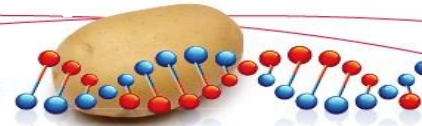




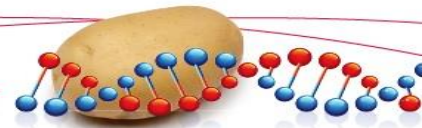
A plant breeding landscape

RESEARCH & DEVELOPMENT

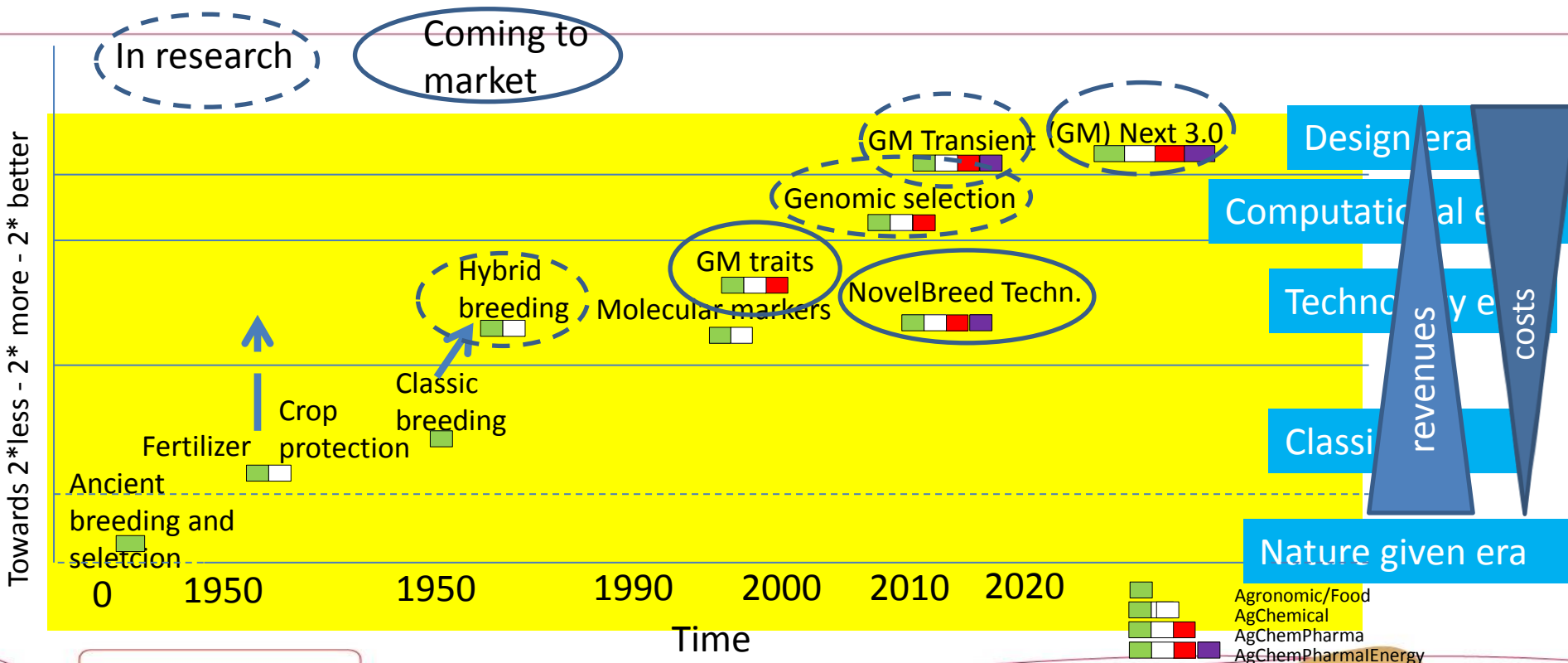


To come

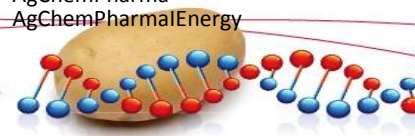
- Crop improvement in time
- Breeder's limits and needs
- Suggestions for science
- NBTs
- Durable resistance



Crop improvement in time



RESEARCH & DEVELOPMENT



Introduction HZPC Group

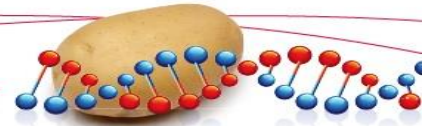


- Since 1898; origin Netherlands
- Number of employees: 262 (FTE)
- Sales of seed potatoes 652.000 ton
- Export/license ca. 75 varieties to 82 countries
- Sales of table potatoes 180.000 ton
- Turnover €275 mio; gm <15%; profit ca €5 mio.
- Shareholders: staff - growers - breeders

Sectors



RESEARCH & DEVELOPMENT



Market success is a 4-fold balance

AgroPhysio



Quality



Resistances

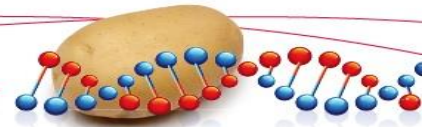


Value chain satisfaction

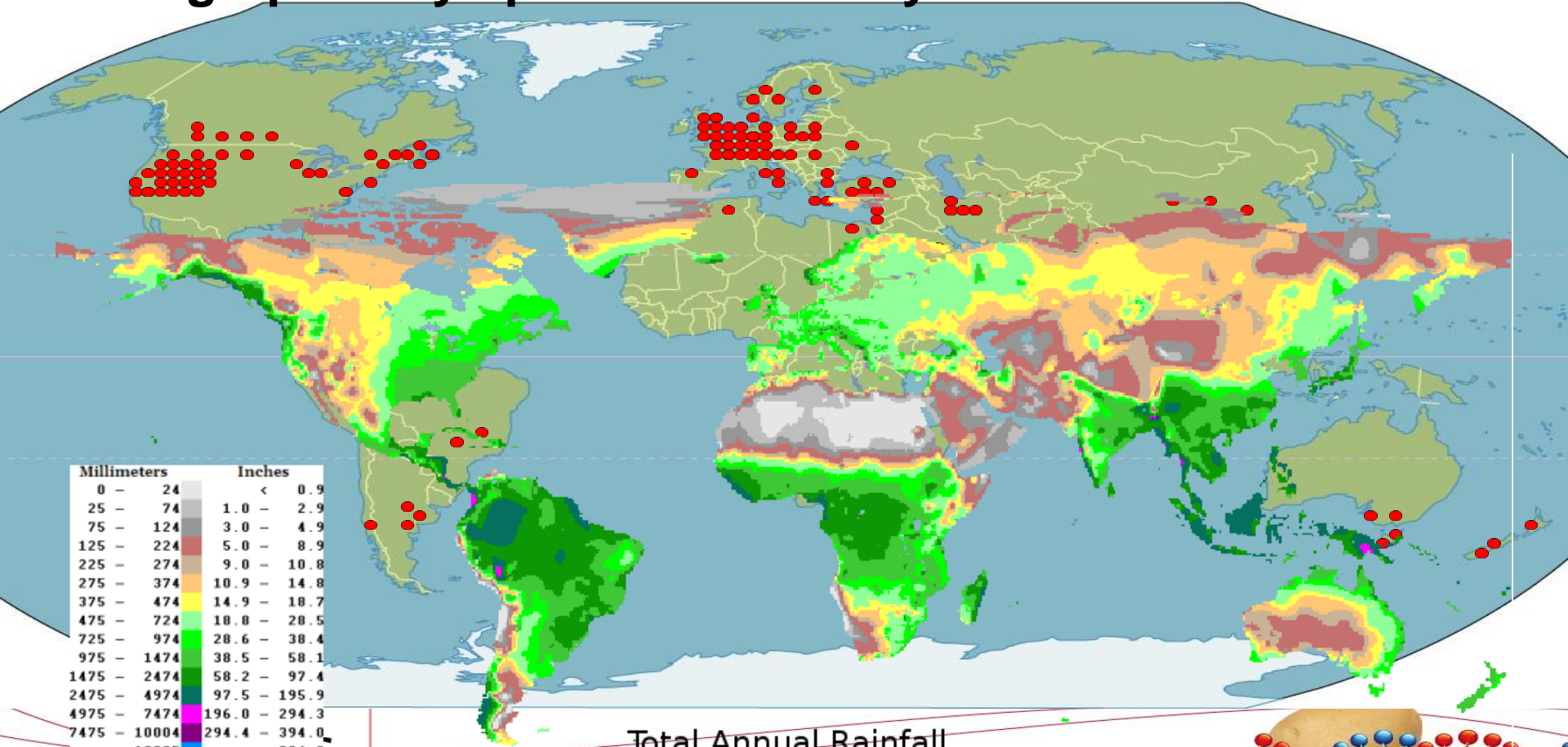


- Achieved by breeding and marketing (60/40; 40/60 ~ 50/50)
- Breeding: about control of heritable traits or performance and speed of recombination

RESEARCH & DEVELOPMENT



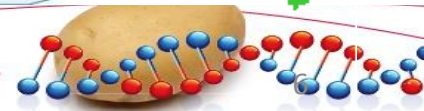
Geographically spread French Fry Factories



Millimeters	Inches
0 - 24	< 0.9
25 - 74	1.0 - 2.9
75 - 124	3.0 - 4.9
125 - 224	5.0 - 8.9
225 - 274	9.0 - 10.8
275 - 374	10.9 - 14.8
375 - 474	14.9 - 18.7
475 - 724	18.8 - 28.5
725 - 974	28.6 - 38.4
975 - 1474	38.5 - 58.1
1475 - 2474	58.2 - 97.4
2475 - 4974	97.5 - 195.9
4975 - 7474	196.0 - 294.3
7475 - 10004	294.4 - 394.0
> 10005	> 394.0

Total Annual Rainfall

RESEARCH & DEVELOPMENT

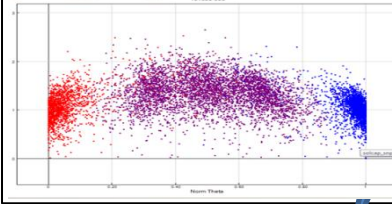


HZPC R&D



- R&D Centre in Netherlands
- 6 internal + 30 contract breeders
- annually 250.000 single hills
- 7 million mini tubers
- Selection in climates
- Towards Genomic breeding

MOLECULAR BIOLOGY & BIOINFORMATICS



PHYTOPATHOLOGY



TRIAL SYSTEM



QUALITY



PRE - BREEDING



Pre & Sector

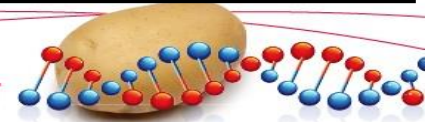
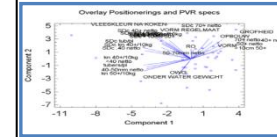


Breeding

QUALITY & FYSIOLOGY

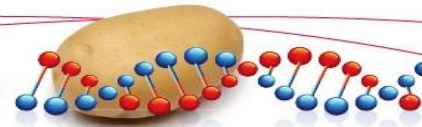


QUANTITATIVE GENETICS



Breeder's limitations

- 40.000 genes * 15 effective alleles is 600k of influences + epigenetical
- Challenge to make the best choice in 600k * 600k opportunities, fixed in series of quadruplex sets of performance.
- 100k potential crosses at present; <1000 are selected by phenotype, offspring, markers and statistical approaches.
- Potato breeding is still subjective at start; rely on yield measurement starts when <3% of initial genetic variation ('single hills') is left
- 8 yrs to identify the variety, 3 yrs to convince added value to customer and produce seed potato volume



Breeder's needs

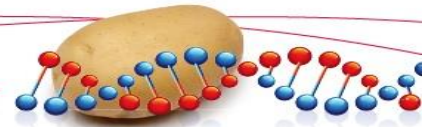
Become more objective with an effective start at crossing and single hill:

Baseline is available

- Phase 1: improved by MAB for resistance Pi R genes, (cyst) nematodes, virus and more; supported by validation protocols for diseases, objective phenotyping, additive genes in a good germplasm background

running and achievable, next,

- Phase 2: grow towards genomic breeding based on performance associated series of SNPs, control allelic contributions, supportive data systems.
- Phase 3: ...



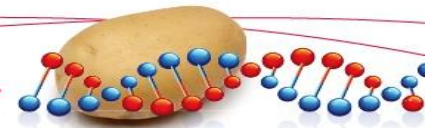
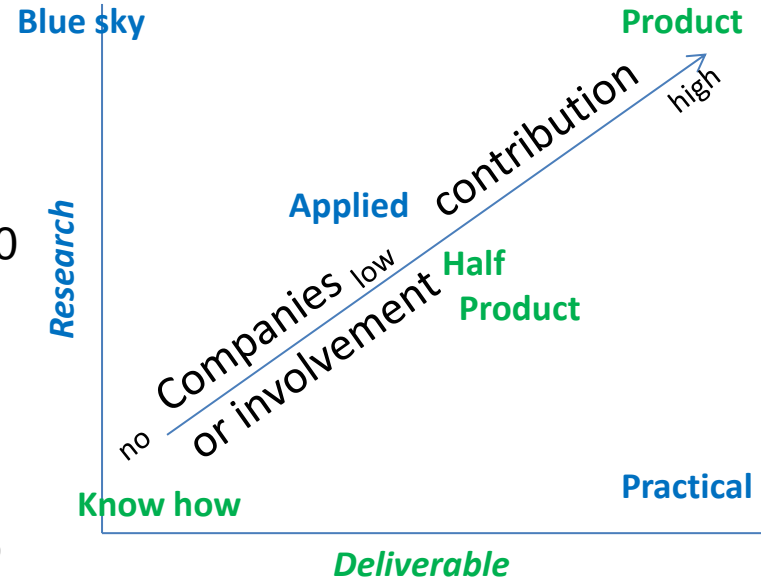
What's needed in/from collaboration with science:

Suggestions/challenges:

- Breeders that can handle the output
- Affordable FTO; more tools is less per unit
- Tuberosum reference genomeSSSSSS
- Broad Data handling systems
- Physiology know how (close the gap towards >100 structural tons/ha)
- Low-no mistake sequencers or an array for all important alleles, or both
- Non gm NBT's to solve/fixation simple traits
- Innovative approaches to unravel complex traits
- Potato companies understand your work better so
Take big steps, Think big, Act collaborative, Focus, Achieve goals in time, Communicate active, Catch

spinn-off

- more



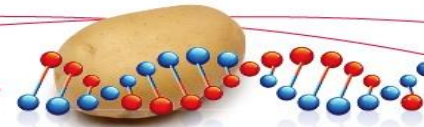
Complexity to come to a logic predictable performance

60-70%	15-25%	15-20%	100%
GENOTYPE	ENVIRONMENT	MANAGEMENT	PHENOTYPE
Controlled input	Variable component	modelling / DSS	Interactive result
DNA sequence, genotyping by sequencing, genomic data	Climatic data Consumer preferences	Physiology Control	Field performance
Epigenome	Detailed metadata	DSS	Plant architecture, growth rate, development, etc.
Methylation	...	QC's	Physiological status
RNA: arrays → RNAseq		Storage	Proteins
		..	Metabolites
			Other compounds
			...

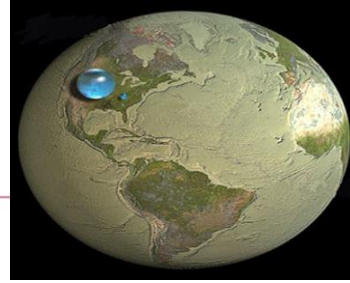
micro RNA's



RESEARCH & DEVELOPMENT



HZPC Challenges (at random)



- Feed the world, locally (food security, carbon print)
- Availability sweet water (salinity)
- High production on poor soils (-2 input, +2 output, better food)
- Raw product storage at low risk (resistances, physiology)
- Low-no losses in the value chain (now 40% food waste)
- Climate extremes within growing season (physiology)
- Secure an affordable legal ownership (build a world wide business)
- Fair profit for future developments (People Planet Profit, Corporate Governance)
- Consistent product on marketable yield & quality (measure, monitor)

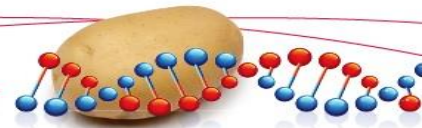


Potato is a great carrier to serve all items!

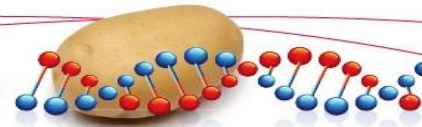
Staple crop nr 4 now,

Potential to move up to 3!

RESEARCH & DEVELOPMENT

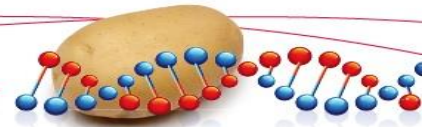


Novel Breeding Technologies



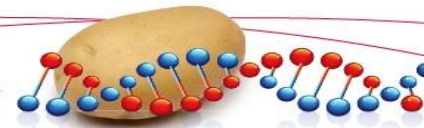
Novel breeding technologies

1. Mutation 'like': SDN1-3, ODM, TALEN, CRISPr, RTDS; techniques can knock out 1 bp up to introduction of genes, protein based or mimic natural repair mechanisms
2. Virus induced gene silencing: (transient) gene silencing mostly used for scientific complementation
3. Cisgenesis: A. tum introduced, only native crossable genes, <20 bp new
4. Grafting: non gm grafted on gm
5. Reverse genetics: redesign parental lines from a hybrid, contains a recombinant step to suppress the meiotic phase
6. RNA dependent DNA methylation; epigenetic change induced by dsRNA methylation
7. Agroinfiltration: local expression of foreign genes, parts of plant can be used for cuttings etc.
8. ..



EU

- Important to know that EU2001/18/EC is **process AND product** based!!! Also the Cartagena protocol is Product and Process based
- Article 2(2) Definition GMO:
 - Part 1: GMO means an organism, with the exception of human beings, in which the genetic material has been altered in a way that does not occur naturally by mating and/or ...= **PRODUCT** based
 - Part 2: genetic modification occurs at least through the use of techniques listed in Annex 1A part 1 and the techniques listed in Annex 1A part 2 are not considered to result in genetic modification= **PROCES** based
- Article 3 is about the application of a gm technique to an organism but will not lead to a GM, listed in Article 1B.
- Legal landscape: scientific base is is <40%, EU *lawyers must be comfortable when taken to court* is >60%

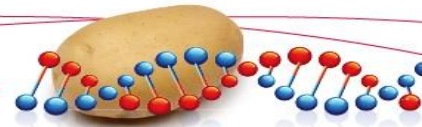


Durability of resistance

Definition: a resistance that holds over many years of agricultural use in various environments, challenged by pathogen recombination and selection in time.

CF9 (downey mildew tomato), Mlo (downey mildew wheat), H1 (Potato cyst nematode), Ve1 (Verticillium's tomato)

- Cf9: appears to be 2-3 genes, not one; **mimic by Cisgenesis**
- Mlo: Susceptibility genes knocked out, hard to replace; **reverse mimic by HIGS**
- H1: highly effective, still to be cloned and learn why durable
- Ve1: apparently gene for gene response with Ave1 Achilles heel

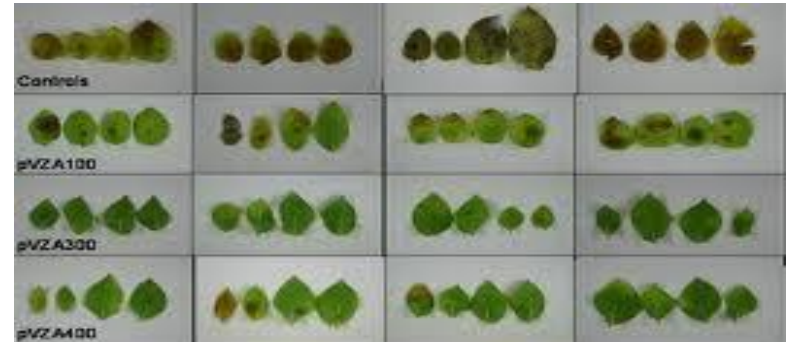


Potato varieties with Pi R-genes

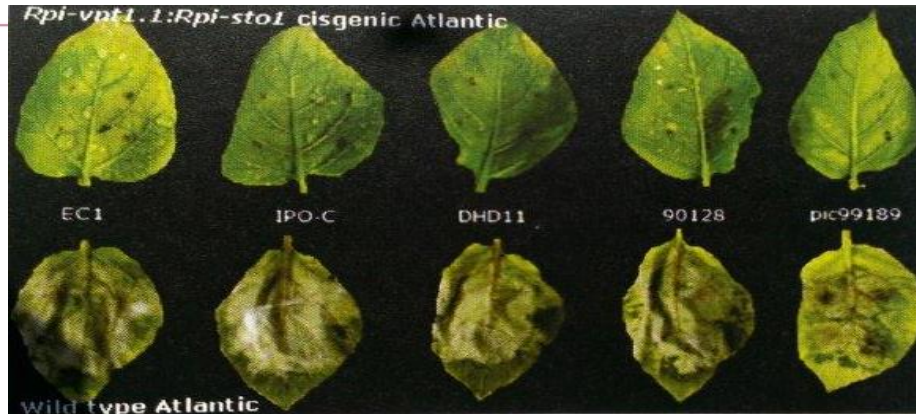
- Innovator, Santé, Escort, Toluca, Bionica, Carolus, Sarpo Mira, Biogold, Fresco, Connect, Raja, Athlete, Coquine, Voyager, Janine, etc ..
- About 10 different R-genes from ca. 5 species are or have been present in listed potato varieties

Aim: Durable Disease Resistance

“Resistance that remains effective over long periods of widespread agricultural use”



Cisgenic ~ mimic durable Cf9 / wild species approach



2013, Kwang Ryong Jo,
Unveiling and deploying
durability of late blight
resistance in potato, thesis.

Year	Genotype	R-gene content	Delay in days & relative						
2011	Bintje	-	2	a
	3151-3	9b	9	.	b	c	.	.	.
	MaR2	2	12	.	b	c	d	.	.
	3020-18	8	15	.	.	c	d	e	.
	edn7727-104	edn2	19	.	.	.	d	e	.
	edn7727-148	10, edn2	22	.	.	.	d	e	.
	3025-53	3a, 3b, 4, 8, 9a, 9b?***	19	.	.	.	d	e	.
	Toluca	blb2	18	.	.	.	d	e	.
	3025-1	abpt, 3a, 3b, 4, 9a, 9b?***	23	.	.	.	d	e	.
	3025-43	3a, 3b, 8	19	.	.	.	d	e	.
	3025-48	3a, 3b, 4, 8, 9b?***	36	e	f
	Sarpo Mira	3a, 3b, 4, smira1, 8	44	e
	MaR8	3a, 3b, 4, 8	43	e
	edn7727-102	edn1, 4, edn2	40	e
	edn7727-158	edn1, 10, edn2	40	e
	MaR9	1, abpt, 3a, 3b, 4, 8, 9a, 9b	114	f
	edn150-4	edn1, 4, edn2	No lesions**	f
				g

Durable resistance:

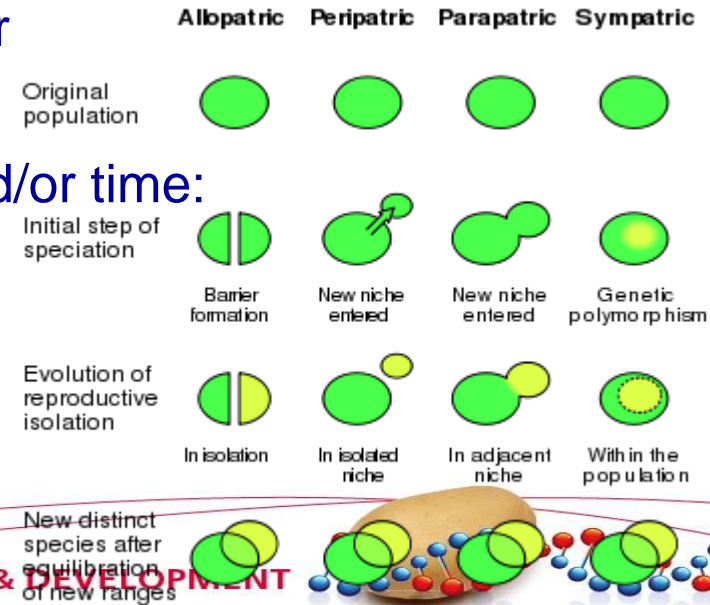
Nature will win, Varieties should hold long ..

Strategies to enhance durability of resistance

- Pyramid/stack multiple genes – increase evolutionary hurdle
- Utilize ‘durable’ or ‘non-host’ resistance
- Match life expectancy of resistance and cultivar

Diversify selection pressure on pathogen

- heterogeneity of resistance genes in space and/or time:
- diversify resistance sources, host & non-hosts
- pipeline with different resistance genes,
- shaping syn- and allopatric evolution,
- multilines and cultivar mixtures.

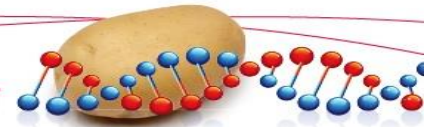


THE 'BOOM-AND-BUST' CYCLE OF DISEASE CONTROL

The Agricultural Consequences of Pathogen Evolution



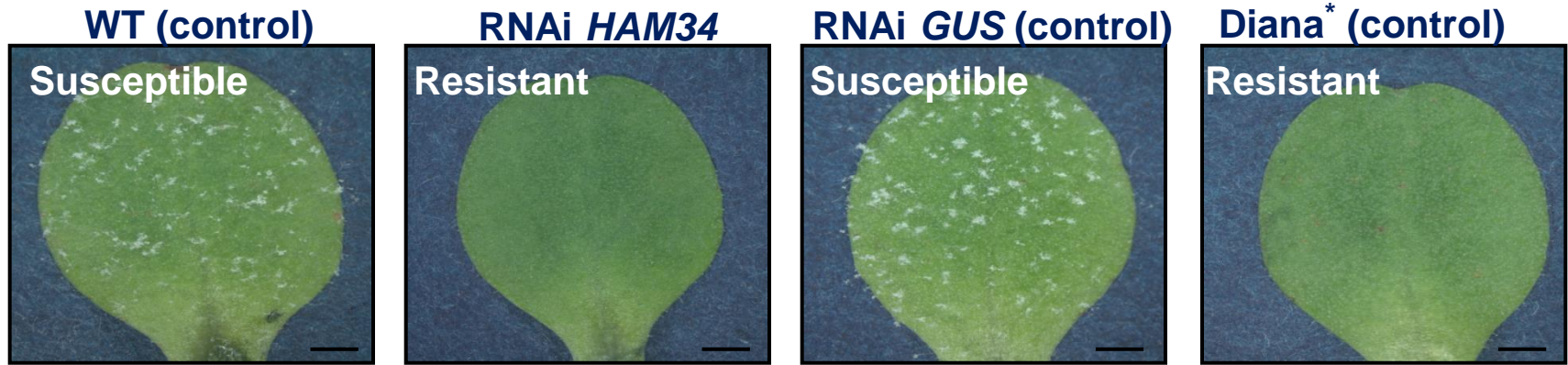
(Suneson, 1960)



Host Induced Gene Silencing

Of six genes tested, two (*HAM34* & *Cellulose Synthase*) resulted in inhibition of *B. lactucae*:

Transgenic T₃ lettuce seedlings expressing RNAi *HAM34* are resistant to *B. lactucae* (8 dpi)



Kindly provided by: Richard Michelmore - Manjula Govindarajulu
<http://michelmorelab.ucdavis.edu/>

RESEARCH & DEVELOPMENT

