Barley Genetics Newsletter (2009) 39:20-23

Historic barley seed samples and their utility

M. W. Leino[1*] and J. Hagenblad[2,3]

[1] Swedish Museum of Cultural History, SE-643 98 Julita, Sweden
[2] Uppsala University, Department of Ecology and Evolution, SE-752 36 Uppsala, Sweden
[3] Norwegian Univ. of Science and Technology, Dep of Biology, N-7491 Trondheim, Norway
*Author for correspondence: Matti.Leino@nordiskamuseet.se

Gene banks around the world hold over 370.000 accessions of barley (van Hintum and Menting, 2003). Although a large number of landraces and obsolete cultivars are stored in genebanks the geographic coverage of this conserved material is far from evenly distributed. For example only a handful of landraces from Northern Europe have been conserved (Jones et al, 2008a). Thus studies of geographic differences in land race barley are hampered by the availability of suitable material.

Another major drawback with gene bank material is the authenticity of the available accessions. Evolutionary forces such as drift and selection may have affected the genetic diversity since the accessions were deposited in gene banks and there is also a risk of contamination from poor propagation practices (Parzies et al., 2000). Furthermore, the true geographic origin of cultivars may be unknown or uncertain.

Recently material from non-viable historic collections has become recognized as an alternative to the use of gene bank material. Herbarium material can be well documented and therefore associated with useful geographic information, but the amount of material available is often limited and successful genotyping can prove problematic (Savolainen et al., 1995). In contrast, seed collections can contain equally well-documented samples with large number of seeds from each accession, thus enabling studies on a population level. Moreover, DNA quality of aged seeds widely surpasses that of other equally old vegetative tissue like straw, leaves and chaff (Lister et al., 2008).

In Sweden there are two large seed collections, both assembled at the turn of the last century. The collection at the Swedish Museum of Cultural History contains 419 different accessions of barley from 1865 – 1918 (Leino et al., 2009). These samples were collected ‘on farm’ from many different counties across Sweden and Finland with a minor addition of accessions from the rest of Europe (Fig. 1). Another seed collection is stored at Vänersborg Museum (Johansson, 2003) and contains 61 barley accessions. The samples in this collection originate from an agricultural exhibition in 1882. The seeds are in both cases stored in sealed glass containers and were displayed on exhibitions and museums but have never regenerated (Fig. 2).

We have genetically analysed part of the collection at the Swedish Museum of Cultural History (Leino *et al.*, 2009). The DNA of these 90 to 140-year-old seeds is severely degraded, but in spite of this degradation PCR amplification is possible, in particular of fragments less than 300 bp long (Fig. 3). We have successfully amplified not only multi-copy genes such ribosomal DNA, but also single copy genes like microsatellites.

Figure 1. Barley harvest in the county Ångermanland, Sweden in 1905. Photo: Petrus Hedström, Swedish Museum of Cultural History.

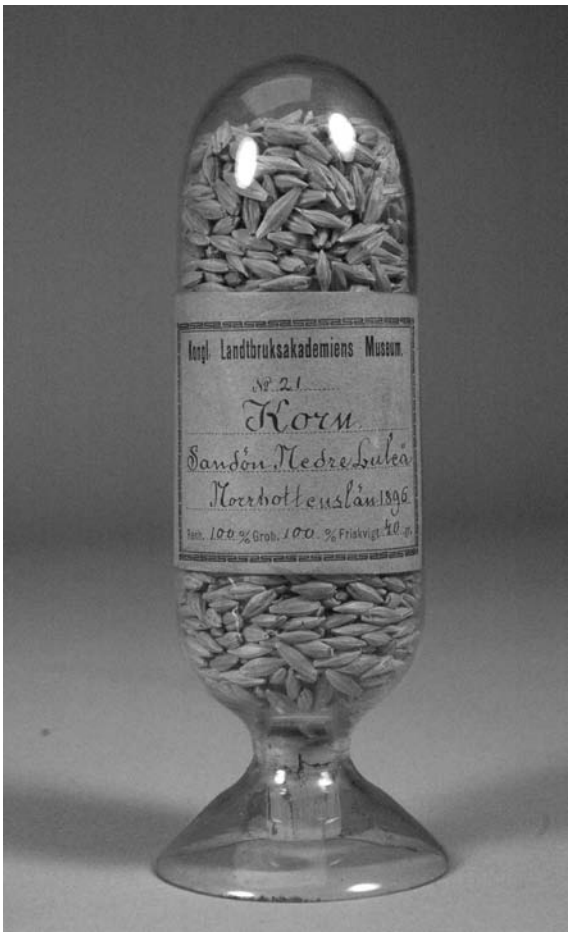

Figure 2. Examples of glass containers with barley seed stored at the museum.

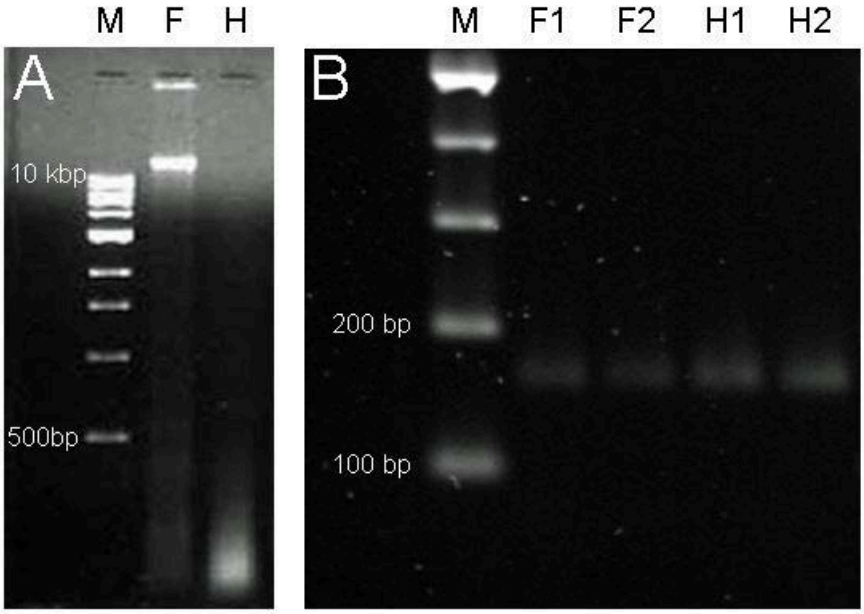

Figure 3. Analysis of DNA quality in fresh and historic barley seed. DNA isolations are made from single seeds. A) Electrophoresed total DNA. B) PCR amplified microsatellite marker fragments. M=DNA size marker, F=fresh seed, H=historic seed.

Recently, we have conducted a population genetic study of Swedish landrace barley using 113 year-old seed samples (Leino and Hagenblad, 2009). Our aim was to investigate the effects of bottlenecks, adaptation and gene flow on genetic diversity within and between landrace populations. The results demonstrated varying levels of within-population variation with some decline in diversity for more Northern populations. We could also detect clear population differentiation, and population structure within Sweden into a Southern and a Northern cluster. These results possibly reflect different introduction routes of barley into Sweden.

Historic material has also been used to study the geographic distribution of functional genes. In a study of the flowering time gene *Ppd-H1* in gene bank material originating across Europe, Jones *et al*. (2008b) showed geographic differentiation, with one allele gradually replacing the other when going north across Europe. Lister *et al*. (2009) repeated the study using historic samples, including several samples from the Vänersborg collection. In this study a much clearer picture was reported with one allele being present north of the Alps and the other in southern Europe. We have found the same pattern in this gene with both alleles being present in gene bank material originating from Sweden, but only a single allele present in historic material from the same region (Knöppel, unpublished). This further illustrates the presence of ambiguous material in genebank collections.

Non-viable historic barley material is a valuable complement to extant gene bank accessions, especially for geographical areas where gene bank material is rare or non-existent. Further studies of neutral and functional markers may shed additional light on the history of barley cultivation and breeding. It is thus imperative that historic material receives the same conservation attention as extant material in our gene banks.

References:

Hintum, Th. van, and Menting, F. 2003. Diversity in *ex situ* genebank collections of barley. In: Bothmer, R. von, Hintum, Th. van, Knüpffer, H., Sato, K. (eds) Diversity in Barley (*Hordeum vulgare)*. Elsevier Science B.V., Amsterdam, The Netherlands. p. 29-52.

Johansson, P. 2003. Prof av vårt svenska frö- Vestra Sveriges landtbruks-museum. In Johansson, P., Aarsrud, S., Öberg, A.-C. (eds.) Det underbara huset: Vänersborgs museum. Vänersborgs museums skriftserie 1:115-122.

Jones, H., Lister, D.L., Bower, M.A., Leigh, F.J., Smith, L.M.J., Jones, M.K. 2008a. Approaches and constraints of using existing landraces and extant plant material to understand agricultural spread in prehistory. Plant. Gen. Res. 6:98-112.

Jones, H., Leigh, F.J., Mackay, I., Bower, M.A., Smith, L.M., Charles, M.P., Jones, G., Jones, M.K., Brown, T.A., Powell, W. 2008b. Population-based resequencing reveals that the flowering time adaptation of cultivated barley originated east of the Fertile Crescent. Mol. Biol. Evol. 25:2211-2219.

Leino, M.W. and Hagenblad J. 2009. 19th century seeds reveal the population genetics of landrace barley (*Hordeum vulgare*). Mol. Biol. Evol. in press.

Leino, M.W., Hagenblad, J., Edqvist, J., Karlsson Strese E.-M. 2009. DNA preservation and utility of a historic seed collection. Seed Sci. Res. 19:125-135.

Lister, D.L., Bower, M.A., Howe, C.J., Jones, M.K. 2008. Extraction and amplification of nuclear DNA from herbarium specimens of emmer wheat: a method for assessing DNA preservation by maximum amplicon length recovery. Taxon 57:254-258.

Lister, D.L., Thaw, S., Bower, M.A., Jones, H., Charles, M.P., Jones, G., Smith, L.M.J., Howe, C.J., Brown, T.A., Jones, M.K. 2009. Latitudinal variation in a photoperiod response gene in European barley: insight into the dynamics of agricultural spread from 'historic' specimens. J Arch. Sci. 36:1092–1098.

Parzies, H.K., Spoor, W., Ennos, R.A. 2000. Genetic diversity of barley landrace accessions (*Hordeum vulgare* ssp. *vulgare*) conserved for different lengths of time in *ex situ* gene banks. Heredity 84:476-486.

Savolainen, V., Cuénoud, P., Spichiger, R., Martinez, M.D., Crèvecoeur, M., Manen, J.-F. 1995. The use of herbarium specimens in DNA phylogenetics: evaluation and improvement. Pl. Syst. Evol. 197:87-98.