PROGRAMA DE PÓS GRADUAÇÃO
EM ALIMENTOS E NUTRIÇÃO
O SEMEAR DA CIÊNCIA

III SIAN *Simpósio de Alimentos e Nutrição*

INTRODUCCIÓN AL MEJORAMIENTO GENÉTICO DE VEGETALES

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UNIVERSIDAD DE LOS LAGOS

FITOGEN
PROGRAMA DE CONSERVACIÓN, MANEJO Y UTILIZACIÓN SUSTENTABLE DE LOS RECURSOS GENÉTICOS VEGETALES.

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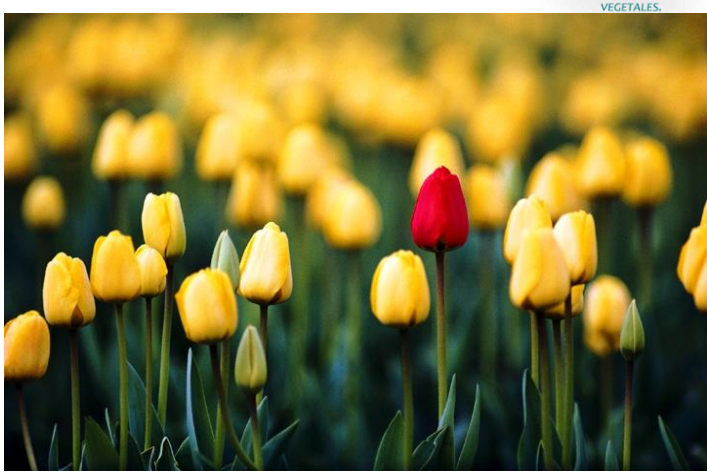
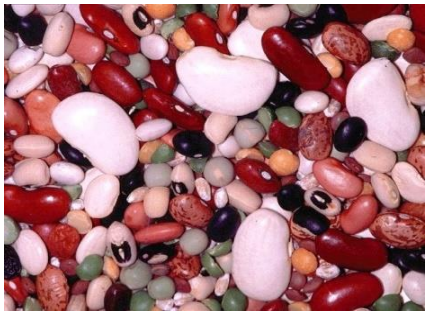
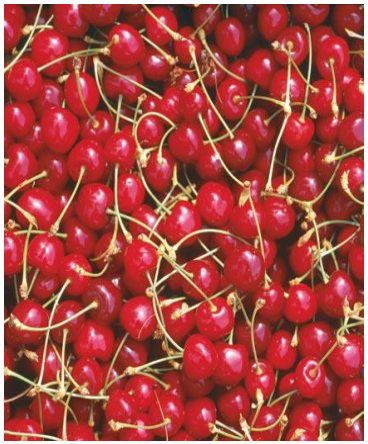







MEJORAMIENTO GENÉTICO

Es la aceleración del proceso evolutivo natural de las especies, con el objetivo de crear nuevas variedades que tengan ventajas para el ser humano y los animales domésticos.

FITOMEJORAMIENTO

MEJORAMIENTO GENÉTICO APLICADO A LAS PLANTAS

Es **arte y la ciencia** de cambiar genéticamente las plantas (Allard, 1967)

FITOMEJORAMIENTO

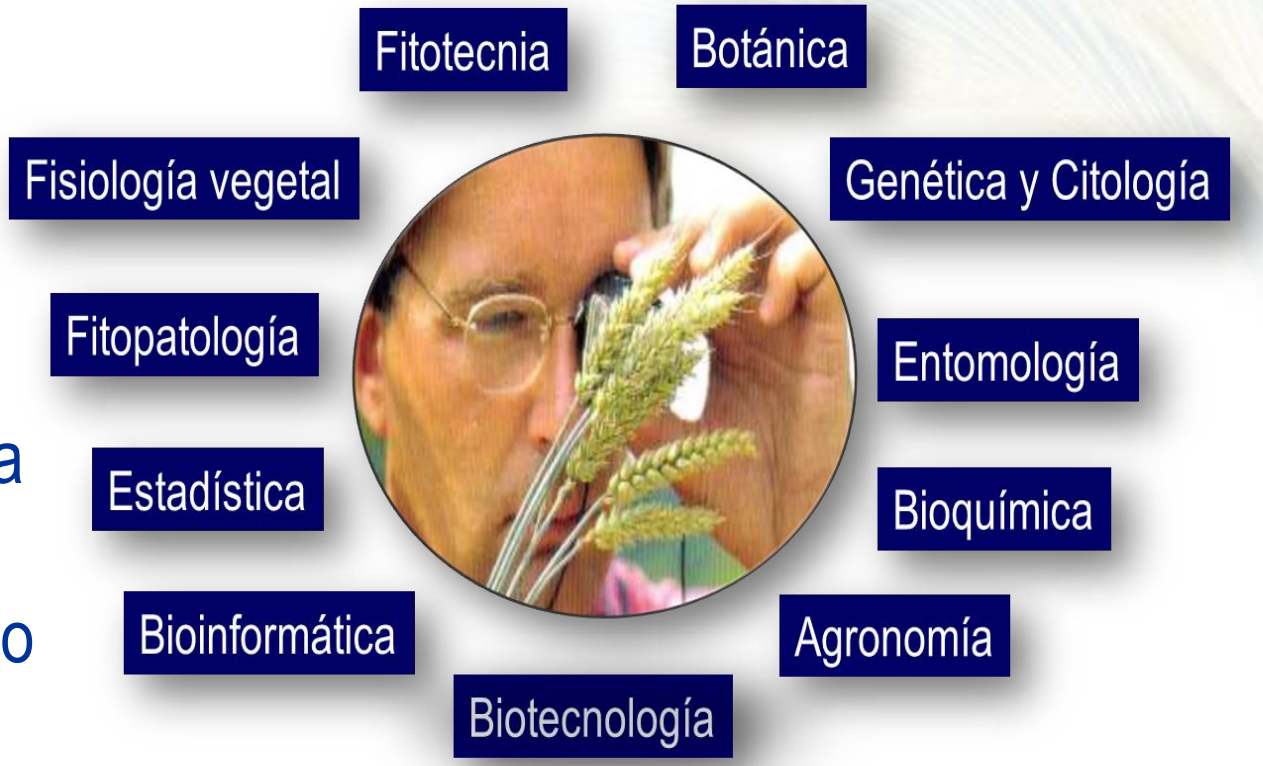
Es **la ciencia, el arte y el negocio** de mejorar las plantas para beneficio humano (Bernardo, 2002)

FITOMEJORAMIENTO

Es un conjunto de principios científicos, métodos, técnicas y estrategias que se aplican para la obtención de genotipos o grupos de genotipos de plantas con características deseables según una serie de objetivos previamente definidos.

FITOMEJORADOR DE HOY

"Un científico que utiliza un sistema organizado de procedimientos y metodologías de manipulación genética para modificar una especie con el objetivo de hacerla más útil para un determinado fin"



PREMISAS PARA LA PLANIFICACIÓN DE UN PROGRAMA DE FITOMEJORAMIENTO

- La existencia de variabilidad o la posibilidad de crearla.
- La capacidad o habilidad de detectar la variabilidad.
- La capacidad de manipular la variabilidad para el desarrollo de una nueva variedad.

OBTENCIÓN DE VARIABILIDAD

Mutaciones, Recombinación, Transferencia horizontal, Selección, Azar

OBJETIVOS DEL FITOMEJORAMIENTO

1. INCREMENTO EN LA PRODUCCIÓN

- Mayor eficiencia fotosintética
- Mayor adaptación
- Mejores caraterísticas agronómica
- Resistencia a enfermedades y plagas
- Resistencia a factores adversos

2. INCREMENTO EN LA CALIDAD

- Alto valor nutritivo
- Mejor sabor, tamaño, color, otros
- Resistencia al transporte y almacenaje
- Reducción de sustancia indeseables

EL ROMPECABEZAS DEL FITOMEJORAMIENTO

CARACTERÍSTICAS GENERALES DEL FITOMEJORAMIENTO

- **ACUMULATIVO**
  - Las nuevas mejoras se deben sumar a las características anteriores.
- **CONTINUO**
  - Cambios de hábitat, técnicas de cultivo, en los mercados, las poblaciones de patógenos.
- **COMPETITIVO**
  - Las nuevas variedades deben superar a las anteriores (ensayos comparativos de rendimiento).

CONSIDERACIONES SOBRE LA ESPECIE Y EL CARACTER A MEJORAR

- Modo reproductivo (autógamo, alógamo o de reproducción vegetativa).
- Cantidad de genes y su momento de expresión.
- Tiempo por generación.
- Número de cromosomas y nivel de ploidia.
- Otros: condiciones de crecimiento, resistencia a enfermedades, etc.

ETAPAS DEL FITOMEJORAMIENTO

ETAPAS DEL FITOMEJORAMIENTO

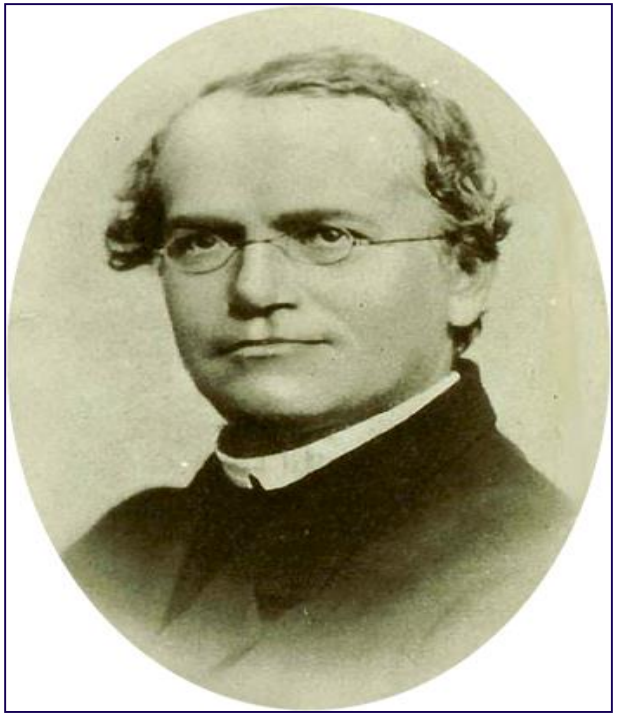
1. Descubrimiento y obtención de variabilidad genética.

ETAPAS DEL FITOMEJORAMIENTO

2. Combinación de la variabilidad disponible.

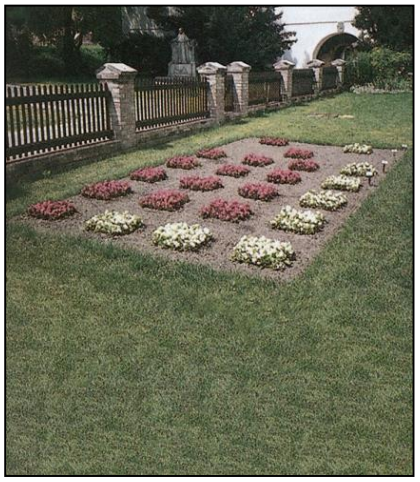
GENÉTICA CLÁSICA

Gregor Mendel
(1822-1884)



El padre de la Genética

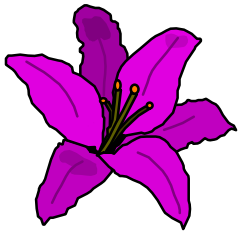
Monasterio Agustino de Santo Tomás de Brunn, actual república Checa



Mendel postula….

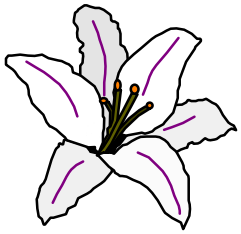
- Ley de la Segregación
- Ley de la Segregación Independiente

P generación



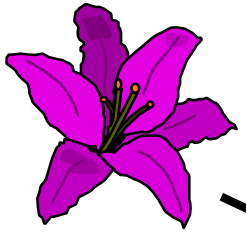
AA

X



aa

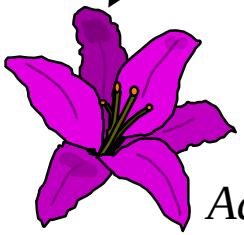
F1 generación



Aa

F2 generación

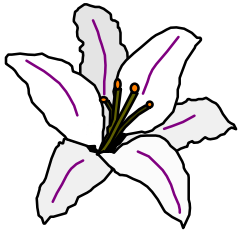
AA



Aa

Aa

aa



Fenotipo: 3:1
Genotipo: 1:2:1

LA SELECCIÓN DE LOS PADRES PARA LOS CRUZAMIENTOS…

DIFICULTAD EN LOS CRUZAMIENTOS (AVENA)

ETAPAS DEL FITOMEJORAMIENTO

2. Combinación de la variabilidad disponible.

ETAPAS DEL FITOMEJORAMIENTO

3. Selección de los individuos con caracteres de interés.

ETAPAS DEL FITOMEJORAMIENTO

3. Selección de los individuos con caracteres de interés.

RESISTENCIA A LA TENDIDURA

RESISTENCIA A LA TENDIDURA

ETAPAS DEL FITOMEJORAMIENTO

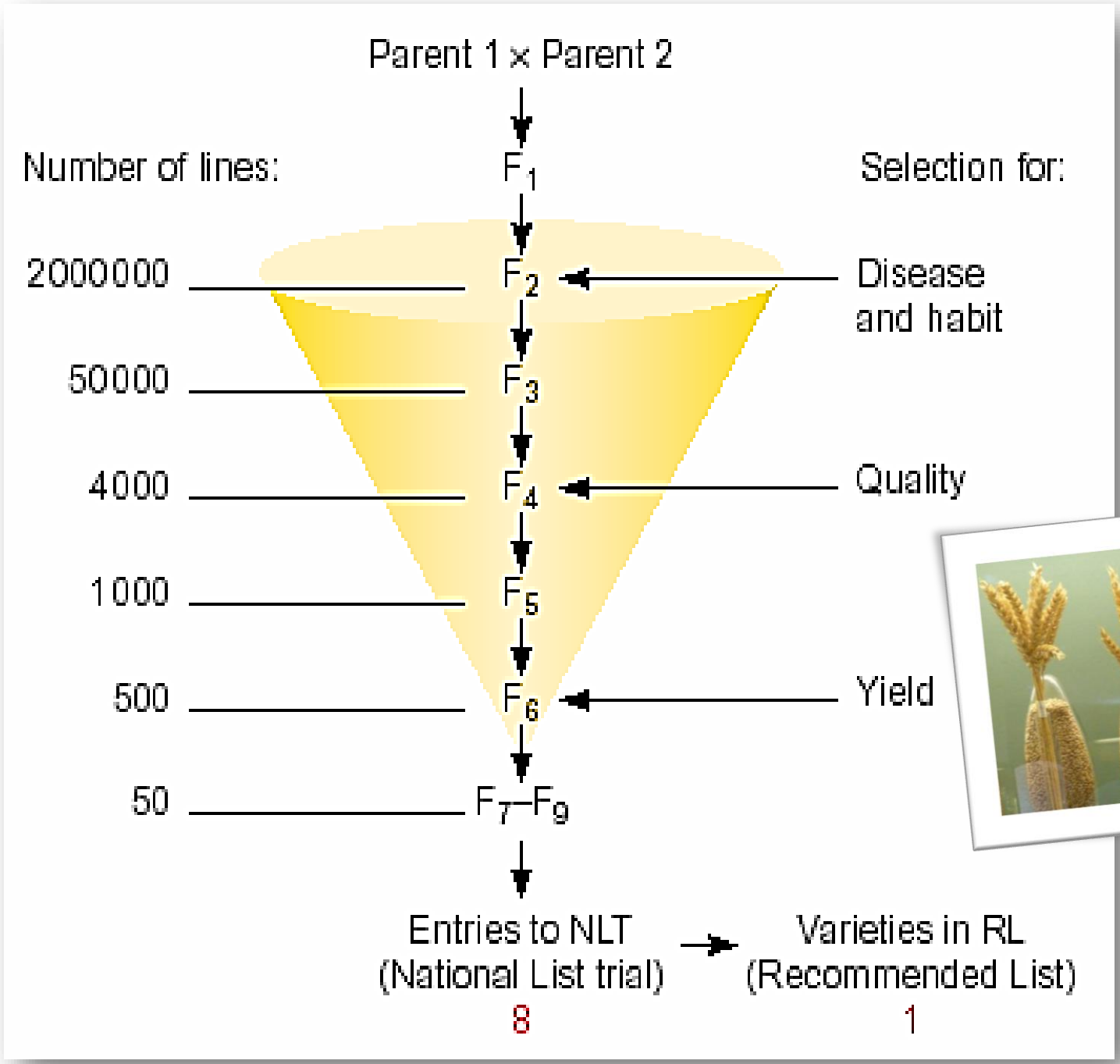
4. Estabilización fenotípica.

ETAPAS DEL FITOMEJORAMIENTO

5. Multiplicación del material seleccionado

ETAPAS DEL FITOMEJORAMIENTO

6. Distribución y comercialización de las nuevas variedades para uso de los agricultores


Parent 1 × Parent 2
Number of lines:
Selection for:
F1
2000000
F2
Disease and habit
50000
F3
4000
F4
Quality
1000
F5
500
F6
Yield
50
F7–F9
Entries to NLT (National List trial)
8
Varieties in RL (Recommended List)
1

MÉTODOS DE SELECCIÓN SEGÚN MODO REPRODUCTIVO

METODOS DE PLANTAS AUTÓGAMAS

- Selección individual
- Selección masal
- Hibridación
  - Pedigree
  - Masal
  - Retrocruzamiento
  - SSD

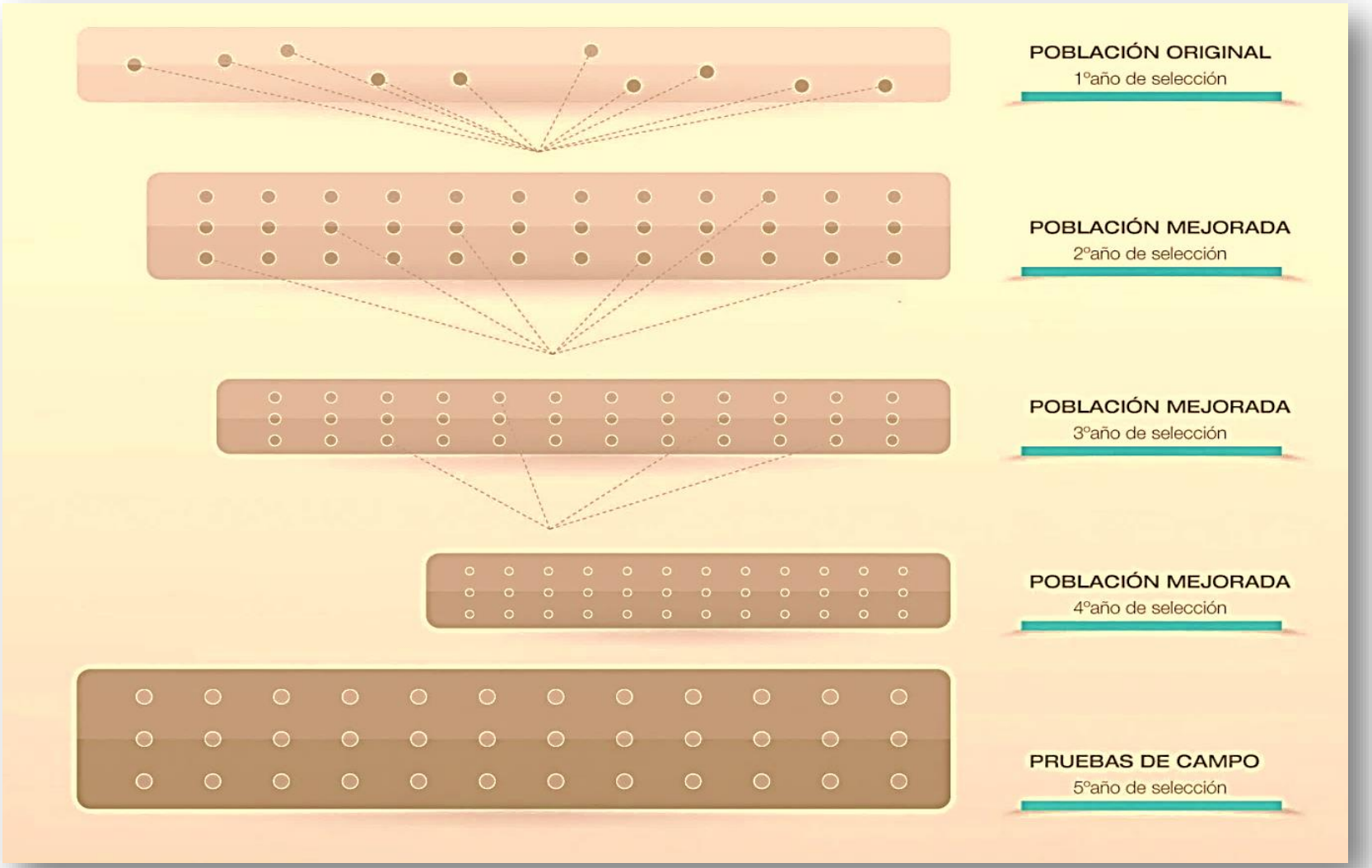
METODOS DE PLANTAS ALÓGAMAS

- Hibridación de líneas puras
- Selección masal
- Retrocruzamiento
- Selección recurrente

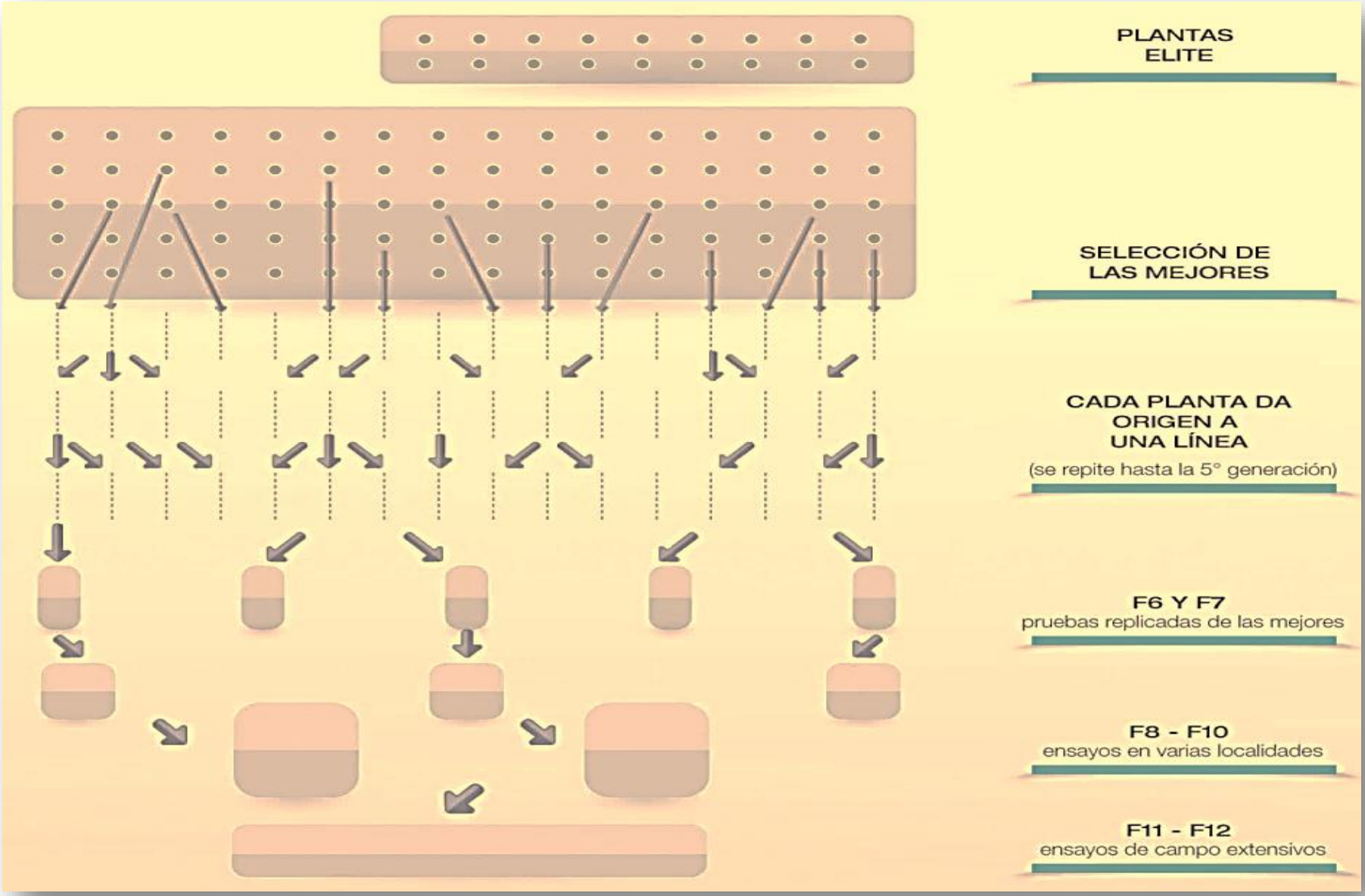
METODOS PARA ESPECIES CON REPRODUCCIÓN VEGETATIVA

- Cruzamientos
- Selección de variedades heterogéneas
- Mutaciones inducidas

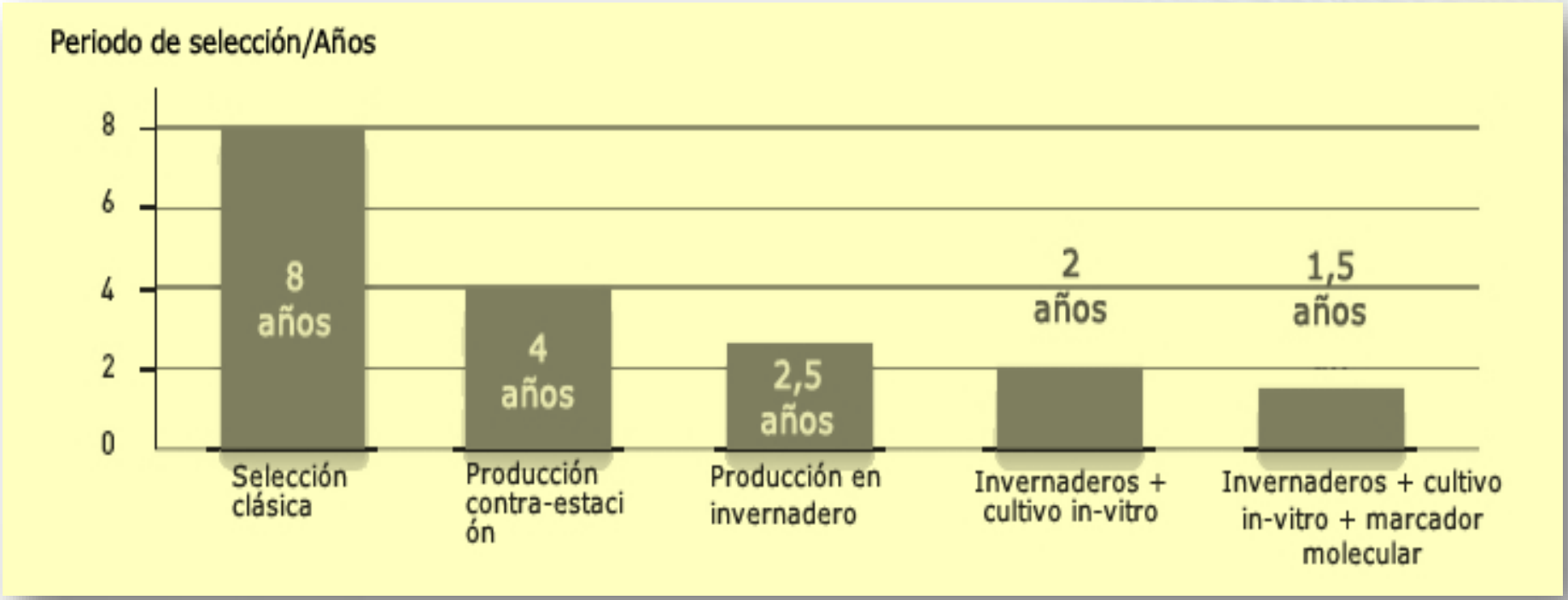
SELECCIÓN MASAL



MÉTODO PEDIGREE



REDUCIR EL PERIODO DE SELECCIÓN

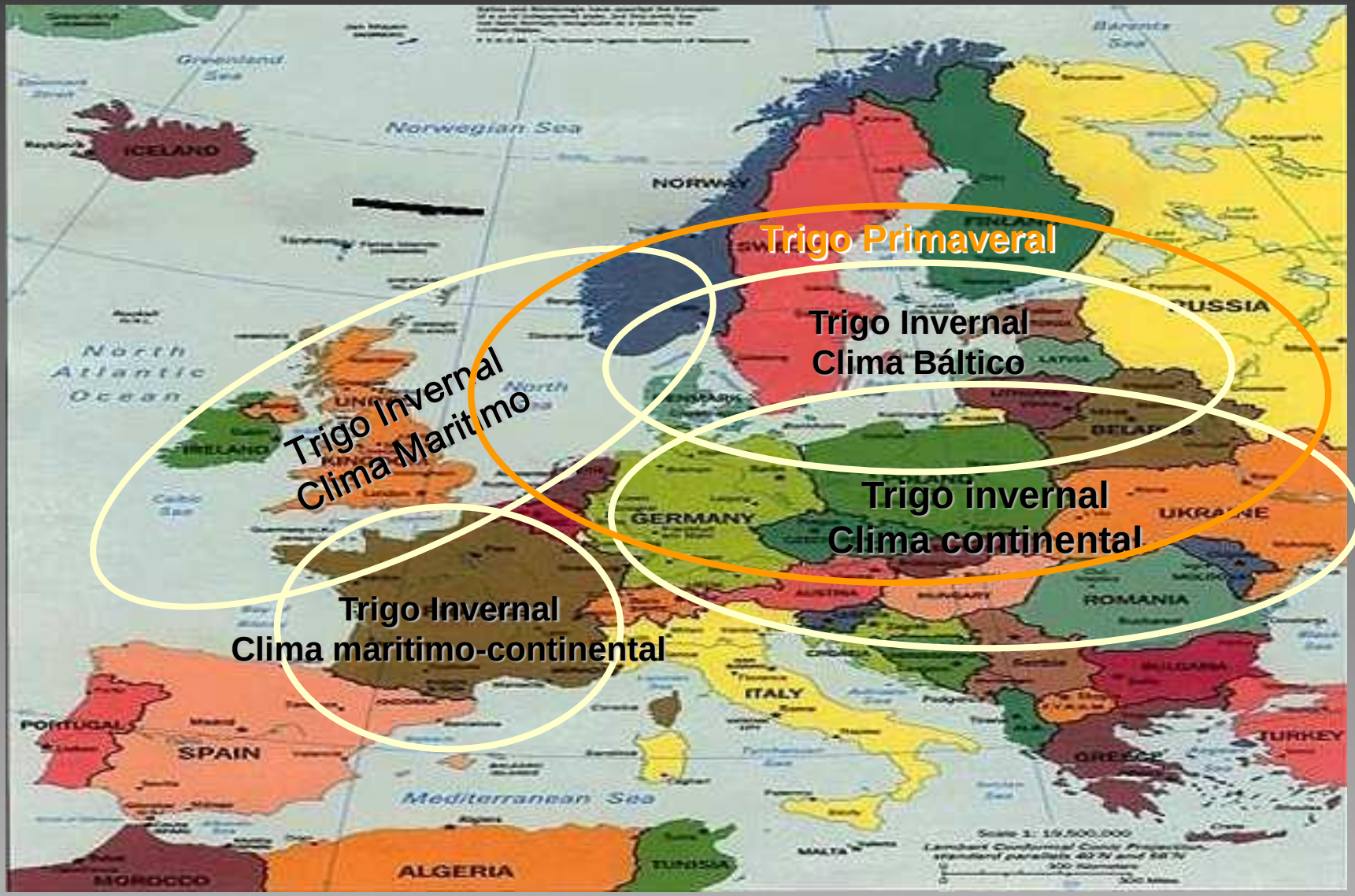


Cada día se buscan **nuevas herramientas** que permitan al fitomejorador ganar tiempo en los ciclos largos del proceso de selección. Además, se busca disminuir la **"toma de plantas al azar"** que hace el mejorador en el campo usando caracteres dependientes del medio ambiente.

PROGRAMA DE MEJORAMIENTO GENÉTICO EN TRIGO (SUECIA)

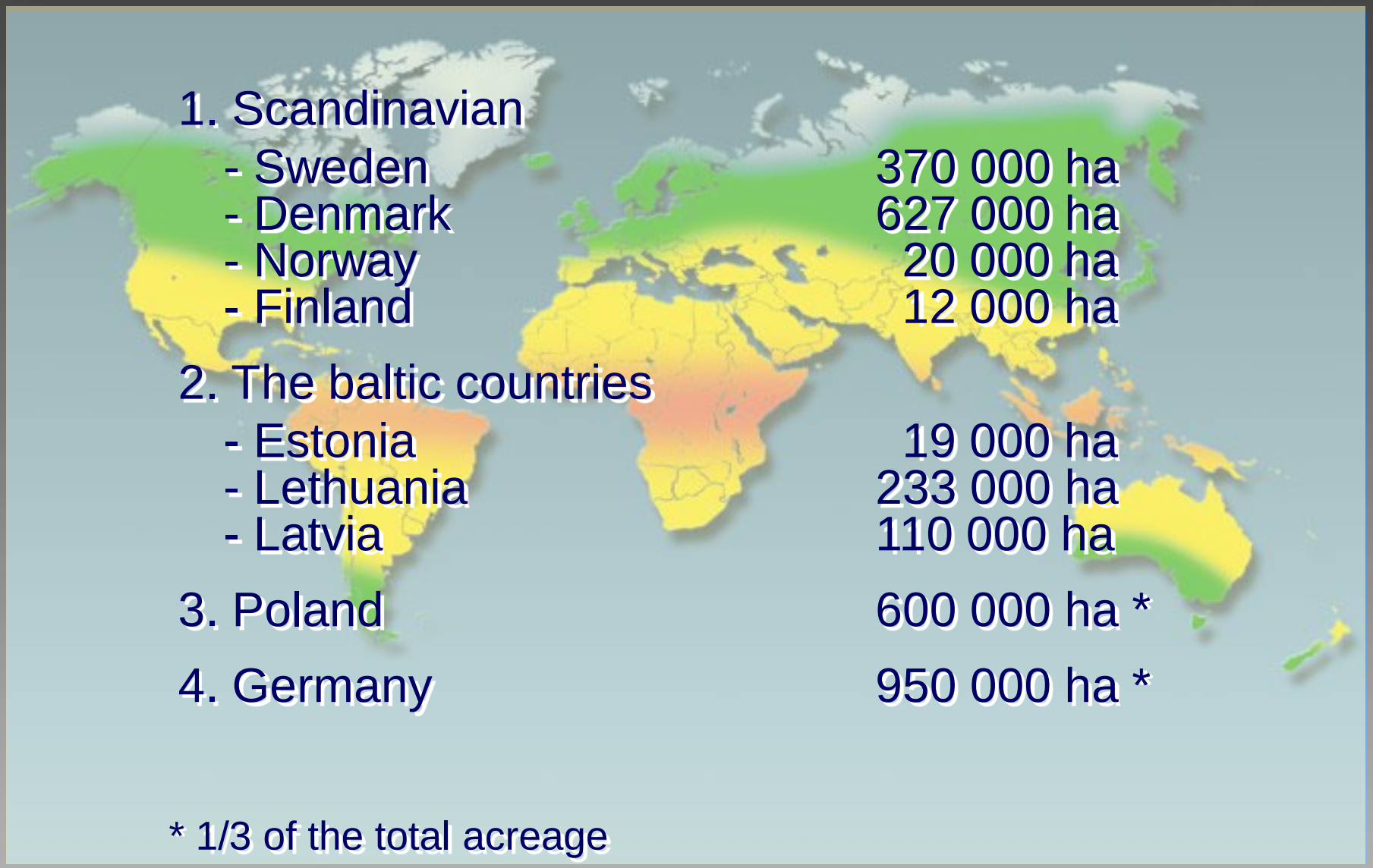
Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Programa de Mejoramiento Genético en Trigo (Suecia)



Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

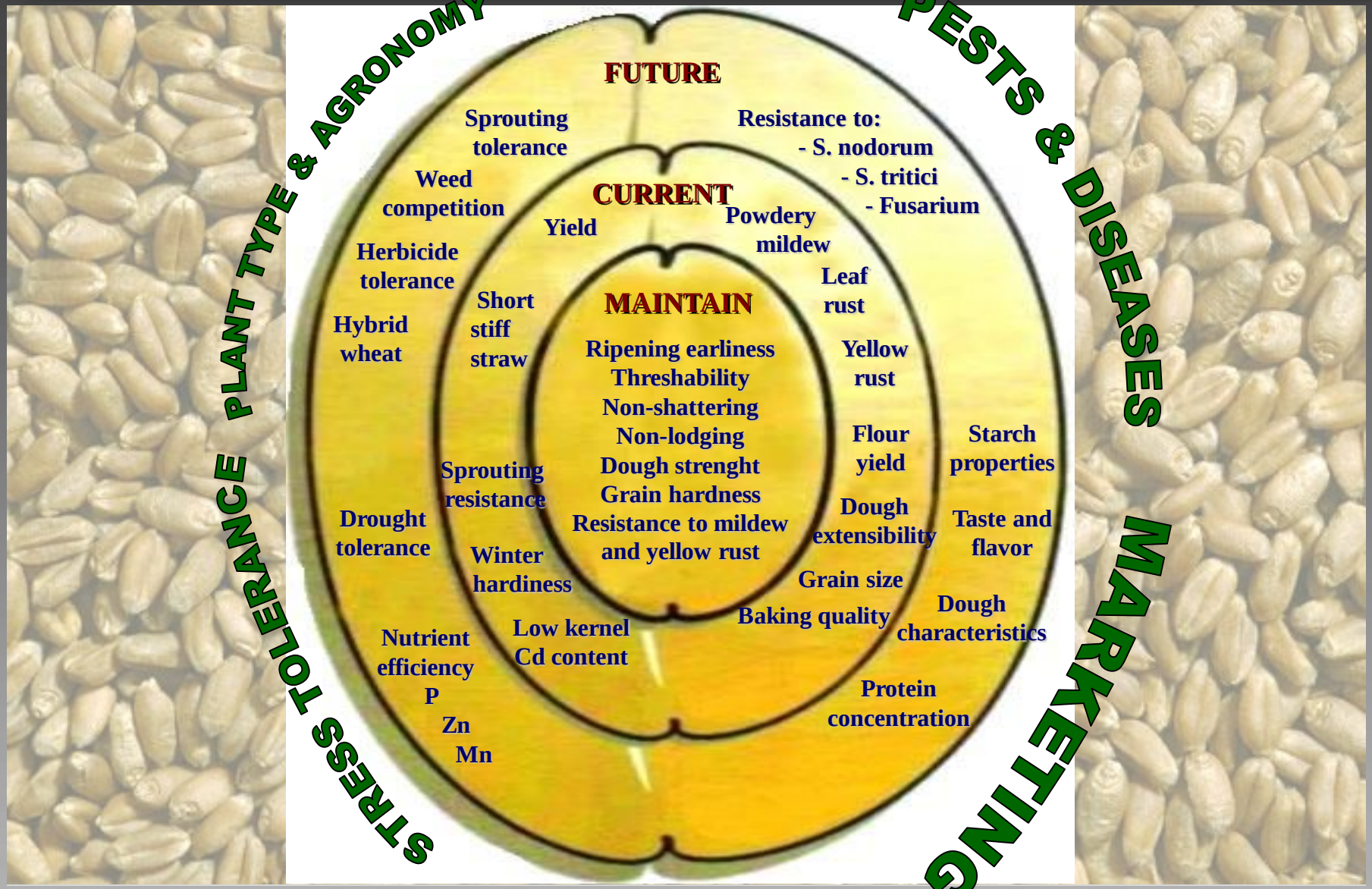
MARKET AND ACREAGE IN WINTER WHEAT



1. Scandinavian	
- Sweden	370 000 ha
- Denmark	627 000 ha
- Norway	20 000 ha
- Finland	12 000 ha
2. The baltic countries	
- Estonia	19 000 ha
- Lethuania	233 000 ha
- Latvia	110 000 ha
3. Poland	600 000 ha *
4. Germany	950 000 ha *

* 1/3 of the total acreage

Objetivos del Programa Mejoramiento en Trigo



Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Objetivos en *Triticum aestivum* (primaveral)

- Buena calidad de molienda del grano (harina).
- Altos niveles de proteina.
- Resistencia a la germinación en el campo.

- Alta producción y bajo tamaño de la caña.
- Madurez temprana (parta norte de Suecia)

Resistencia a las heladas/hielo

Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Amount of winter wheat in Landskrona

Type of material	Year 2001
• Conventional crosses (field + glasshouse)	500
• F1 hybrids	740
• Double Haploid material (No. of plants)	4 000
• F2 – F6	32 000
• Preliminary yield trails (PYT)	1 240
• Advanced yield trails (AYT) (incl. hybrids)	1 300

Cruzamientos Convencionales...

Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Enmasculación de los Machos en Trigo

Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Los mejoradores estudian cuidadosamente el material...

Oscar Díaz – Wheat Breeder – Svalöf Weibull 1999-2004

Variedades tempranas y tardías...

Oscar Díaz – Wheat Breeder – Svalöf Weibull 1999-2004

Selección de las líneas avanzadas (5-6 años)

Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Evaluación de la producción de una variedad

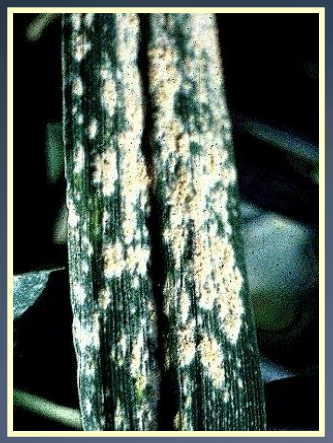
Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Resistance breeding in wheat

Objectives of resistance breeding in wheat

- Find ”new” genes conferring disease resistance.
- Transfer ”new” genes to adapted materials.
- Combine new and already known genes to get a durable resistance.
- Find and keep new pathogene isolates which are useful in the selection work.
- Develop and improve testing methods.
- Develop DNA markers for resistance genes (MAS).

Main target diseases of wheat in Sweden



Blumeria graminis

Puccinia triticina

Puccinia striiformis

Stagonospora nodorum

Septoria tritici

T. caries

T. contraversa

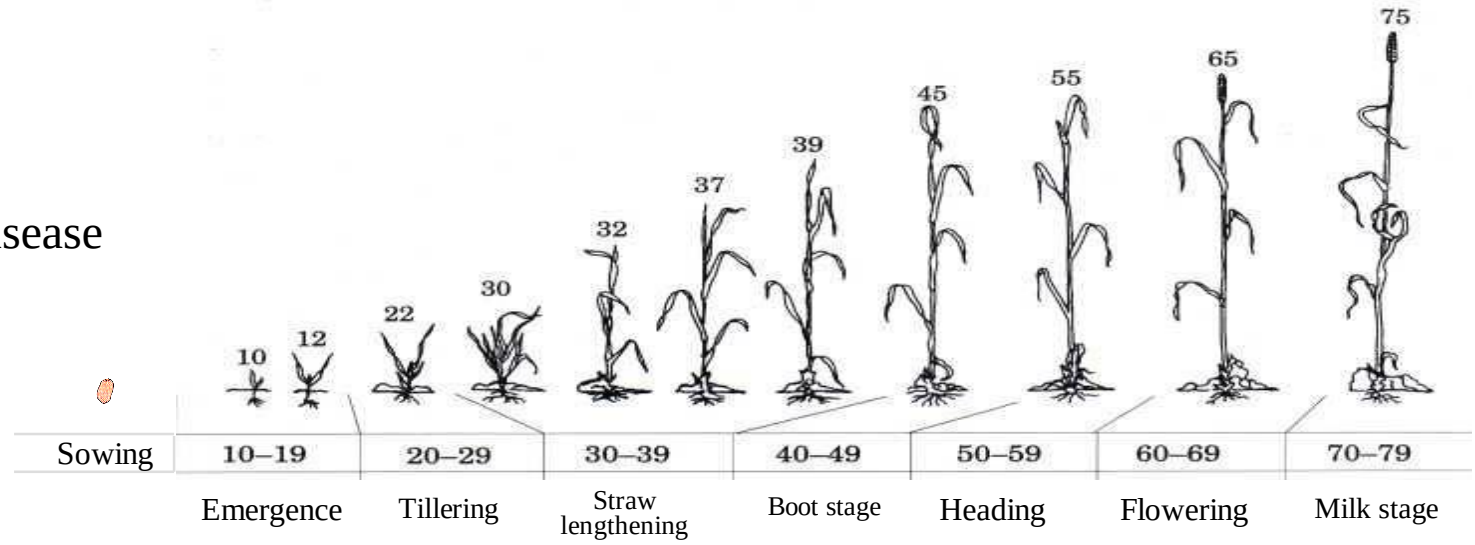
D. tritici-repentis

When the fungus (disease) can be observed in the field (wheat)

Fungus in the field

Control of the disease

Seed treatment



- Yellow rust
- Brown rust
- Mildew
- Common bunt
- Dwarf bunt
- *S. nodorum*
- *S. tritici*
- Tan spot
- Eyespot
- Take all
- Scab
- Snow mold

Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Breeding wheat for disease resistance: strategies

- Test method
  - Greenhouse and/or field
  - Plant test or progeny test
  - Test for large breeding material
- Crosses
  - Resistance sources X cultivated varieties
- Backcrosses
  - Straight backcross (same parent)
  - Balanced backcross (different selected parents)
  - Combination of resistance to different diseases
  - Combination of resistance, quality and yield
- Selection
  - Testing and selection of resistant progenies
- Field trials at several locations in Europe
  - Test for resistance, quality and yield
- Searching for new resistance sources
  - Genebanks
  - Within *Triticum* species and near related species

Resistance – what type of resistance?

Inheritance:
- single gene
- oligogenic
- polygenic

Expression:
- complete or partial resistance
- seedling resistance
  or
- adult plant resistance
- specific resistance (vertical)
  or
- unspecific resistance (horizontal)

Breeding strategy for mildew resistance

- Resistance testing in glasshouse using several races
  – Resistance confirmed in the field.

- Resistance based on several major genes with 2-3 genes (or more) in each variety.

- Extensive work with ”new” resistance genes from cultivated and wild progenitors.

Scoring wheat for mildew resistance

Material	Lines tested	P. mildew isolates			
		MDa	Egt1	46	Mhu
F2	650 x 3	X	X	X	---
F3	7 870 x 3	X	X	X	---
F4	7 133 x 3	X	X	X	---
F5	3 837 x 3	X	X	X	---
F6	460 x 3	X	X	X	---
F4 (bunt)	339 x 3	X	X	X	---
F5 (bunt)	837 x 3	X	X	X	---
PYT	1 500 x 4	X	X	X	X
AYT	300 x 4	X	X	X	X
Elite	200 x 4	X	X	X	X
DH	60 x 4	X	X	X	X
Polish mat.	20 x 4	X	X	X	X
TOTAL	71 698 winter wheat lines tested				

Infectando el material con Oidio (un hongo)

Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Scoring wheat for mildew resistance

Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Analizando el nivel de infección

Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Breeding strategy for leaf rust resistance

- Resistance based on a few major genes and quantitative resistance (probably polygenic).
- Field testing are more important than greenhouse tests.
- Extensive work with ”new” resistance genes from cultivated wheat and wild progenitors.

Seedling infection types – specific resistance

Specific resistance
Quality rating scale

0 Immune:
No signs of disease

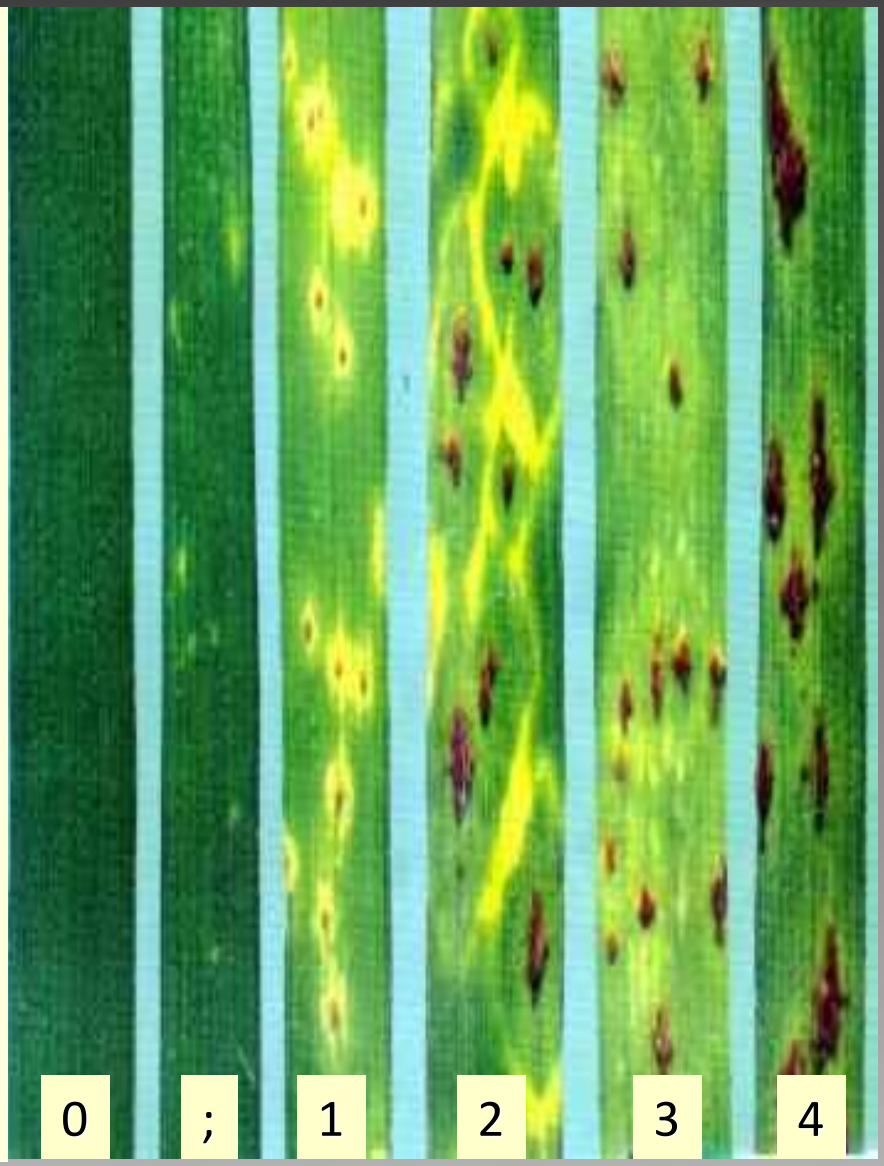
; Practically immune:
No pustules but hypersensitive flecking

1 Very resistant:
Pustules small and isolated.
Hypersensitive flecking.

2 Moderately resistant:
Pustules small to medium.
Surrounded by chlorotic or necrotic areas.

3 Moderately susceptible:
Pustules medium in size. No necrosis.

4 Susceptible:
Pustules large. Often united. No necros



Breeding for multiple disease resistances

F2 – selection in seedling tests

Breeding for resistance to mildew and leaf rust

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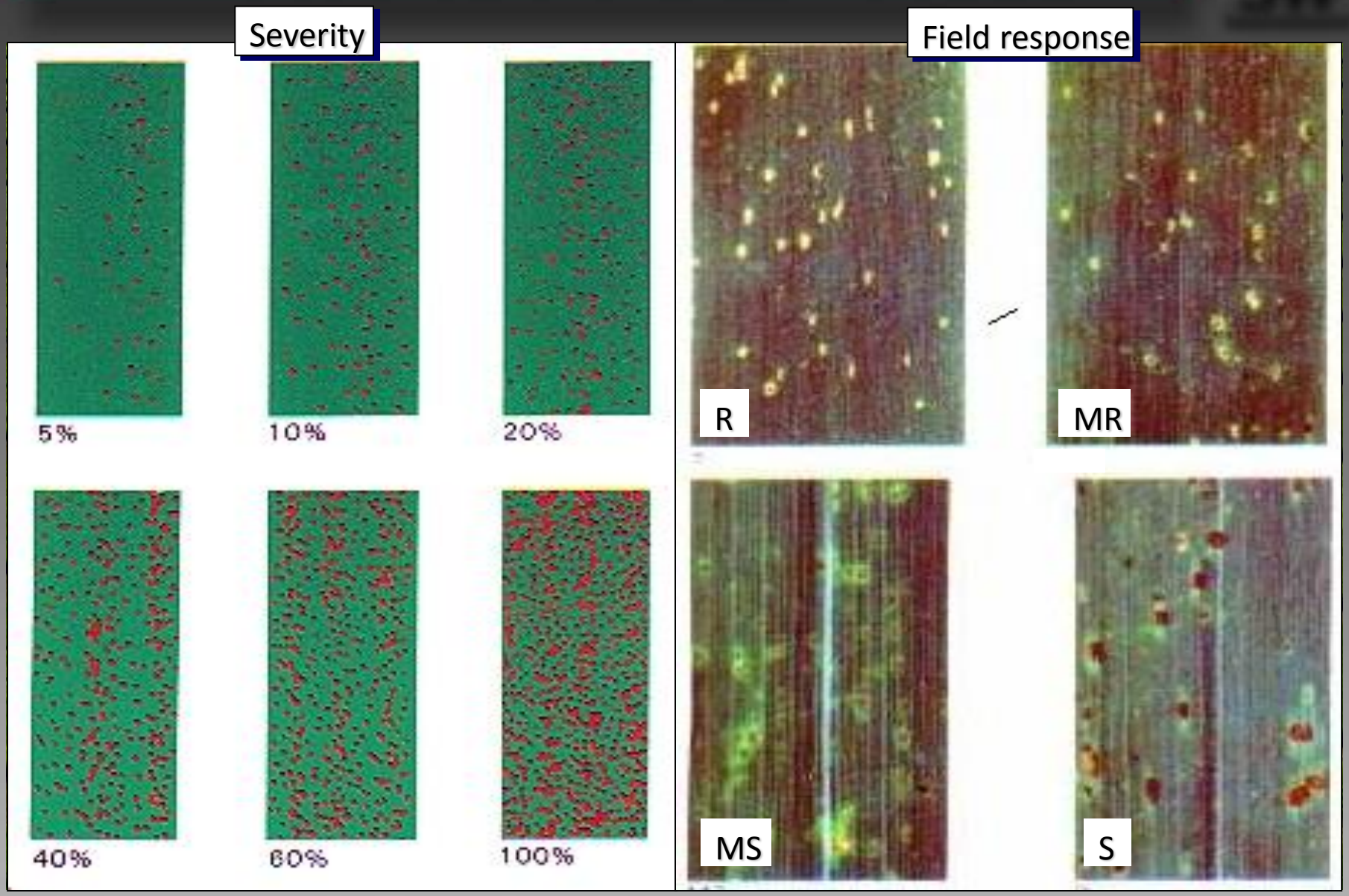
Resistance breeding – Tests in the field



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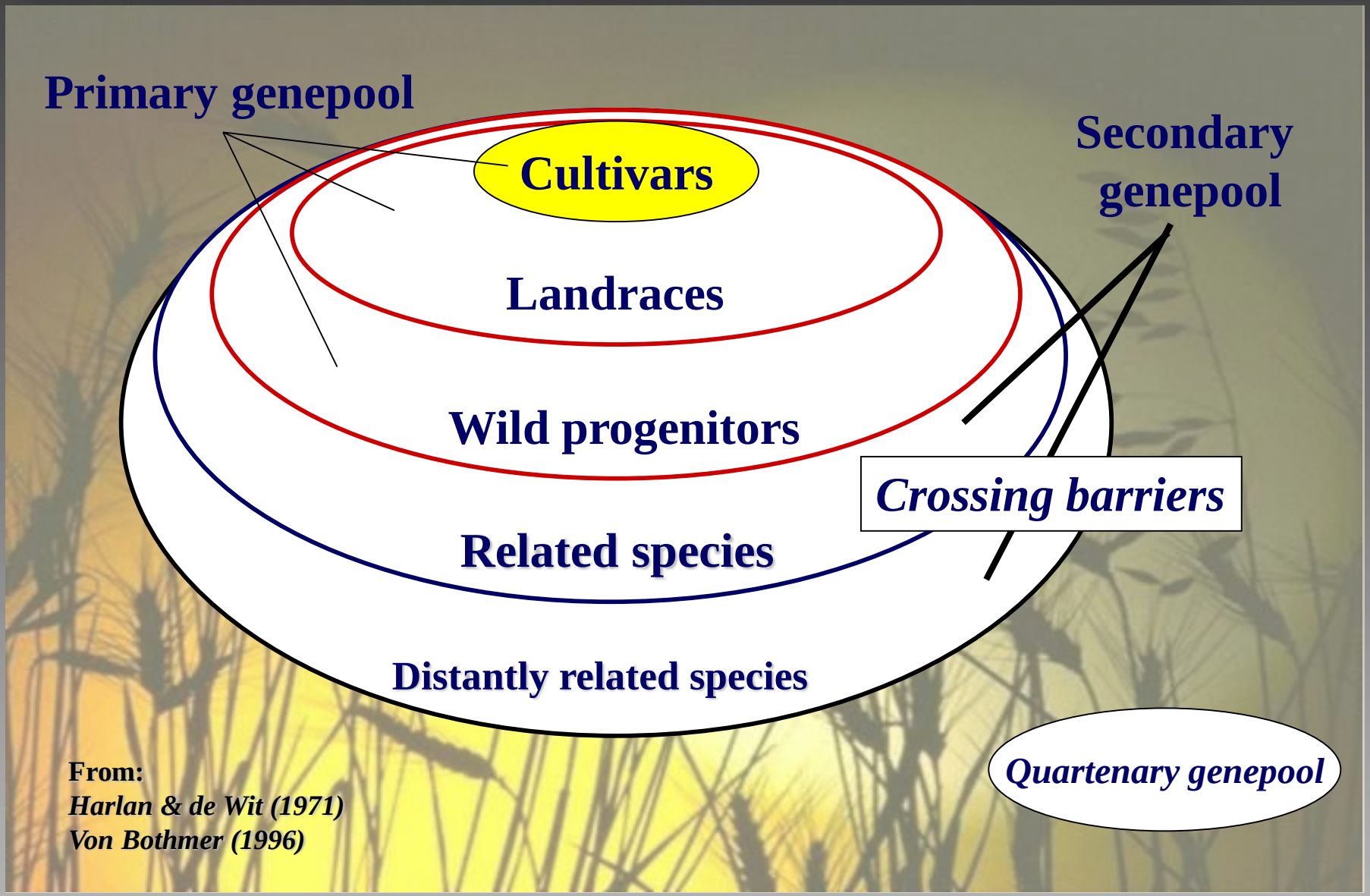
GENES DE RECURSOS FITOGENÉTICOS

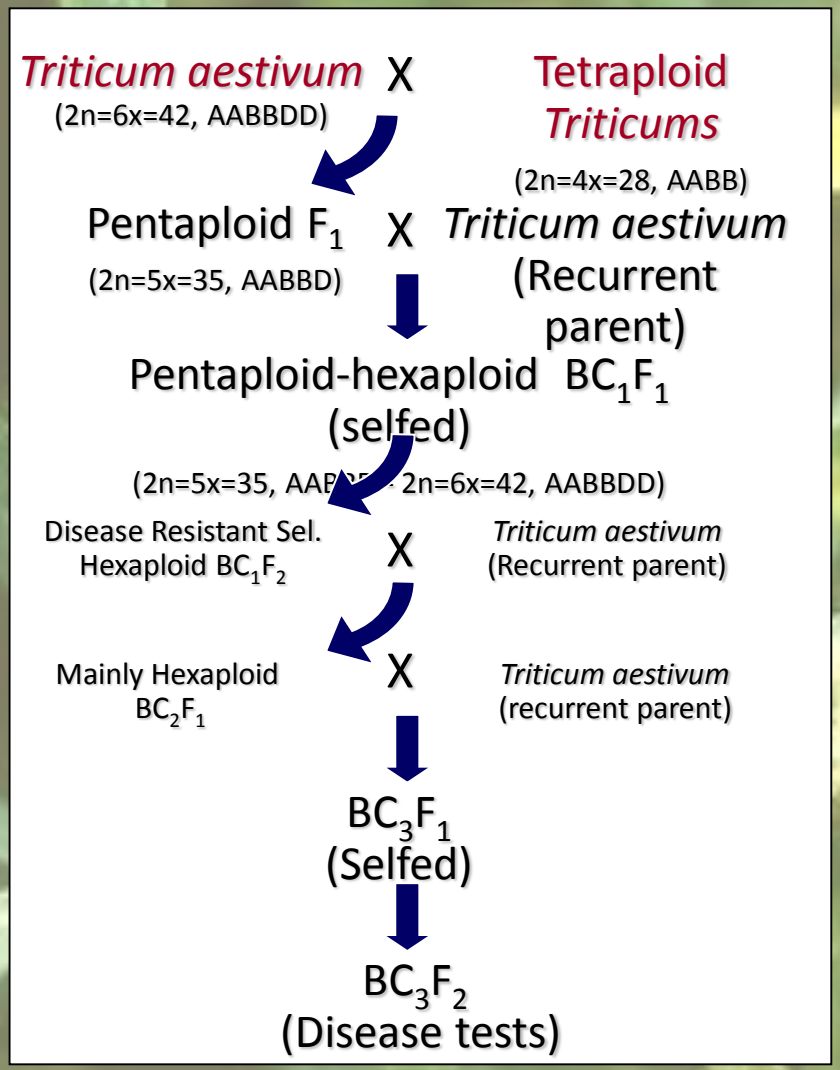


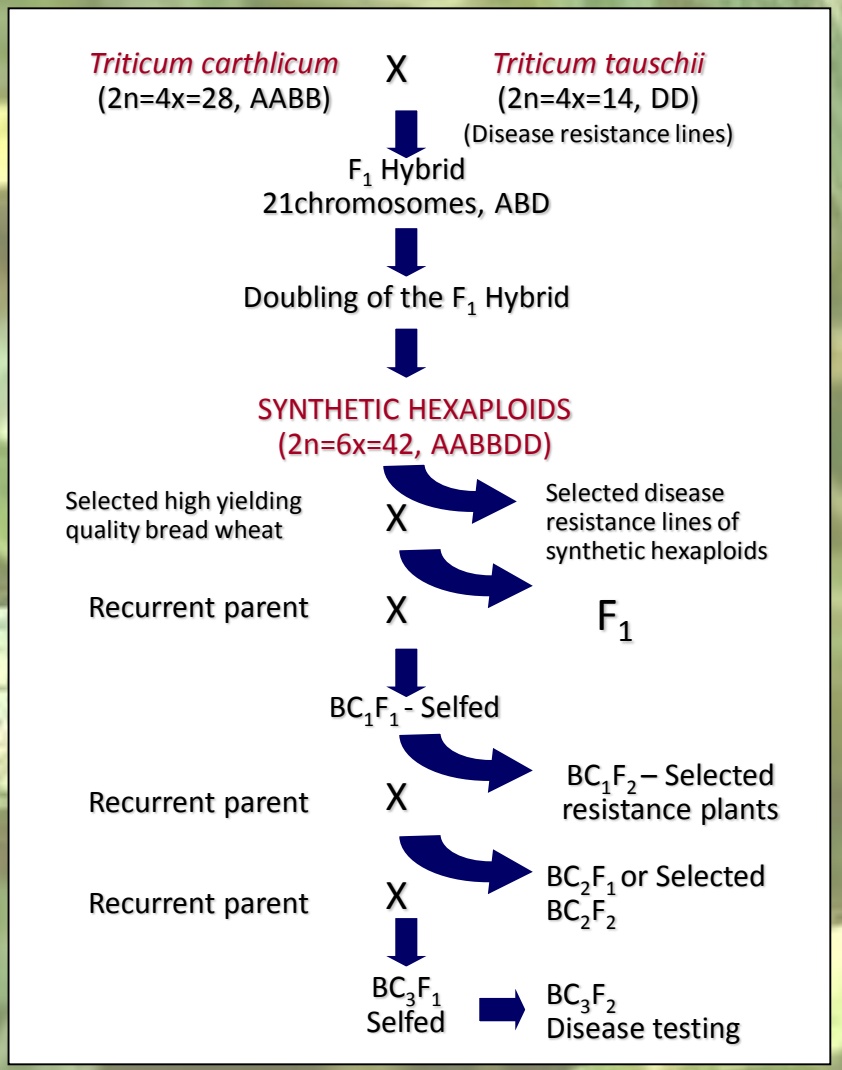
From:
Harlan & de Wit (1971)
Von Bothmer (1996)

Gene sources for resistance breeding in wheat

- *Tritordeum lines*
  (Durum wheat *x Hordeum chilensis*)
  → - Powdery mildew
  - Leaf and yellow rust
  - Septoria tritici
  - Common bunt
- *Triticoleymus lines*
  (*Triticum* ssp. x *Leymus* ssp.)
  → - Powdery mildew
  - Brown rust
- *Triticum* ssp.
  (*T. aestivum*, *T. carthlicum*, *T. durum*, *T. turgidum*, *T. dicoccom*, *T. dicoccoides*, *T. tauschii*, *T. monoccocum*)
  → - Powdery mildew
  - Brown rust
  - Yellow rust

Introduction of disease resistance genes from *Triticum* species





Shuttle breeding in Chile

Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Double haploid techniques at Svalöf Weibull

- Double haploid (DH) provide wheat breeders with pure lines in a single generation saving time in the breeding of new cultivars.

- DHs are also used for *genetic mapping* and for the *development of transformation systems* in wheat.

- In winter wheat, we usually produce about 4 000 lines of DH in order to speed up the breeding program.

Maize method (DH production)

Crosses of winter wheat X maize (sweet corn)

After 1-2 days, hormone treatment of spikes with 2,4-D

After 18-21 days, embryo rescue

Embryos are placed *in vitro* for growing for one month or more (green haploid plants, no albinos)

Transplanting of haploid plants to soil

Colchicine treatment

Vernalization

Marker Assisted Selection (MAS) in wheat

Microsatellites (SSR)

1. Evaluate the genetic variation and relationship of winter and spring wheat varieties using SSR markers:
   - Establish the genetic distance between varieties to make decisions on future crosses.
   - Provide a genetic fingerprint for each variety, which may be used for the implementation of plant breeders rights.

2. Identification of SSR markers linked to genes for resistance to several fungus diseases in wheat:
   - Monitor the transfer of disease resistance genes in breeding material
   - Pyramid resistance genes.
   - Combine resistance to different diseases.
   - Simplify resistance tests.

Hybrid wheat breeding in Sweden

- Wheat hybrids are direct progenies with improved characters compared to the average level of their parents (heterosis) or to the best parent (heterobeltiosis)

- In 1998, a chemical hybridization agent (gametocite) was made available to use in research on hybrid wheat. Crossing blocks have been sown since 1998 for the production of hybrid seed.

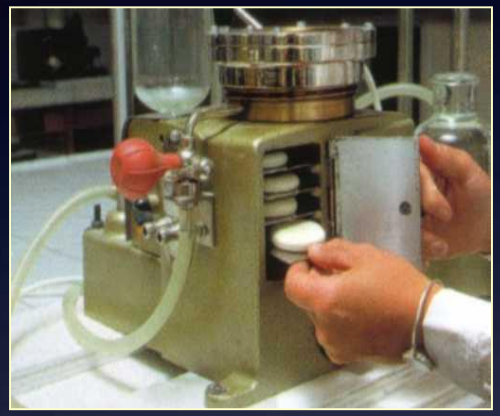
- To study heterosis, general and specific combining ability, yield trials have been carried out.

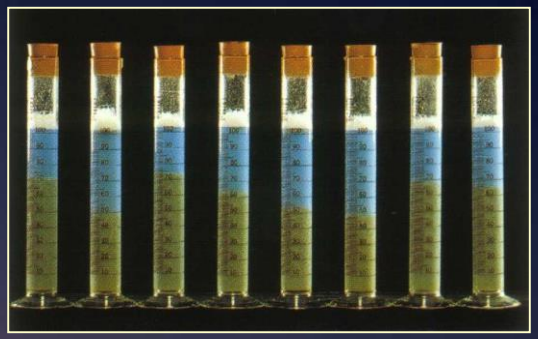
Hybrid wheat crossing blocks 2001

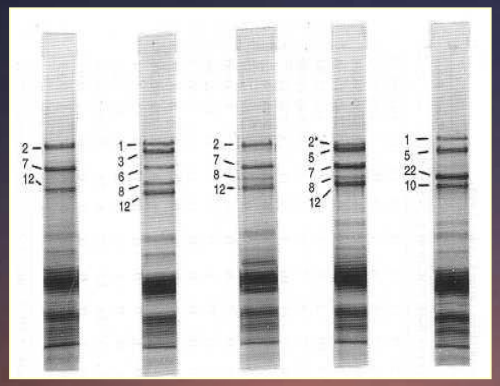
Triticale																			
SW 48246																			
SW 48246																			
101	102	103	104	105	106	107	108	109	110	111	112	113	114	115	116	117	118	119	120
SW 48246																			
121	122	123	124	125	126	127	128	129	130	131	132	133	134	135	136	137	138	139	140
SW 48246																			
SW 48246																			
Triticale																			
SW 48313																			
SW 48313																			
201	202	203	204	205	206	207	208	209	210	211	212	213	214	215	216	217	218	219	220
SW 48313																			
221	222	223	224	225	226	227	228	229	230	231	232	233	234	235	236	237	238	239	240
SW 48313																			
SW 48313																			
Triticale																			
Bill																			
Bill																			
301	302	303	304	305	306	307	308	309	310	311	312	313	314	315	316	317	318	319	320
Bill																			
321	322	323	324	325	326	327	328	329	330	331	332	333	334	335	336	337	338	339	340
Bill																			
Bill																			
Triticale																			
SW 48341																			
SW 48341																			
401	402	403	404	405	406	407	408	409	410	411	412	413	414	415	416	417	418	419	420
SW 48341																			
421	422	423	424	425	426	427	428	429	430	431	432	433	434	435	436	437	438	439	440
SW 48341																			
SW 48341																			
HK Pontus																			

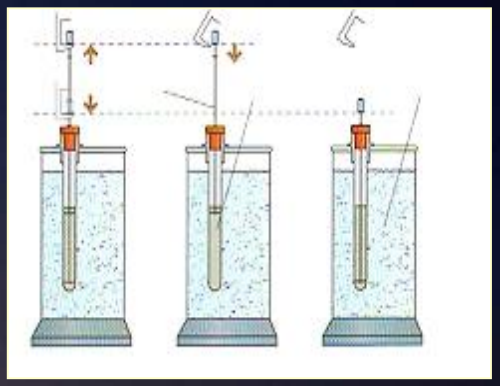
F1 hybrid wheat yield trials 2001 (2 repl)

QUALITY IN WHEAT









Resistance to sprouting in wheat

Dr. Oscar Díaz – Coordinador del Programa de Fitomejoramiento – Svalöf Weibull, Suecia, 1999-2004

Why do the wheat grains sprout?

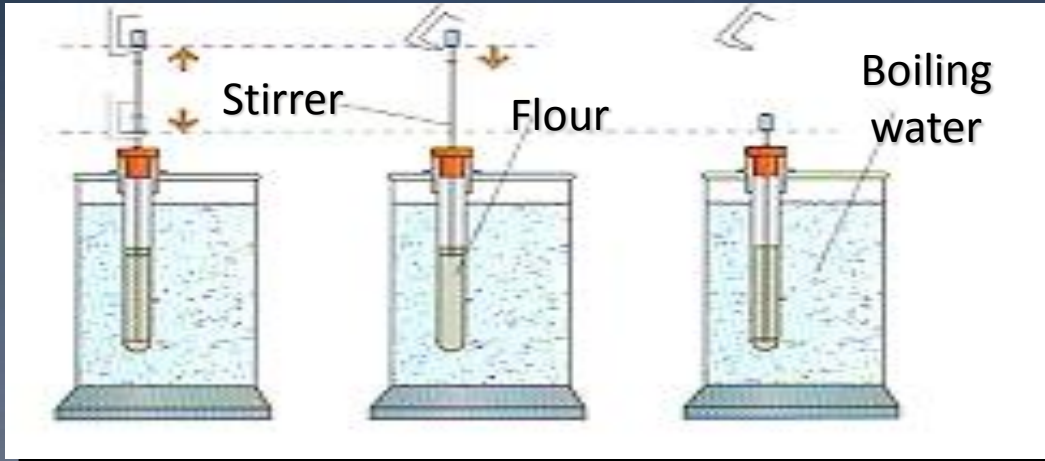
- Under rainy conditions, the grains of some wheat varieties germinate in the ear either before or at harvest-ripeness.
- Research has shown that just-matured wheat grains are usually not able to germinate (*sprout*). They first need a rest period (*dormancy*).
- Research has also shown that length of the dormant period depends on the weather before harvest: *high temperatures and low air humidities in the dough-ripe stage result in a shortering of dormancy and vice versa.*

Why do the wheat grains sprout?

- The sprouting process is initiated by an increase in *α-amylose activity in the grains*, although no external signs of germination are yet visible. Only 1-2 days after the start of the sprouting process, small rootlets appear at he base of the grains.

- The α-amylose activity in the grains and flour has proved to be a good indicator of the degree of sprouting.

Hagberg-Perten Falling Number Method



- A flour suspension is heated in a boiling-water bath so that the starch will be gelatinized.
- The starch is decomposed in sugars and dextrins by the amylases in the sample.
- The higher the α-amylase activity, the less viscous the suspension will be.
- 7 g of flour and 25 ml of water are introduced in the tube with a stirrer. The tube is placed in a boiling-water. The flour suspension is stirred for 60 seconds.

Falling Number and suitability for bread making

Falling number	Suitability class
<< 120	High sprouting level *(not suitable for bread or biscuit making)*
120 – 180	Sprouted wheat *(may be mixed with an unsprouted wheat lot)*
180 – 200	Low sprouted wheat
200 – 250	Unsprouted wheat
250 – 300	Unsprouted wheat *(should be mixed with malt flour or sprouted wheat)*
>> 300	Unsprouted wheat *(has to be mixed with malt flour or sprouted wheat)*

Sprouting as a bread making quality problem

- Bread made from sprouted wheat has an inferior quality because of the high α-amylase activity, which will break down too much starch into dextrins.

- The bread (crumb) becomes sticky and unpleasant to chew.
- The bread is also difficult to slice.
- The α-amylase activity should not be too low, since a certain amount of sugar is needed as substrate for the yeast.

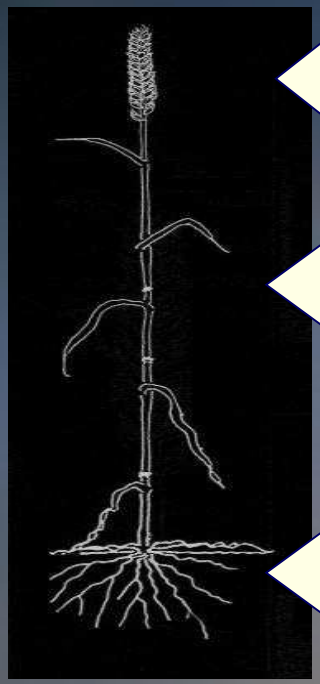
Strategies for sprouting resistance in Sweden

- A breeding program for sprouting resistance has been carried at Svalöf Weibull during many years. The key of this program is that one of the parent in the crosses must have a high falling number (> 350).
- All the wheat material from the advanced yield trial (AYT) is regularly tested by the Hagberg-Perten method. All the varieties with the falling number lower than 220 are discarded.
- A chamber have been specially designed to test the material in years where rainfall is not too much. A total of 25 spikes are selected from each variety and placed into the chamber during 4 days at 15-18 °C. Thereafter, the spikes are dry (25 °C) and the falling number is tested.

Cadmium in Wheat

- Cadmium (Cd) is a heavy metal with potential toxicity to the human.
- The problem was discovered in Japan (“Itai-Itai”). The water was contaminated by cadmium from the zinc-mines.
- Research has shown that high levels of Cd in wheat can cause unwell function of the kidneys, a decalcification of the skeleton, brittleness of the bones and neurological injury.
- The risk groups are young women with low amounts of iron in the blood, people with diabetes and smoker.

Factors affecting the upptake and transport of Cd to the kernel



60 µg/kg dm

200 µg/kg dm

400 µg/kg dm

- Flour
- Bran
- Germ

Cd in the kernel

Cd in the straw and leaves

Cd in the roots

Upptaking of Cd

Genetic Mecanisms?

Genetic Mekanisms?

Genetic mecanisms?

pH, Ca++, Z++, NO_3++

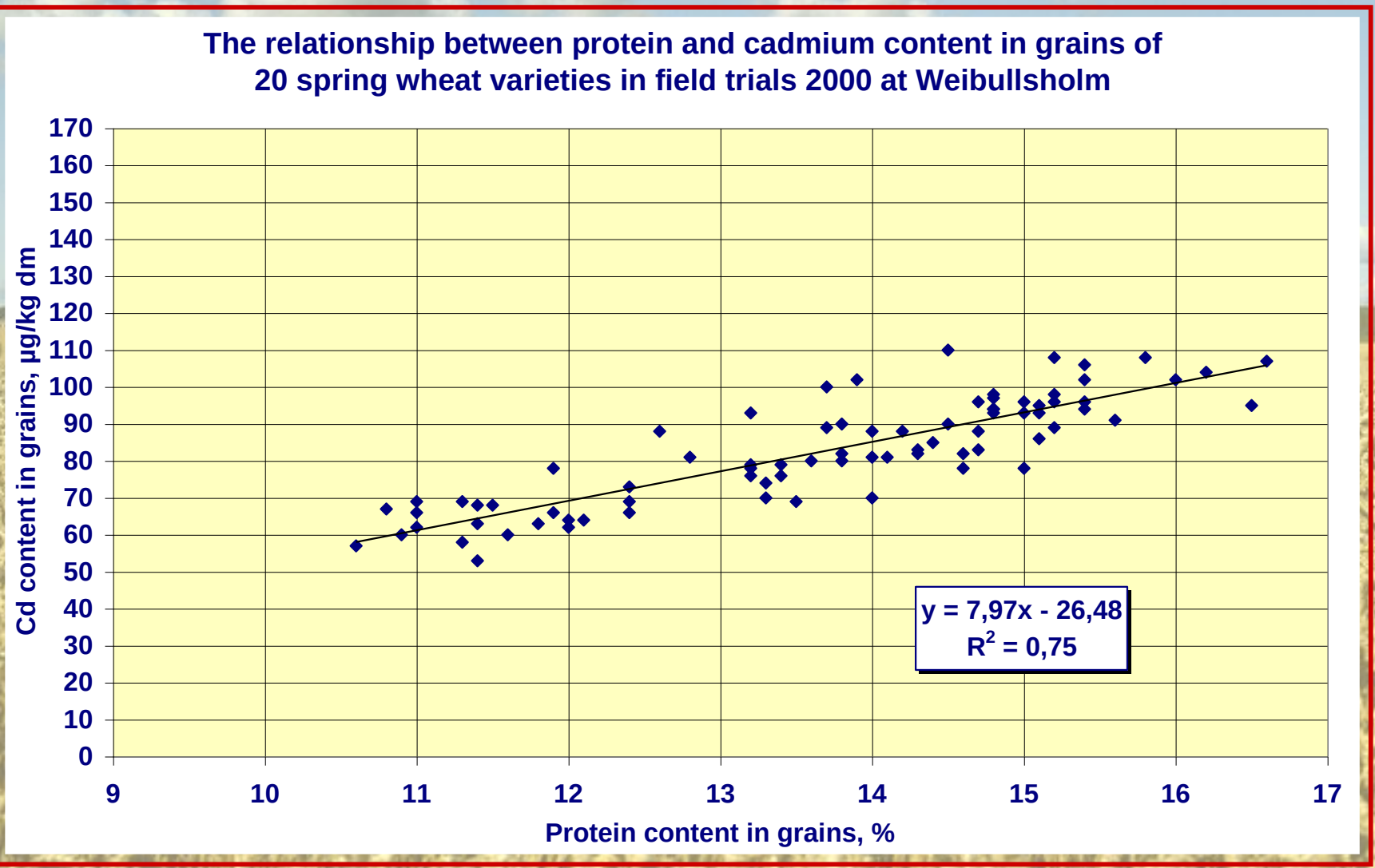
Weather conditions

Acid rain from the air

- Cd in the soil
- Mineralisation

- Fertilizers
- Phosphorus
- Sewage sludge
- Manure

Cd and protein content in spring wheat



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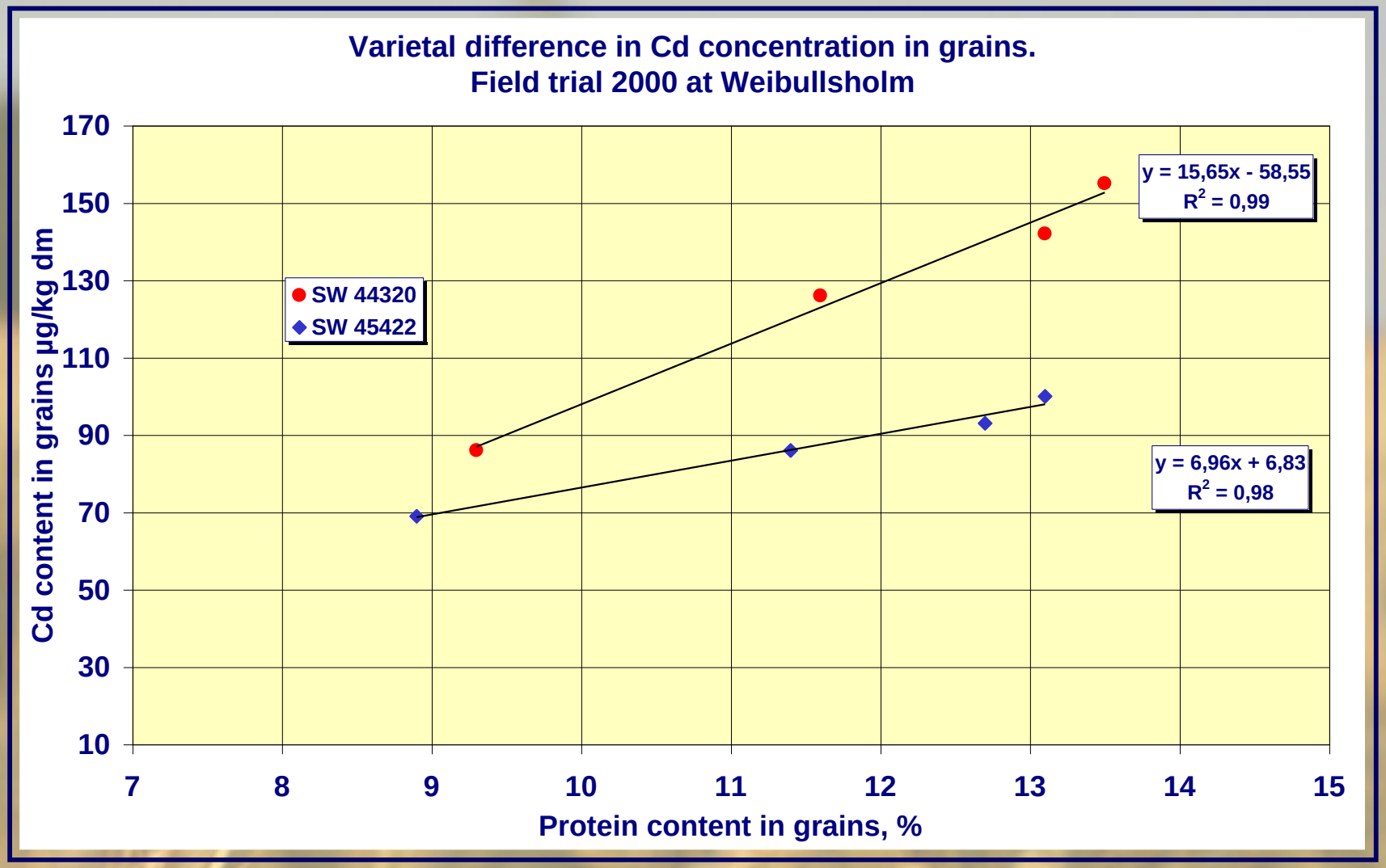
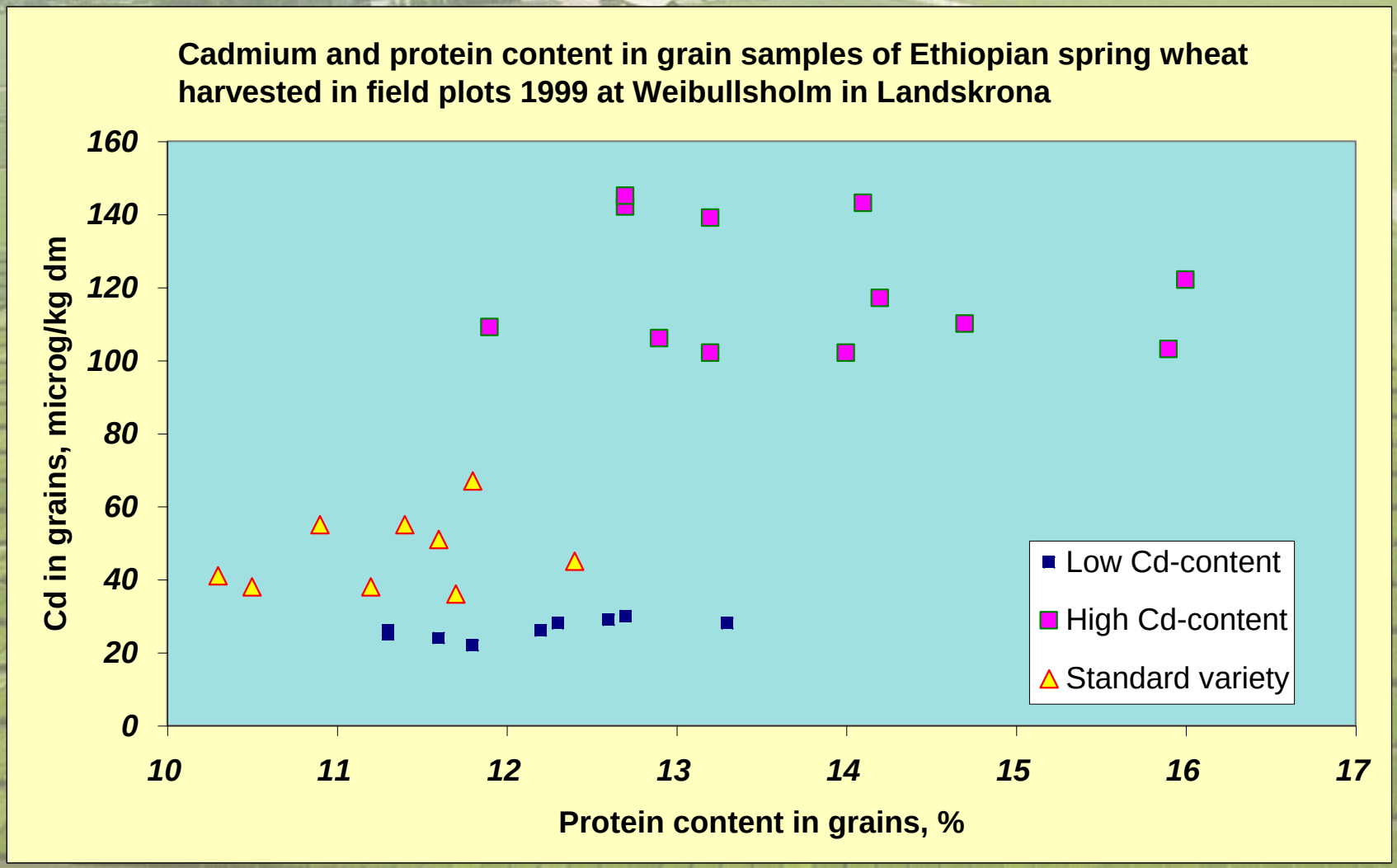
Varietal difference in Cd content – winter wheat



Ethiopian spring wheat



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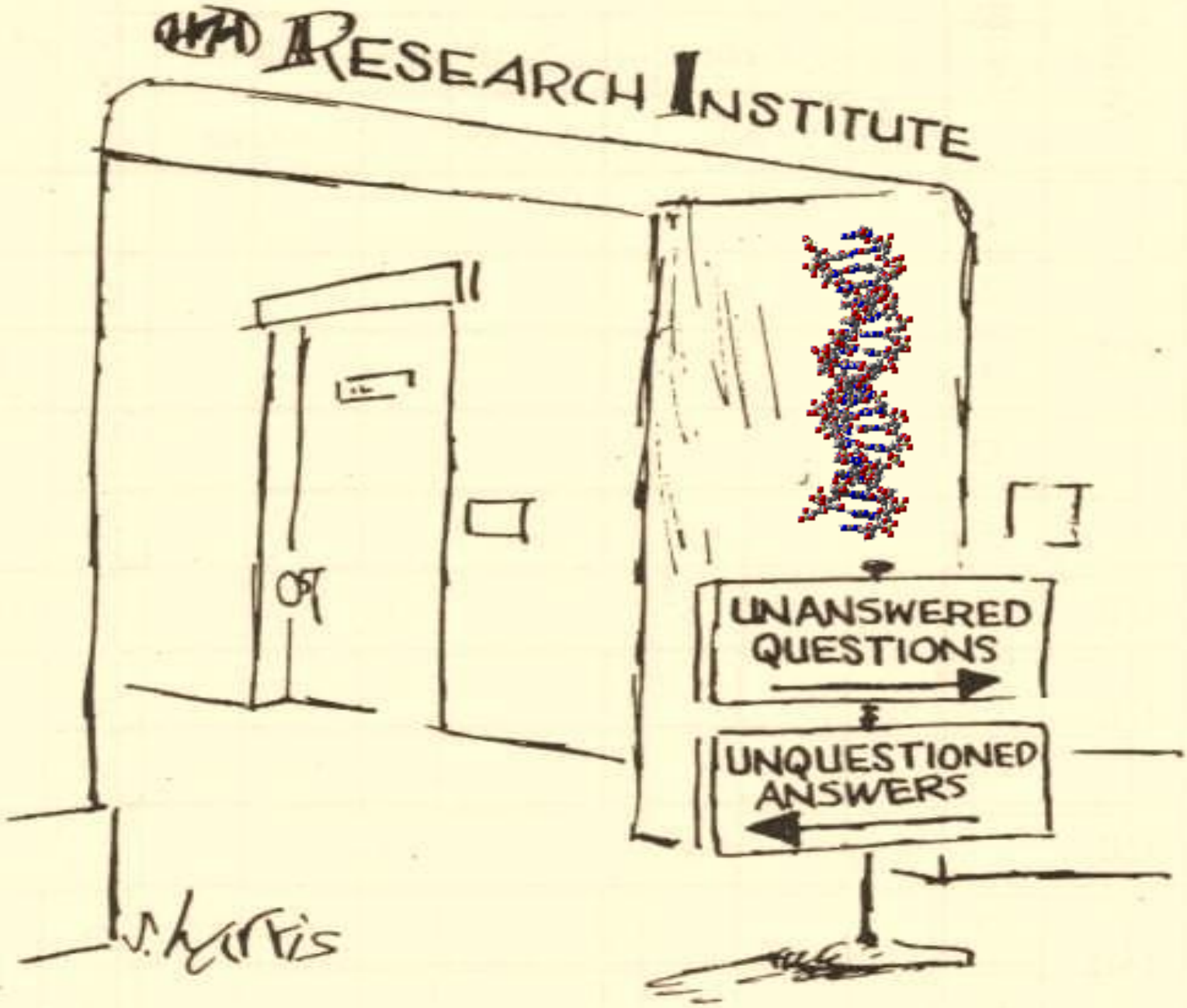
Soñando con el trigo del futuro...

Sir Rowland Biffin, one of the first English wheat breeders...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