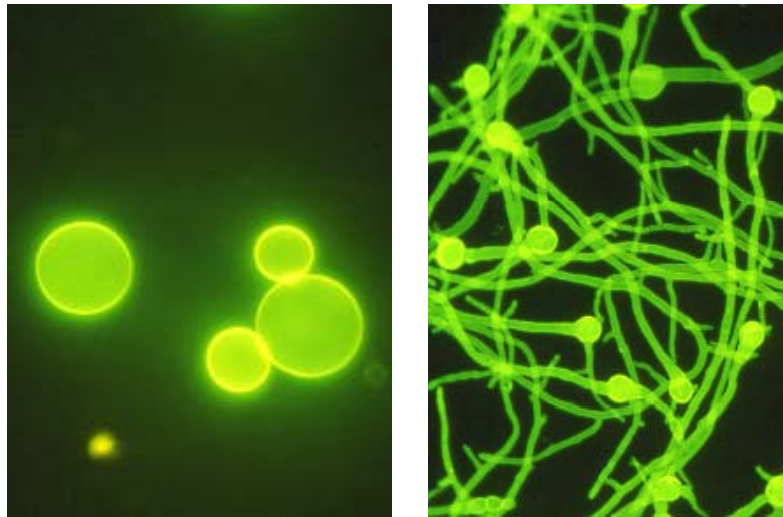
(DE ACUERDO CON EL CONCEPTO DE MAX DELBRÜCK: USAR SIEMPRE EL MODELO MÁS SIMPLE PARA RESOLVER LOS PROBLEMAS MÁS DIFÍCILES)

1. SON EUCARIOTES SIMPLES
2. SE CULTIVAN CON FACILIDAD EN EL LABORATORIO
3. SUS PROCESOS DIFERENCIATIVOS SON REPRODUCIBLES
4. ALTERNAN CICLOS SEXUALES Y ASEXUALES
5. SON HAPLOIDES DURANTE LA MAYOR PARTE DE SU CICLO DE VIDA
6. SE PUEDEN OBTENER DIPLOIDES O HETEROCARIONES
7. SE PUEDEN TRANSFORMAR GENETICAMENTE
8. SE CUENTA CON TODAS LAS ARMAS DE GENETICA MOLECULAR PARA SU ESTUDIO
9. SE CONOCE LA SECUENCIA GENOMICA DE VARIAS ESPECIES

## **DIMORFISMO:**

La propiedad que poseen diversos hongos para crecer en forma de micelio o de levadura, dependiendo de las condiciones del medio ambiente

El dimorfismo se puede considerar como un fenómeno de diferenciación.



## **VENTAJAS:**

1. Es reversible
2. Es dispensable
3. Es reproducible en el laboratorio

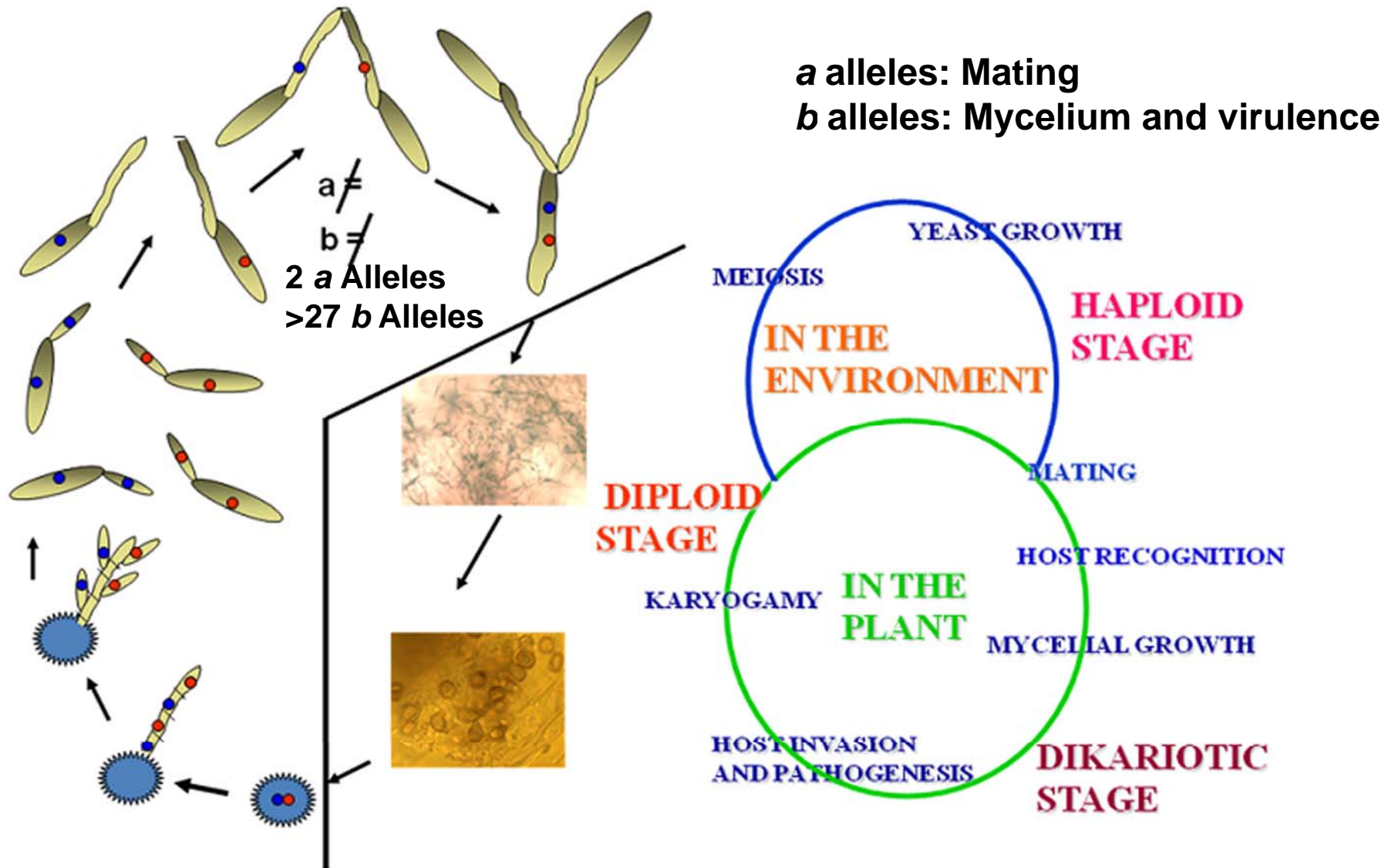
# EL MODELO DE ESTUDIO: EL HONGO BASIDIOMICOTA *Ustilago maydis*

## ORIGIN AND NATIONALITY OF *Ustilago maydis*

The association of *Ustilago maydis* with grasses as teozintles originated about 50 million years ago, but the current population of *U. maydis* originated with the domestication and cultivation of maize in Mexico, which occurred about 8000 to 10,000 years. This process totally transformed the population of *U. maydis*, replacing those of the native teozintles strains derived from corn. Subsequently *U. maydis* followed the distribution of domesticated maize from Mexico ...

Georgina May *et al.*, 2002; 2006

# *Ustilago maydis* life cycle



# INFECCIÓN NATURAL POR *Ustilago maydis*



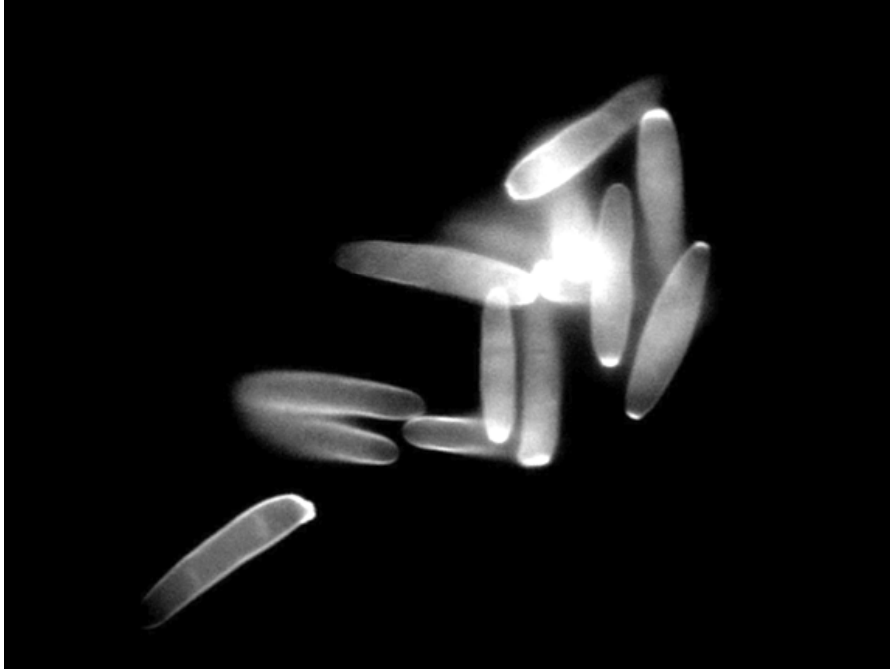
**MAÍZ**



**TEOZINTLE**

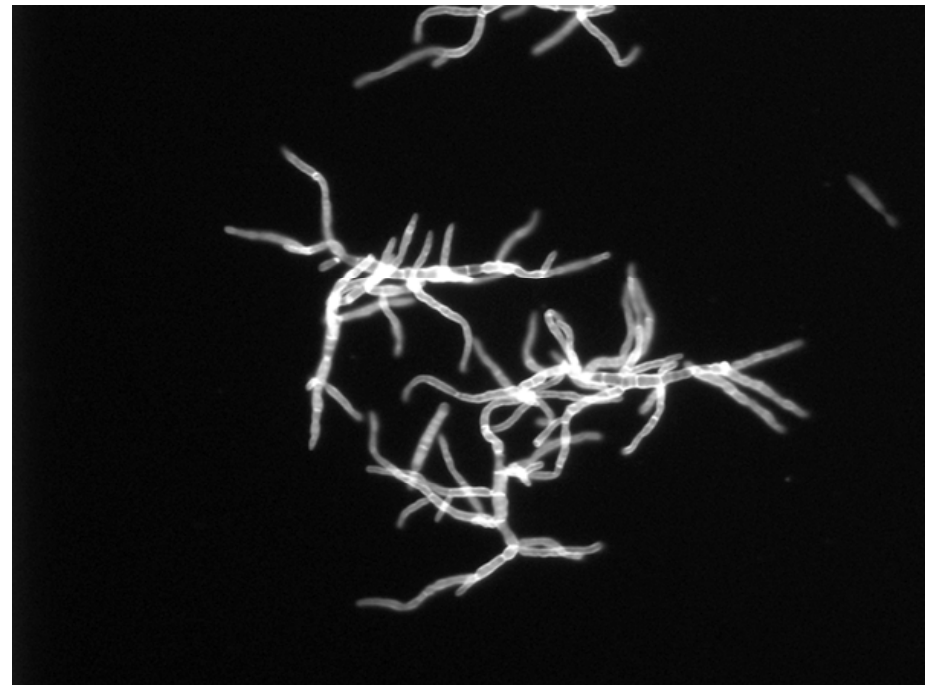
**SOLO CEPAS DICARIÓTICAS O DIPLOIDES HACEN MICELIO Y SON VIRULENTAS**  
**(a1bX/a2/bY)**

**DIMORFISMO *in vitro* DE *Ustilago maydis***



**pH 7**

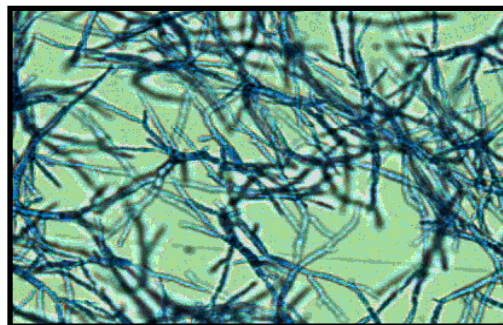
**pH 3**



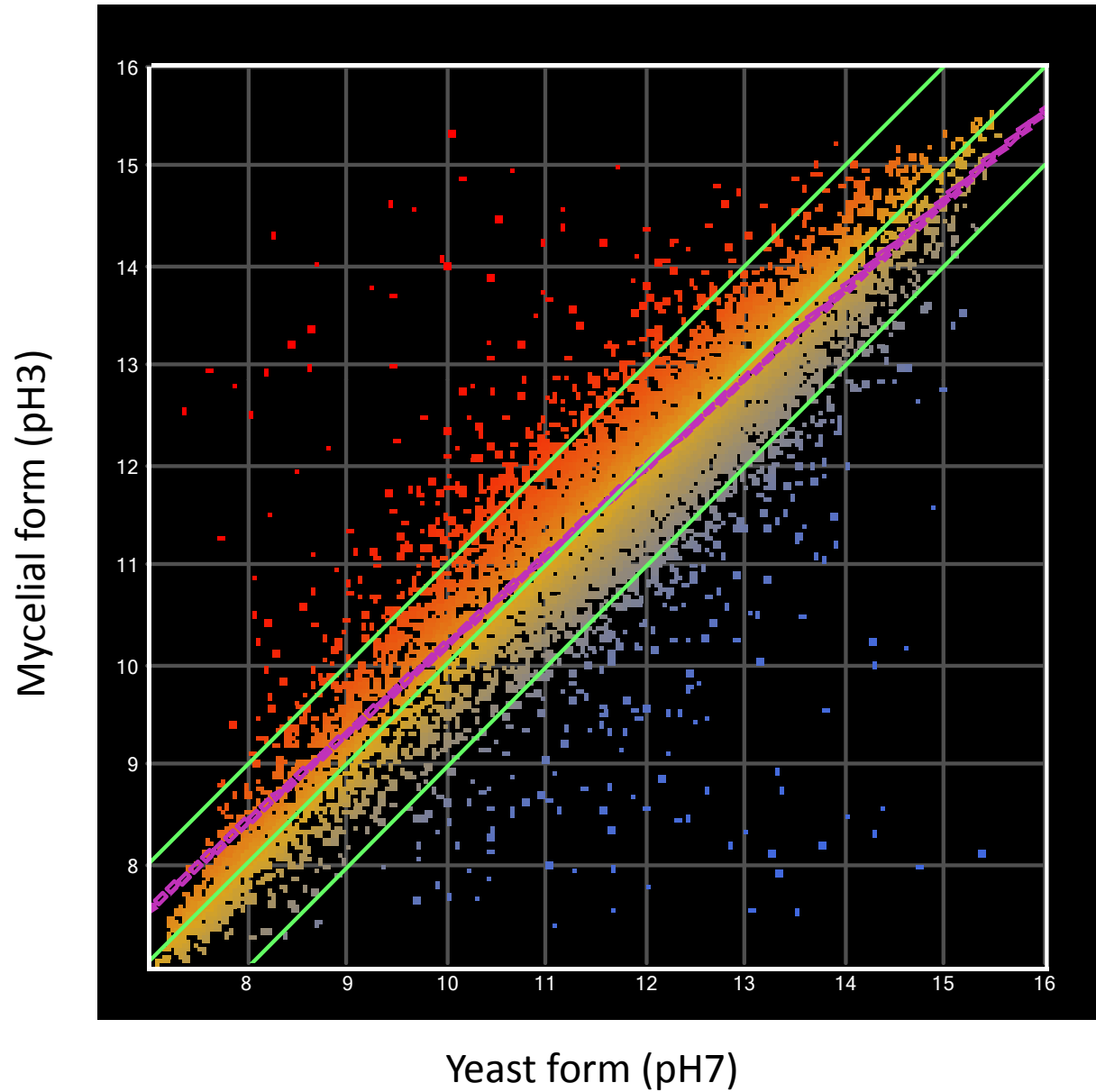


**INDUCCIÓN DE MICELIO *in vitro* POR pH en DIFERENTES  
CEPAS DE *Ustilago maydis***

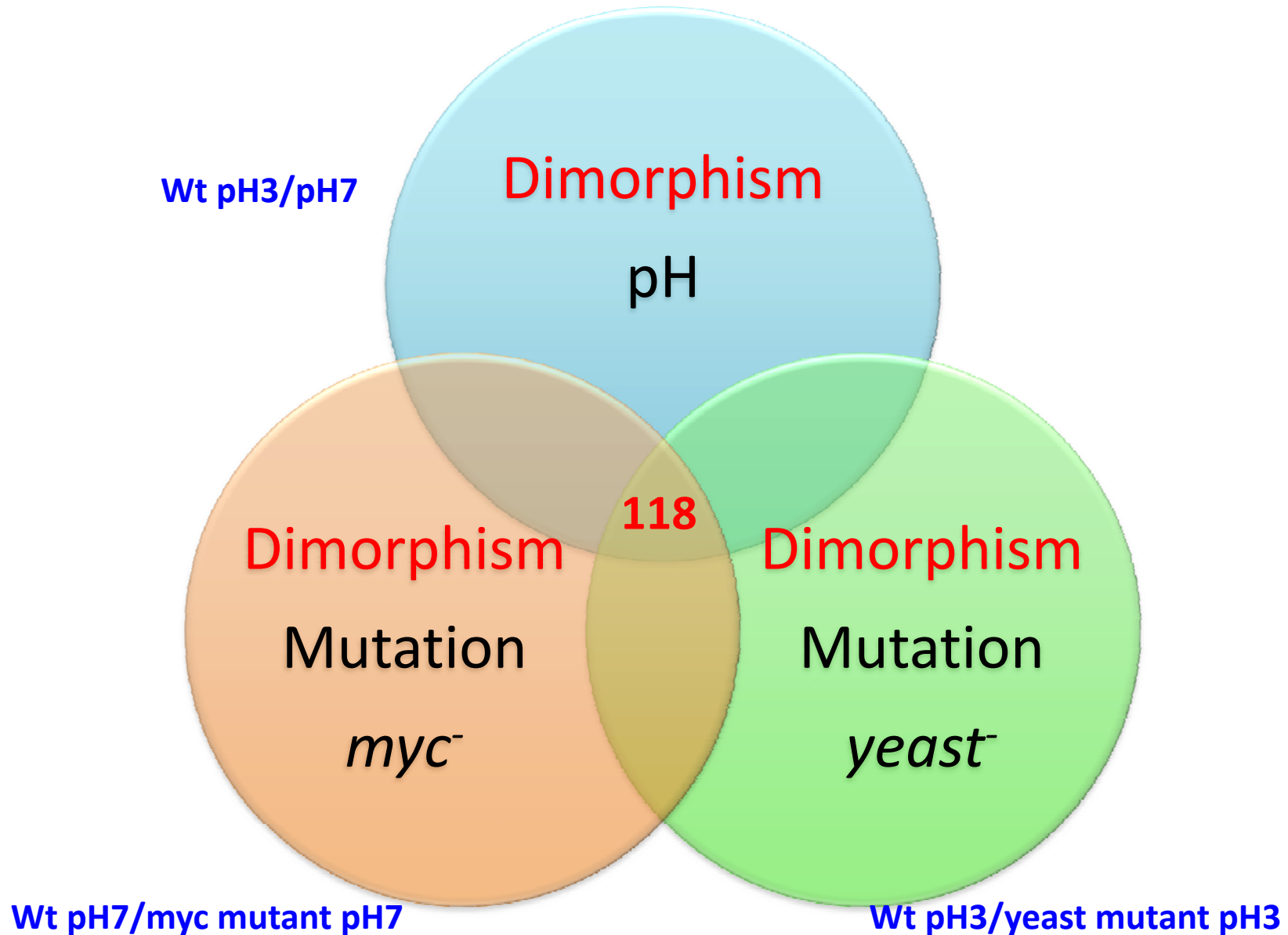
CEPA	GENOTIPO	FENOTIPO	MICELIO (%)
D12	<i>a1b1/a2b2</i>	DIPLOIDE	98
FB1	<i>a1b1</i>	HAPLOIDE	74
FB2	<i>a2b2</i>	HAPLOIDE	78
RK1662	<i>a1bE2</i>	MUTANTE EN <i>bW</i>	81
RK1723	<i>a1bW2</i>	MUTANTE EN <i>bW</i>	80
RK1607	<i>a1bE1</i>	MUTANTE EN <i>bE</i>	76
RK1725	<i>a1bW1</i>	MUTANTE EN <i>bW</i>	79
RK1447	<i>a1b</i>	MUTANTE TOTAL EN <i>b</i>	81



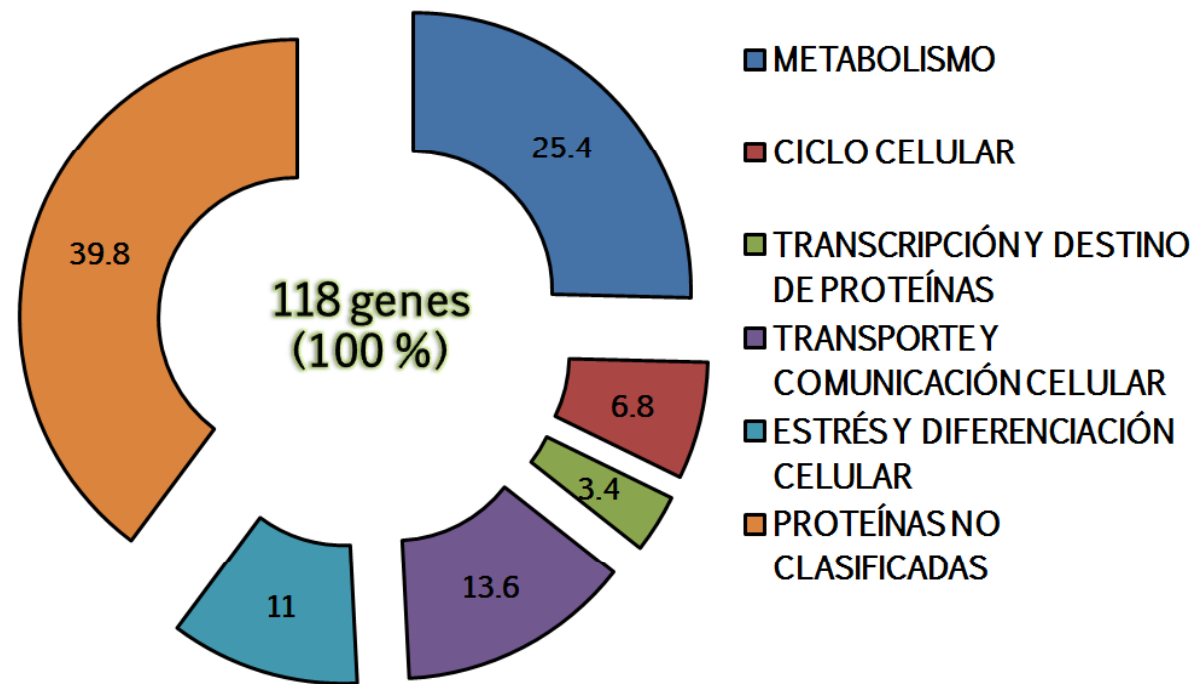
# ANALYSIS OF GENE EXPRESSION BY USE OF MICROARRAYS



# How to identify dimorphism specific genes



# AGRUPACIÓN FUNCIONAL DE LOS GENES DE *Ustilago maydis* ESPECÍFICOS DEL DIMORFISMO *in vitro* DIFERENCIALMENTE EXPRESADOS



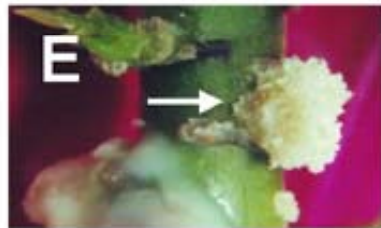
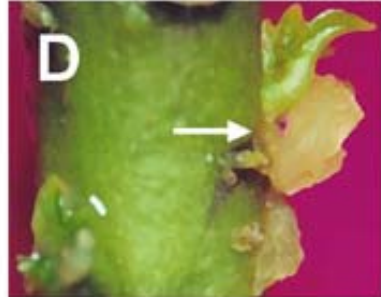


## PLANTS EXPERIMENTALLY INOCULATED WITH *U. maydis*

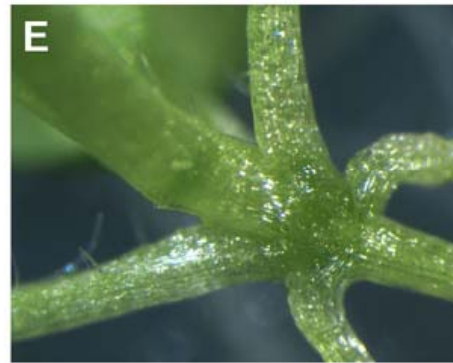
TAXONOMIC GRUP	SCIENTIFIC NAME	COMMON NAME
ANGIOSPERMS	<i>Ginkgo biloba</i>	Ginkgo
DICOTS	<i>Carica papaya</i>	Papaya
	<i>Solanum tuberosum</i>	Potato
	<i>Nicotiana tabacum</i>	Tobbaco
	<i>Saintpaulea sp</i>	African Violet
	<i>Phaseolus vulgaris</i>	Bean
	<i>Arabidopsis thaliana</i>	Arabidopsis
MONOCOTS	<i>Zea mays</i>	Maize
	<i>Allium sativum</i>	Garlic
	<i>Asparagus officinalis</i>	Asparagus
	<i>Sorghum bicolor</i>	Sorghum

## SYMPTOMS IN HOSTS

MYCELIAL GROWTH, CHLOROSIS, ANTHOCYANINS, NECROSIS, DWARFING. ROOT INCREASE, FLOWERING ACCELERATION, TUMORS

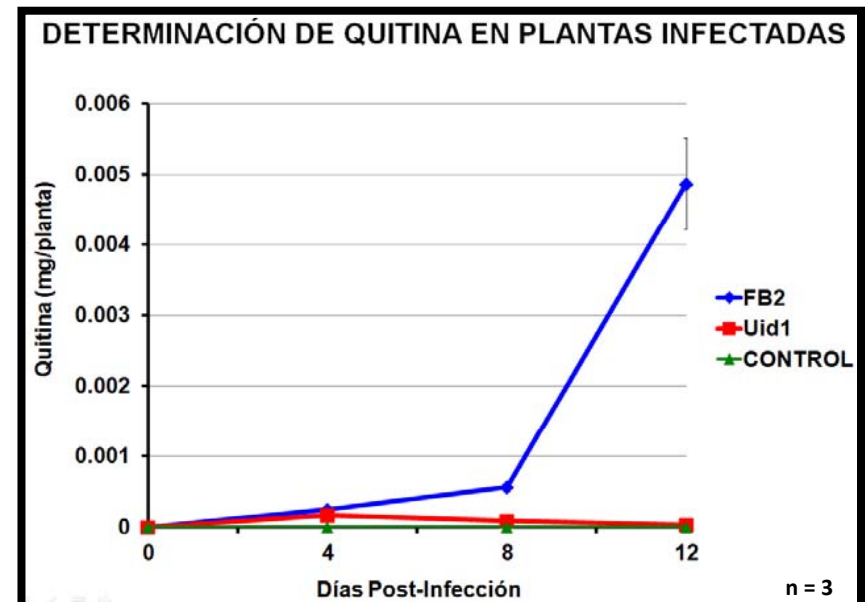
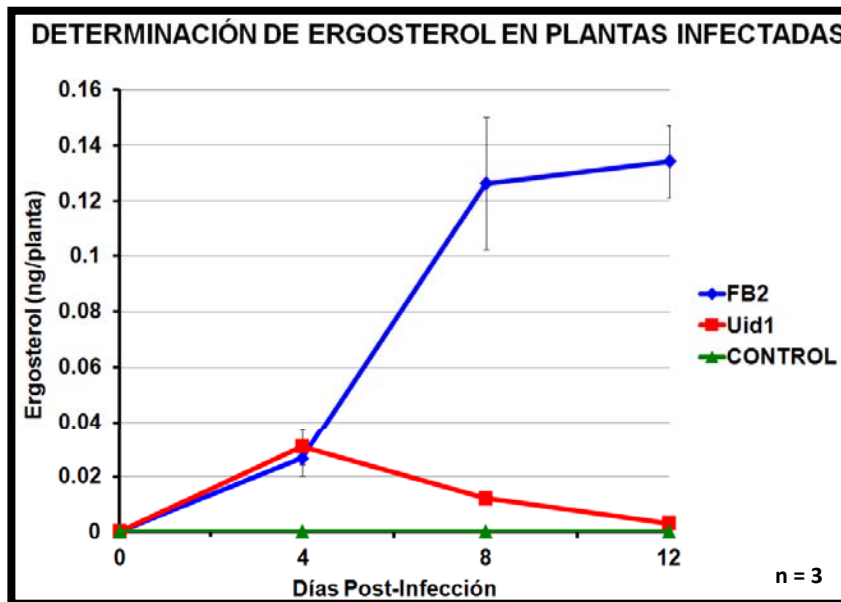
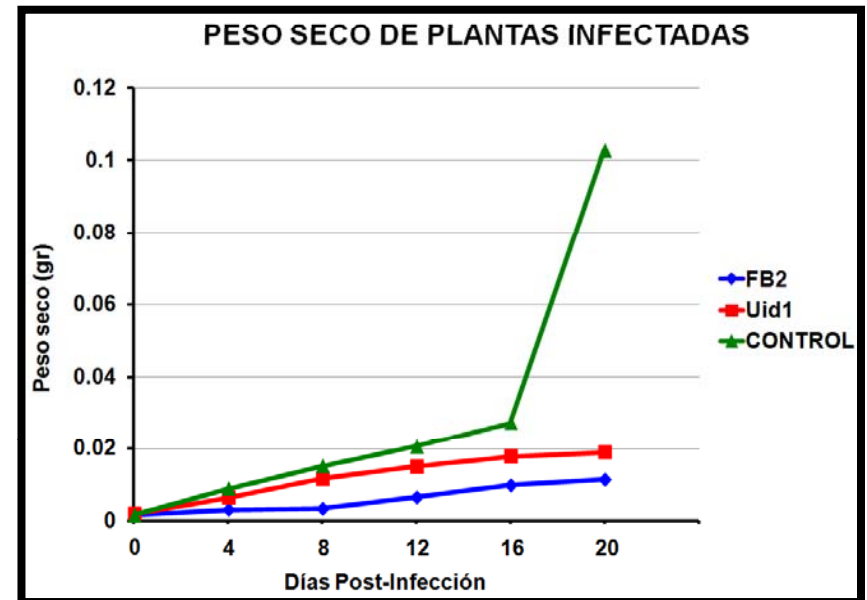
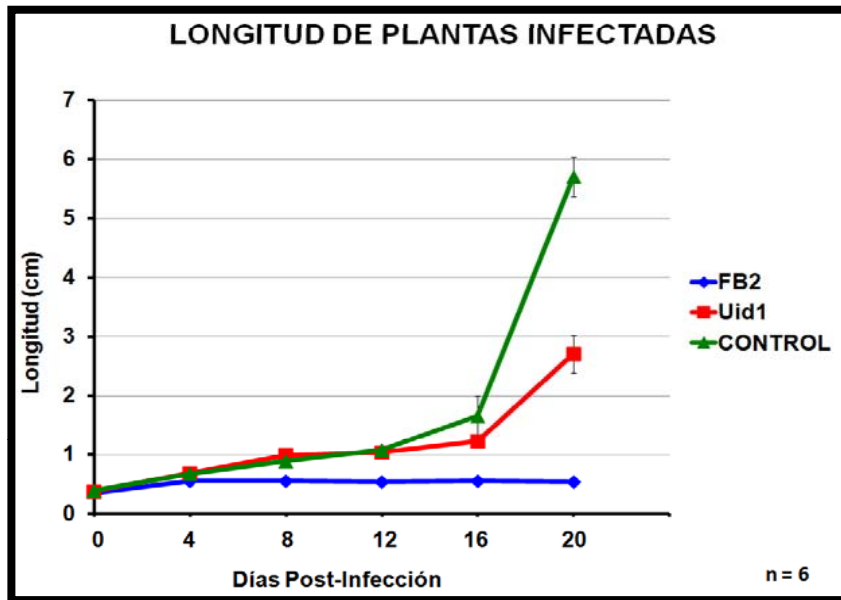


PAPAYA



ARABIDOPSIS

# Desarrollo de plantas infectadas y crecimiento de *U. maydis* dentro del tejido de *A. thaliana*

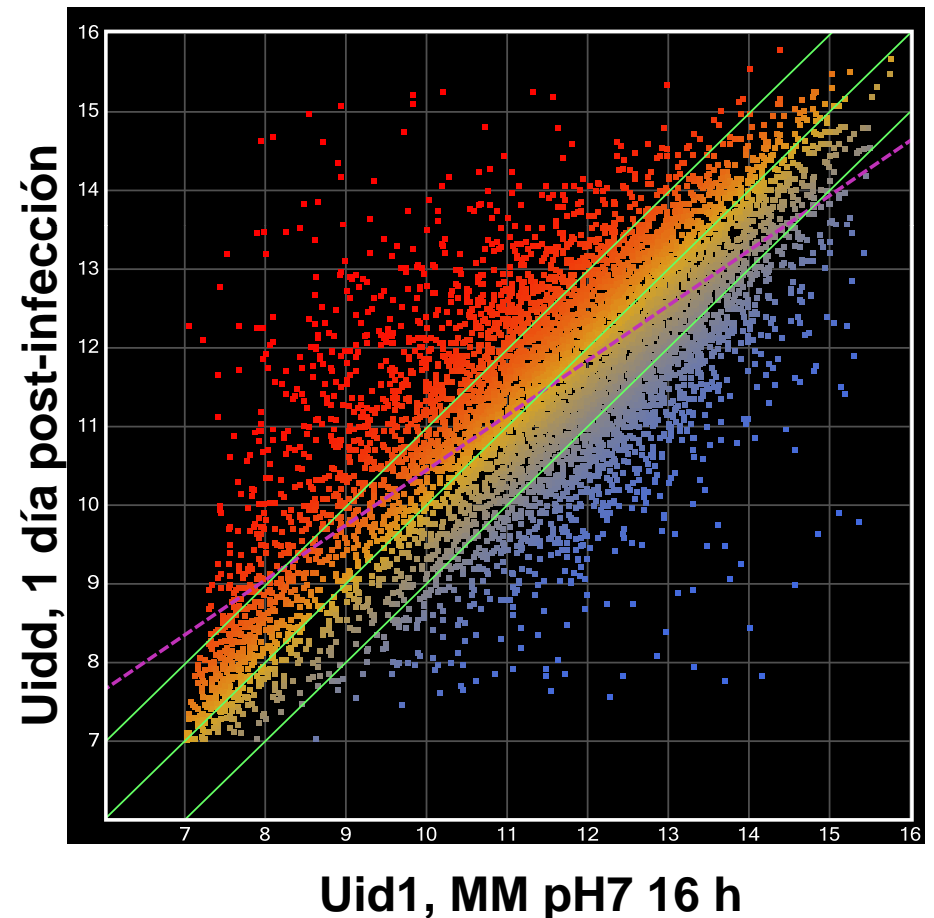
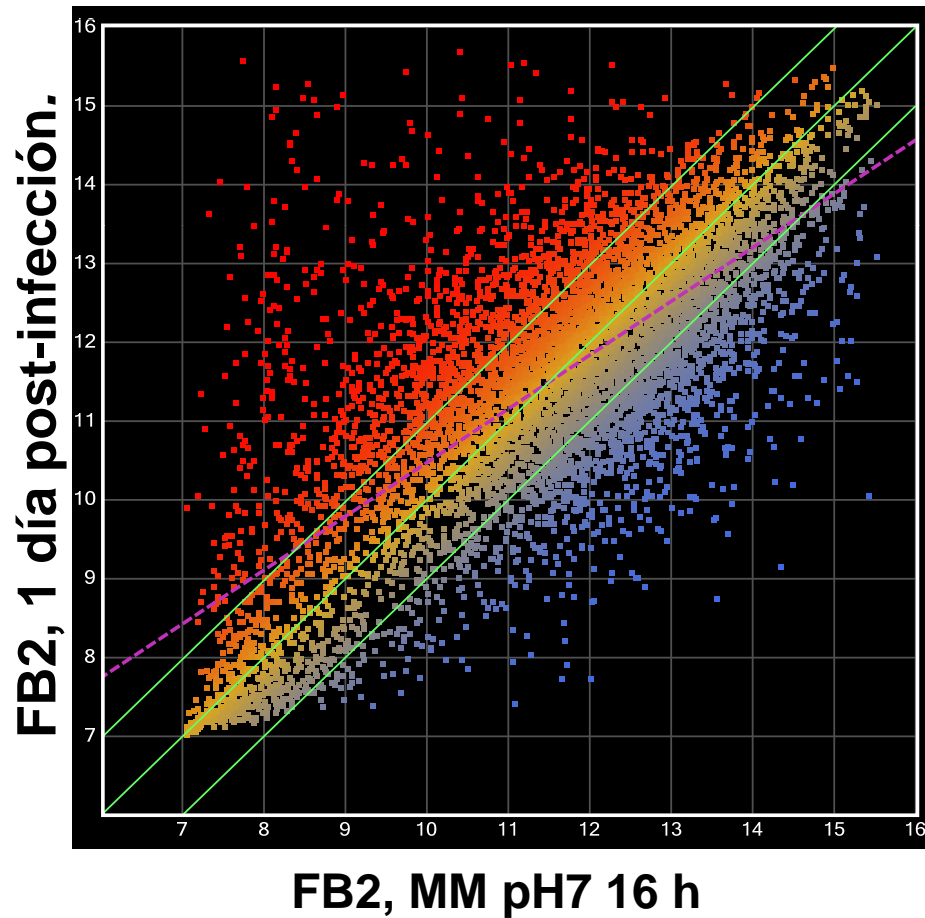




**CAMBIO DE EXPRESIÓN DE GENES DE *Ustilago maydis* EN LA TRANSICIÓN DE VIDA SAPRÓFITA A PARÁSITA EN *Arabidopsis thaliana***

**HAPLOIDE**

**DIPLOIDE**



## Genes diferenciales en *Ustilago maydis* durante su infección en *Arabidopsis thaliana*

Cepa Haploide de <i>U. maydis</i>	Días Post-infección de la cepa FB2 de <i>U. maydis</i> en <i>A. thaliana</i>			
	1	2	4	8
Genes diferenciales	2637	2317	2504	2322
<b>Sobre-expresados</b>	<b>1295</b>	<b>1134</b>	<b>1217</b>	<b>1154</b>
<b>Reprimidos</b>	<b>1342</b>	<b>1183</b>	<b>1287</b>	<b>1168</b>

Cepa Diploide de <i>U. maydis</i>	Días Post-infección de la cepa Uid1 de <i>U. maydis</i> en <i>A. thaliana</i>			
	1	2	4	8
Genes diferenciales	2390	2421	2164	2163
<b>Sobre-expresados</b>	<b>1171</b>	<b>1204</b>	<b>1086</b>	<b>1063</b>
<b>Reprimidos</b>	<b>1219</b>	<b>1217</b>	<b>1078</b>	<b>1100</b>

## EXPRESIÓN DE GENES REGULADOS POR EL HETERODÍMERO bE/bW DURANTE LA INFECCIÓN POR *U. maydis*

Genes diferencialmente regulados por el heterodímero en la cepa diploide  
Ninguno en la cepa haploide

Gene		Description	Fold Change
<b>um03172</b>	<b>Rbf1</b>	<b>Related to Zinc finger protein</b>	<b>2.310 up</b>
um02331	Kpp6	Kpp6 - MAP kinase	15.332 up
um03541		Related to Meiotic expression upregulated protein 26	3.218 up
um04654		Related to U2 snRNP protein A`	2.057 up
um02104		Probable Regulator of G-Protein Signaling Protein	2.586 up
um01008	ADN polimerasa epsilon	Probable POL2 - DNA polymerase epsilon, catalytic subunit A	3.895 down
um03758	Clb1	Clb1 - B-type cyclin 1	4.337 down
um04791	Cln1	Related to G1/S-specific cyclin	38.484 down

# Genes de *U. maydis* codificantes de enzimas degradadoras de la pared celular de plantas

33 genes descritos en el genoma de *U. maydis* (2006)

## Genes expresados diferencialmente solo en la cepa haploide (FB2)

Gen	Descripción	Nivel de expresión
um03411	probable endo-1,4-beta-xylanase	3.058 up
um04816	related to Endoglucanase 1 precursor	2.984 up
um02356	related to beta-galactosidase precursor	3.815 up
um03416	Crg1 - carbon source-regulated protein (putative arabinase)	9.944 up
um06075	related to beta-glucosidase	2.077 up

## Genes expresados diferencialmente solo en la cepa diploide (Uid1)

Gen	Descripción	Nivel de expresión
um10671	related to Pectin lyase B precursor	2.721 down
um00235	probable EXG1 - exo-beta-1,3-glucanase (I/II), major isoform	5.705 up



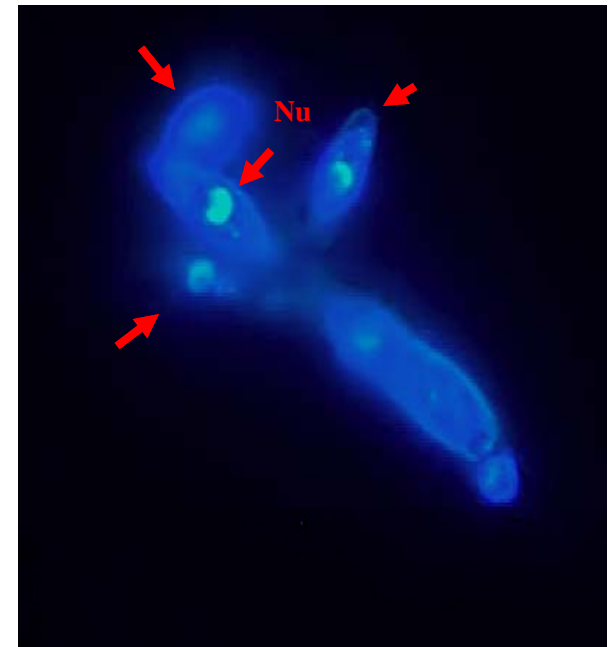
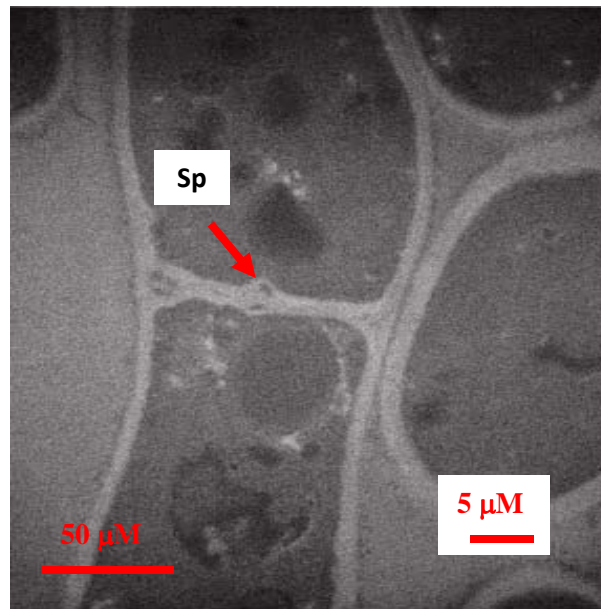
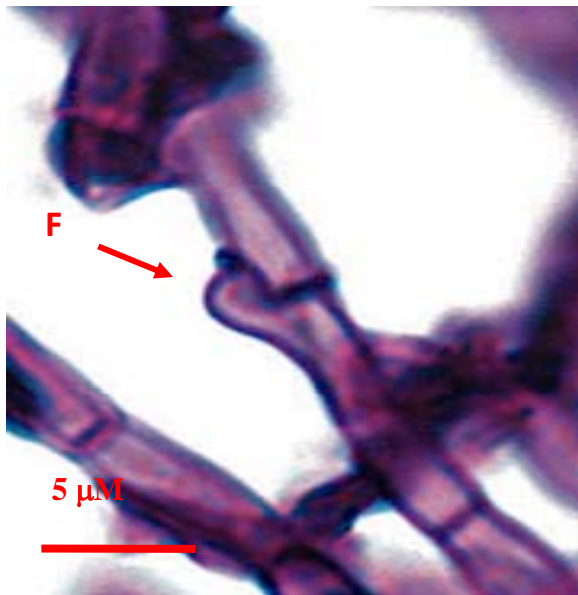
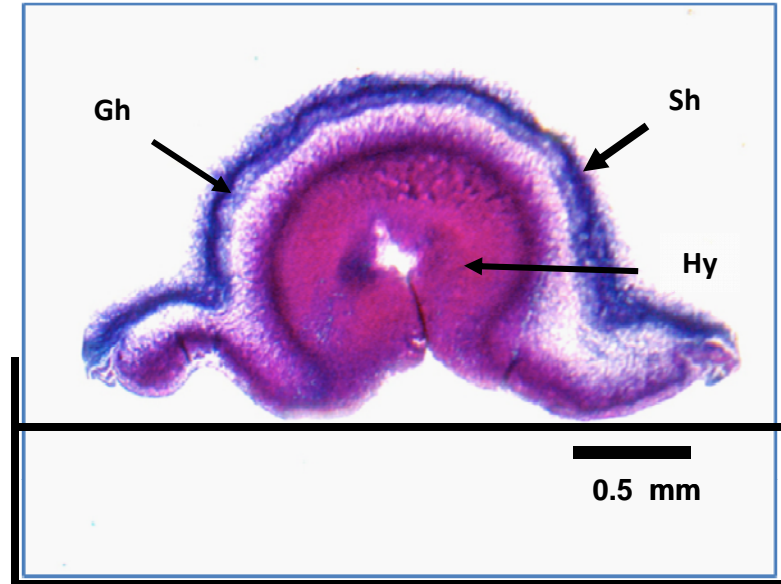
28 12 2003

**SIMPLE KEY TO THE ORDERS OF THE SUB-CLASS  
HETEROBASIDIOMYCETIDAE**

- A. Basidiocarp usually well developed; mostly saprobic, some species parasitic on plants or scale insects *Tremellales*
- AA. Basidiocarp lacking or poorly developed; mostly parasitic on vascular plants
- B. Teleutospores present; plant parasites
- C. Basidiospores produced on sterigmata; forcibly discharged *Uredinales*
- CC. Basidiospores sessile; not forcibly discharged *Ustilaginales*
- BB. Teleutospores lacking; resting spores may be present; saprobic Family *Sporobolomycetaceae*<sup>1</sup>

<sup>1</sup> A family of Basidiomycetes of uncertain affinity which cannot be included at present in any of the existing orders.

# Formation of fruiting bodies









*Zea mays*  
"Corn"  
\$0.40 Dls



*Claviceps gigantea*  
(Ascomycota)  
"Horse Tooth"  
\$ 0.00



*Sporisorium reilianum*  
(Basidiomycota)  
"head smut"  
\$ 0.00



*Ustilago maydis*  
(Basidiomycota)  
"HUITLACOCHÉ"  
\$2.40 Dls

# ¿ QUE ES LA CIENCIA?

**"CIENCIA ES EL CONOCIMIENTO CIERTO DE LAS COSAS POR SUS PRINCIPIOS Y CAUSAS"**

Diccionario de la Lengua Española

**"CIENCIA ES EL CUERPO DE DOCTRINA METODICAMENTE FORMADO Y ORDENADO QUE CONSTITUYE UNA RAMA PARTICULAR DEL HUMANO SABER"**

Id

**"LA CIENCIA ES UNA FORMA DE EXPLICAR LA REALIDAD SIN RECURRIR A DOGMAS, MILAGROS O AL PRINCIPIO DE AUTORIDAD"**

Marcelino Cerejido

NO HAY CIENCIA BASICA  
Y CIENCIA APLICADA.  
EXISTE TAN SOLO LA  
CIENCIA  
Y SUS APLICACIONES



LOUIS PASTEUR

The NIH will most assuredly continue its strong tradition of supporting basic research, which it defines as systematic study directed toward fuller knowledge or understanding of the fundamental aspects of phenomena and of observable facts without specific applications in mind. Since 2003, the proportion of NIH funds spent on basic research, defined in this way, has ranged from 53 to 57%, standing at 54% for fiscal year (FY) 2012.

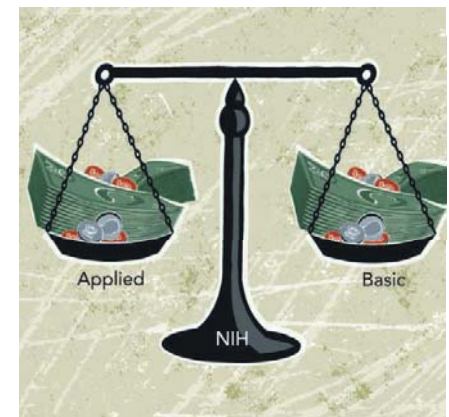
Basic research also accounts for most of the 135 Nobel Prizes won by NIH-supported scientists, including the 2011 awards to Bruce Beutler and Jules Hoffmann for their discoveries about innate immunity, and the late Ralph Steinman for adaptive immunity.

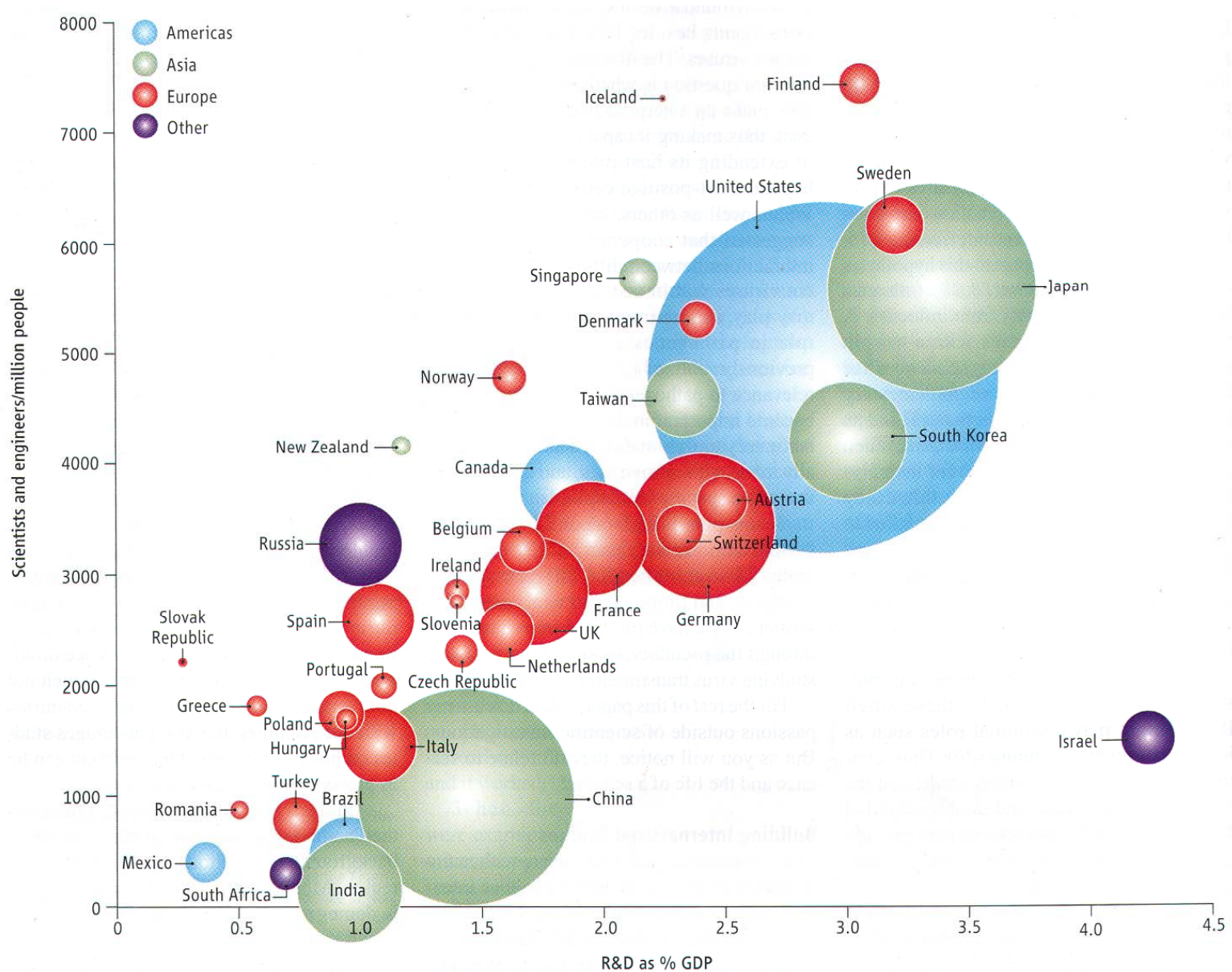
In this time of severe budget constraints, Americans need to know that **today's basic research is the engine that powers tomorrow's therapeutic discoveries**. They need to know that basic research is the type of science that the private sector, which requires rapid returns on investment, cannot afford to fund. **They need to know that, because it is impossible to predict whence the next treatment may emerge, the nation must support a broad portfolio of basic research.**



Francis S. Collins is director of the U.S. National Institutes of Health, Bethesda, MD.

SCIENCE VOL 337 AUGUST 3, 2012





**Fig. 4.** World of R&D in 2010. Size of circle reflects the relative amount of annual R&D spending by the country noted. [Reproduced by permission of *R&D Magazine* (28)]

# LA CARRERA EN EL AVANCE DE LA CIENCIA

“Don’t you come back into my office asking for more money until 2010.” Brazilian president Luiz Inácio Lula da Silva’s playful admonishment to his science minister came after the president’s announcement last week of a remarkable US\$28-billion package for science and technology over the next three years. The spending promised is equivalent to 1.5% of the country’s GDP — currently, science receives about 1% of GDP.



**PAÍSES DESARROLLADOS**

n absence  
Brazil is  
of all the  
ally each  
07 report  
economic  
pment. To  
ent’s new  
ensive  
for

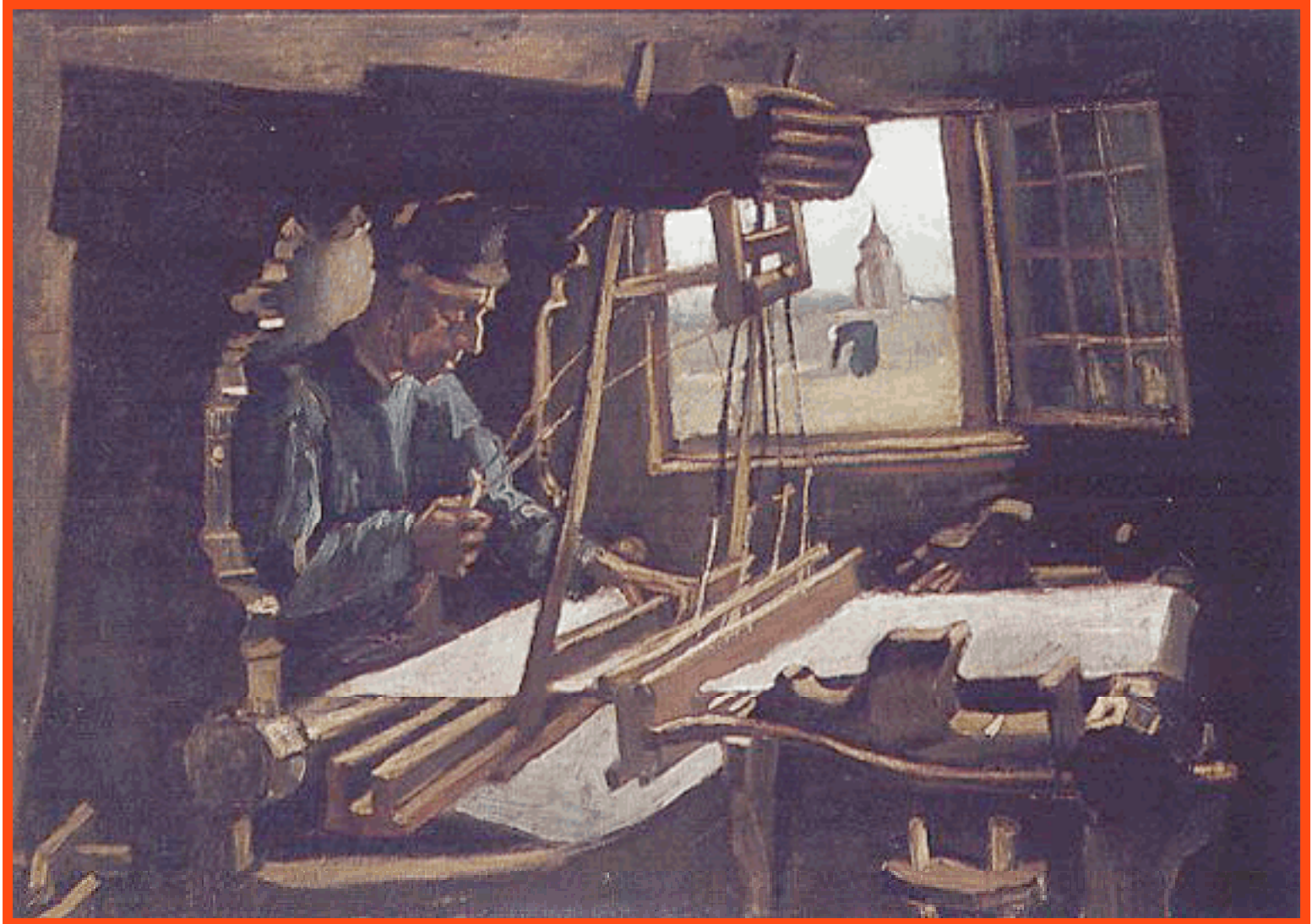


research  
get it th  
three t  
says. “  
compe  
No  
warn  
duri:  
ca  
s  
s



**MÉXICO**

## EL CIENTÍFICO COMO TEJEDOR DE HIPÓTESIS Y EXPERIMENTOS



**¡GRACIAS POR SU ATENCIÓN!**