

Conserving and utilising plant germplasm and genetic resources

Biodiversity and genetic diversity

Biodiversity, which is variation among species, can only occur by means of variation within species. Genetic variation is a measure of the variation that exists in the genetic make-up of individuals within populations. It is the product of evolution. The theory of evolution is at times crudely known as the concept of the 'survival of the fittest'. Variation allows populations to adapt to a changed or changing environment. What happens if a plant is required to adapt with ecological changes? Variation is essential not only for the survival and reproduction of plant populations, but also for the very existence and the preservation of species diversity. Information about the genetic diversity of agricultural plants helps scientists and farmers to form strategies to preserve and protect the diversity of plants on and off-farm.

Why do we have gene banks?

In 1970, a disease epidemic struck the maize crop in the United States because of genetic uniformity, which resulted from the widespread reliance on cytoplasmic male sterility (CMS-T) for hybrid seed production. Later identified as 'race T' of the fungus *Helminthosporium maydis*, it shortly became known as the Southern corn leaf blight. In the early 2000s, an aggressive fungus spread from Africa and headed for the global centres of wheat-growing areas. This crisis is well documented and was known as the 2000s wheat rust (Ug99) crisis. It was brought partially under control only when a resistant accession (collected from North Africa) in Germany's IPK seed bank was found, hence the significance of the preservation of genetic diversity for generations to come. Potato cyst nematode (PCN) is the latest threat in 2016 that destroys more than 80% of the crop and remains active for a long time once established.

On a frozen arctic island, more than 850,000 samples of seeds are being stored at the Global Seed Vault to protect the world's food supply from epidemics, disasters or wars that might wipe out irreplaceable seed varieties. It is an international back-up centre for national and international plant gene banks around the globe. These gene banks are crucial resources for sustainable agriculture. Currently there are just about 2,000 gene banks worldwide holding 7.5 million accessions or individually collected packets of seeds. These genetic resources are strategic global assets. A sensible investor would be passionate about generating satisfactory yields with the lowest cost, lowest risk practices. Genetic resources held at the gene banks are a safety net against the loss of valuable germplasm. Guaranteed support for national gene banks is essential for the preservation of genetic diversity of food and feed crops and the protection of food resources. These assets conserve rich gene pools and help feed the host country as well as the world.

The relatives of today's crop and forage species, the ancestors and families of today's crop and forage plants, promise farmers an effective weapon to adapt to, rather than fight, climate variation. Priceless genes can be lost if these genetic resources are not collected, characterised, regenerated and maintained. Gene banks have the responsibility of routine processes and jobs, many interlinked, needing strong management.





Seeds of some cultivars developed from germplasm at AgResearch's Margot Forde Germplasm Centre

The seed collection at gene banks includes important traditional cultivars, their non-domesticated relatives, advanced as well as obsolete commercial varieties and breeding material. However to make this collection useable by breeders, scientists and the end users (farmers), a limited number of the accessions in a collection of each species (with a minimum amount of repetitiveness) must be provided by gene banks. This subset must represent the genetic spectrum of the whole collection. Any data obtained from the collection, including passport (collection site) and characterisation (molecular or non-molecular) data, can be used to select this 'core collection', which will then have as much genetic diversity as possible depending on its size. Genetic variability within the core collection of a species increases the chance of at least a few individuals being resistant to diseases or tolerant to abiotic stress such as drought, ensuring the survival of rare genes in that species.

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Climate change – threat or opportunity?

Climate is rapidly changing, putting our agricultural systems under more strain than ever before. Agriculture needs to acclimatise to tolerate more unusual weather patterns and varying conditions such as increased occurrence of low or high rainfall, heat waves and salinity.

The prospect of climate change in New Zealand based on patterns and forecasts by NIWA will inevitably force agricultural practice to change, adopt and adapt. The pace of this change must be planned in two tiers:

- *Incremental* – by doing better what we are doing now for the next 10 years
- *Transformational* – meaning something has to give/change and there are consequences. This needs to begin today and yield new products in the next 10 to 15 years.

Incremental change

Continuous testing and characterisation is recommended for the incremental change approach to develop new cultivars of current forage species used in New Zealand. The end products must match the system, environment, soils and management. Red clover is an example of a current species without exploitation of its potential genepool for this country. There are pros and cons in using red clover as a forage in New Zealand. The positive attributes are quality and intake, wide adaptation, seasonality of yield, ability to grow in mixed swards and, indeed, free nitrogen. The negative sides include low seed yield, poor persistence and under-grazing by animals. A focus for future research on red clover germplasm might be to explore germplasm with more bioactive components and the mechanism by which it can increase protein use efficiency in livestock rumen.



Depositing NZ's forage seed collection at the Global Seed Vault.
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The expanded understanding of the needs of a future-ready agriculture by scientists, the industry and farmers has directed research toward increased acknowledgement of the benefits of new forage species.

Hybrids of current and new species may also be another end product of the incremental change approach. These hybrids are implausible to transpire in nature, except when there are exceptional circumstances, e.g. very remote populations without other populations from the same species. The aim of these types of hybridisation would be to increase root biomass (persistence), shoot biomass, phosphate use efficiency, drought tolerance, disease resistance, nitrogen use efficiency etc that have been diluted in the genepool of the current forage species in New Zealand. However to show these features the hybrid may be dependent on coordinately controlled activity of plant proteins and metabolites associated with environmental factors.

Strategies for cost reduction of fertiliser in pastures are aimed at the inefficiencies of white clover. Some hybrids of white clover (*Trifolium repens*) and its closely-related wild species (*Trifolium uniflorum*) have shown higher biomass than white clover at Olsen P 9-20, increasing phosphate use efficiency (PUE) for future white clover hybrids. Adding to other effects of change, the water availability will be down by ~5-7.5% between 2030 and 2045 as a result of less precipitation or rainfall. Other hybrids of this cross-combination have maintained 10% soil moisture at 20 cm after almost four months with no irrigation, promising some white clover alternatives for this scenario. If they were irrigated weekly they maintained 30% of soil moisture at 20 cm. This is impressive knowing that soil moisture content is critical in biomass accumulation, run-off, soil conservation and evapotranspiration.

Transformational change

Products or species/cultivars resulting from the transformational change approach will be harder to take in and accept. Not only will there be novel species, new practices will also be needed for the successful implementation of this approach. However the expanded understanding of the needs of a future-ready agriculture by scientists, the industry and farmers has directed research toward increased acknowledgement of the benefits of new forage species.

Resilience and adaptability to different soil types and environments is a very important factor in the selection of species and their cultivars in summer dry and dryland agriculture. Subterranean clover (*Trifolium subterraneum* L.) is the most widely-sown species in Australia with three subspecies. The promise of this species to offer nutritious forage for farm animals and increase soil quality was recognised by A.W. Howard of Australia. Subspecies *subterraneum* tolerates grazing if managed well. It grows favourably in well-drained soils, while subspecies *yannicum* is a perfect option for poorly-drained soils, both adapted to soil pH_{Ca} 4.5-6.5. Subspecies *brachycalycinum*, however, is recommended for cracking clays and stony soils and grows in pH_{Ca} 6.0-9.0.

There are 45 registered cultivars of subterranean clover in the public registrar or with the Plant Breeders Rights (PBR) Office in Australia, eight introductions from the wild and 21 crossbreds. Also, the germplasm of this species has been recently rationalised to a representative 97 accessions (core collection) out of the original 3,000

wild populations. This unique resource provides an unprecedented opportunity for New Zealand to discover new traits in this species across its diverse hill country. In summer dry hill country, it seems, we continuously fight against the hostile atmospheric conditions. The core collection consists of subterranean clover populations with potential drought tolerance (range of annual rainfall at collection site: 275-1200 mm), which can lead into developing cultivars for this region. Cold tolerance screening of this core collection is in progress in Germany, the results of which can be used to select the appropriate approach for developing cold tolerant subterranean clover for New Zealand. The availability of high-throughput screening of this trait via simple and non-invasive methods will speed up this development process.

Estimating the depth of a plant's underground structure is also a criterion for agricultural practices, with the aim of both yield and preservation of the soil and environment, and leads to sensible research decisions. *Biserrula* (*Biserrula pelecinus* L.) is a hard-seeded annual forage legume persistent in Mediterranean farming systems. It has a deep root system (more than 2m in unrestricted soils) and a significant seed yield of 300-1500 kg/ha, with an average of 17 seeds per pod. If low contamination seed packages, with the highest vigour in combination with the right germplasm and with up to an average of 24 seeds per pod, are chosen then the maximum yield is certainly achievable. It is adapted to a wide range of acid and alkaline soil types, and under heavy grazing adopts a prostrate growth habit that protects it from over-grazing. Appropriate management strategies and another forage species are needed when feeding animals with *biserrula* due to potential photosensitivity effects. It is also well suited to intensive crop-pasture rotations, thus minimising impacts on soil (health, nutrient deficiency etc). *Biserrula* has provided herbage yield as high as 11 t/ha in Western Australia.

NZ's gene bank contributing to change

New Zealand's only standard and internationally registered gene bank, the Margot Forde Germplasm Centre (MFGC), is based in AgResearch's Grasslands Campus in Palmerston North. The Centre has a set of tools, data and expertise to collect germplasm from around the globe and accumulate the diverse seed collections. It is also capable of making this collection of use to the breeders and research community for plant improvement and cultivar development. The information used by staff is mainly categorised into three groups: origin data, phenotypic data and genotypic data.

Origin data

The first group or origin data comprises: ecological data such as information about the structure or dynamics of the collected population and its neighbouring species; geographical data such as latitude and longitude or the GPS coordinates of the collection site on a map; and

passport data such as soil pH, soil texture, climate.

Phenotypic data

The second group or phenotypic data comprises: agronomic data such as flowering time, hardseededness, cold tolerance, drought tolerance; morphological data such as hairiness, leaf colour patterns; and biochemical or physiological data such as primary and secondary compounds, chlorophyll content. To produce secondary compounds, plants operate an extremely complex process where factors such as biosynthesis, accumulation and expression play important roles.

The complexity of these phenotypic traits must therefore be taken into account at the time of the germplasm rationalisation process. Techniques ranging from traditional chromatography to mass spectrometry can be used to obtain and interpret these data. Techniques such as gas chromatography mass spectrometry combine both powerful methods to identify compounds that are extremely hard to detect and conduct quantitative measurements.

Genotypic data

The third group or genotypic data comprises: DNA marker data such as amplified fragment length polymorphisms (AFLPs); simple sequence repeats (SSRs); single nucleotide polymorphisms (SNPs); and DNA sequencing data such as genotyping by sequencing, whole genome sequencing, exome sequencing. The existence of many of the markers, as well as their differences, and methodologies for their use require cautious consideration before choosing such methods. Sequencing means the identification of the arrangement of chemical compounds – coded as A, T, C and G – that form a genome.

Multivariate data for faster breeding

All these three main sets of data are interrelated. A good example of this is the different morphological types from populations collected in islands that normally show the uppermost genetic variation and the broadest range in phenotypic variation as well. In this particular case, origin, phenotypic and genotypic data are highly correlated. Together, these three sets of data provide 'multivariate data' to increase the chance of selecting the right and promising germplasm or population for faster breeding by targeting traits of interest. This practice is called pre-breeding and the process is currently being streamlined at the Centre.

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Staff member working inside the cold room at the Margot Forde Germplasm Centre