Exploiting the wealth of genebank collections in breeding

Kees Reinink, Rijk Zwaan
Why genebanking?

• cultural historic reasons (museum function)
• to use old varieties in demonstrations, research and teaching
• to compare modern day varieties with varieties that were important in ancient days
• **to preserve genetic variation for use in breeding**

cv. “Wonder van Voorburg”
Genetic variation as basic substrate of plant breeding

- Breeders continuously work on genetic variation to select better varieties.
- The type of genetic variation that breeders are searching for varies depending on breeding goals and crop.
Genetic variation as basic substrate of plant breeding

- In staple food crops generally yield and yield related traits such as resistance to biotic and abiotic stress dominate the breeding goals.
- In vegetables and ornamentals many other traits (size, taste, vitamin content, colour, shelf life etc.) can be important in breeding.
Breeders are interested in specific traits or genes in genebank accessions

- Genebank accessions as such only seldom are of interest to a breeder.
- Breeders search for traits that add value to elite material.
- Phenotypic descriptions are mostly of little value. A minimum descriptors list is sufficient.
- Important is a correct botanical classification of the species.
Introgressing a trait from a genebank accession

- The breeder must try not to lose genes involved in the expression of the trait.
- Remove all linkage drag
- When introgressing traits from a very distant plant type or from wild material it is often difficult to recognize linkage drag, as most of the material displays an unwanted phenotype.
- Only when the material has advanced to the level of elite material, linkage drag can clearly be recognized.
Interspecific cross between *Lactuca serriola* and *L. sativa*
Introgressing a trait from a gene bank accession

- Introgression is most easy when it concerns monogenic traits with a clear expression.
- Many resistance genes fall into this category.
- Genebank material has been widely used as source of new resistance genes for breeding programs.
Example 1: Virus resistances claimed in *Capsicum annuum* (pepper)

- Chilli veinal mosaic virus  
- Cucumber mosaic virus  
- Paprika mild mottle virus  
- Pepper mild mottle virus  
- Pepper mottle virus  
- Pepper yellow mosaic virus  
- Potato Y virus  
- Tobacco etch virus  
- Tobacco mild green mosaic virus  
- Tobacco mosaic virus  
- Tomato mosaic virus  
- Tomato spotted wilt virus

(from ISF website http://www.worldseed.org)
Example 2: Fungus and insect resistances claimed in *Lactuca sativa* (lettuce)

**Fungi:**
- *Bremia lactucae* Downy mildew
- *Fusarium oxysporum f. sp. lactucae* Fusarium wilt
- *Microdochium panattonianum* Anthracnose

**Insects:**
- *Macrosiphum euphorbiae* Potato aphid
- *Myzus persicae* Green peach aphid
- *Nasonovia ribisnigrri* Lettuce leaf aphid
- *Pemphigus bursarius* Lettuce root aphid

(from ISF website http://www.worldseed.org)
Introgression of more complex traits

Traits with a more complex inheritance and a more quantitative expression are much more difficult to introgress from gene bank accessions into elite material.

1. extreme variation after wide or interspecific crosses obscure the effect of the genes of interest
2. difficult to keep several genes together
3. problem with linkage drag increases with the number of genes to be introgressed
Use of modern genomic tools

- Genomic tools revolutionize plant breeding.
- Also impact the use of gene bank material in breeding programs.
- In most crops a large number of molecular markers is now available.
- For many crops the genome sequence is available or under construction.
Use of modern genomic tools

- Use of markers to follow the introgression of genes and reduce risk of losing genes.
- Minimize the size of introgression fragments.
- Creation of Backcross Introgression Library (BIL) lines → localisation of candidate genes.
- Candidate gene and (eco)tilling approaches.
- Mutation breeding as alternative source of genetic variation.
BIL population of *Lactuca saligna* in lettuce background

L. *sativa* cv. ‘Olof’

L. *sativa* cv. ‘Olof’ + saligna 8.2

(M. Jeuken, Wageningen University)
BIL population of *Solanum pennellii* in tomato line (Zamir)

- *S. pennellii* LA716
- *S. lycopersicum* M82

→ Lines with yellow or orange fruits
BIL population of cucumber – segregation for colour

White
White green
Light green
Green
Dark green

P1
P2
Candidate gene approaches

- Applying genetic knowledge from one species to another
- Genetic knowledge developed in model species or other crop species.
- Huge capacity to screen populations by sequencing or tilling for certain allelic variants.
- In the near future large parts of (core) genebank collections will be resequenced.
- Core-collections will be redefined based on sequence information to have maximal allelic content.
Example: *eIF4E* gene

Organization of *Cm-eIF4E* gene

(Nieto et al., BMC Plant Biol. 2007; 7: 34)

- eIF4E is part of a protein complex with an essential role in mRNA translation.
- eIF4E is a susceptibility factor required for plant virus multiplication.
Example: *eIF4E* gene

- Mutants of eIF4E are associated with resistance of several crops to several viruses:
  - Melon Necrotic Spot Virus
  - Lettuce Mosaic Virus
  - Pea seed-borne mosaic virus
  - Tobacco etch virus (TEV) and Pepper veinal mottle virus infection in pepper
  - PVY virus in tomato
  - Barley yellow mosaic virus
  and many many others….
Example: Mlo gene

- Mlo’s belong to the family of seven-transmembrane (7TM) domain proteins.
- Some Mlo proteins act as “susceptibility” factor.
- Mutations in these Mlo genes result in broad spectrum resistance to powdery mildew in:
  - barley
  - Arabidopsis
  - tomato
  - grapevine
  - ……. 
Revival of mutation breeding

• Screening of large collections for a specific mutation is easy.
• Both genebank and mutant population can be source of new alleles. (ecotilling $\leftrightarrow$ tilling).
• Next step will be targeted mutation breeding (allele surgery, genome editing).
Mutants for shelf life in cucumber

184 Ethylene insensitive mutant

186 control

185 Ethylene insensitive mutant
Mutants for Russet spotting in lettuce

control

ethylene insensitive mutant
Conclusions 1

• New tools greatly increase the capacity of breeders to evaluate and absorb new genetic variation from genebank collections.

• Breeders would like to have access to collections that are as large as possible to search for alleles of interest.

• Genebanks can serve breeders by open access to a well maintained collection
  • preferably sequenced
  • information and accessions accessible through the Internet
Conclusions 2

- Collections can be further enlarged by collection trips organized in cooperation with breeding companies.
- Genebanks can play a broker function when access to germplasm is restricted for whatever reason.
Thank you for your attention