



**BMT/9/9 Add.**

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**INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS**  
GENEVA

**WORKING GROUP ON BIOCHEMICAL AND MOLECULAR  
TECHNIQUES AND DNA PROFILING IN PARTICULAR**

**Ninth Session**  
**Washington, D.C., June 21 to 23, 2005**

**SNPS IN BARLEY: A POTENTIAL "OPTION 1" APPROACH**

*Document prepared by experts from the United Kingdom*

1. The BMT agreed that, where agreed by the relevant experts, the presentations made at the meeting should be made available in the BMT document section of the UPOV website, as addenda to the relevant documents. This document contains a copy of the presentation made by Mr. Robert Cooke (United Kingdom) for document BMT/9/9.



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## ***SNPs in Barley***

**SNPs in Barley:**

- the 5 loci had separation coefficients of between 0.12 - 0.49;
- the 132 varieties could be separated into 20 groups (with 1 - 16 varieties);
- the overall separation rate was 98% (criterion = one locus difference);
- allelic status of ABG601 correlated to winter/spring character - linked to *Sgh1* locus (vernalisation)?

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## ***SNPs in Barley***

*vrn1 vrn2* Spring DV92: *VRN2* mutation → Inactive repressor → Flowering without vernalization

*vrn1 Vrn2* Winter G1777 G3116: Vernalization → Inactivation of *Vrn2* repressor → *vrn1* promoter → Flowering only after vernalization

*Vrn1 Vrn2* Spring G2528: Promoter mutation → *VRN2* not recognized → Flowering without vernalization

**Molecular Basis of Vernalisation:** Genetic studies have shown that variation at two major gene loci underlies the differing vernalisation requirement of spring versus winter cereals. The loci, designated *Vrn1* and *Vrn2*, have been comparatively mapped.

***VrnH1* - chromosome 5H**  
***VrnH2* - chromosome 4H**

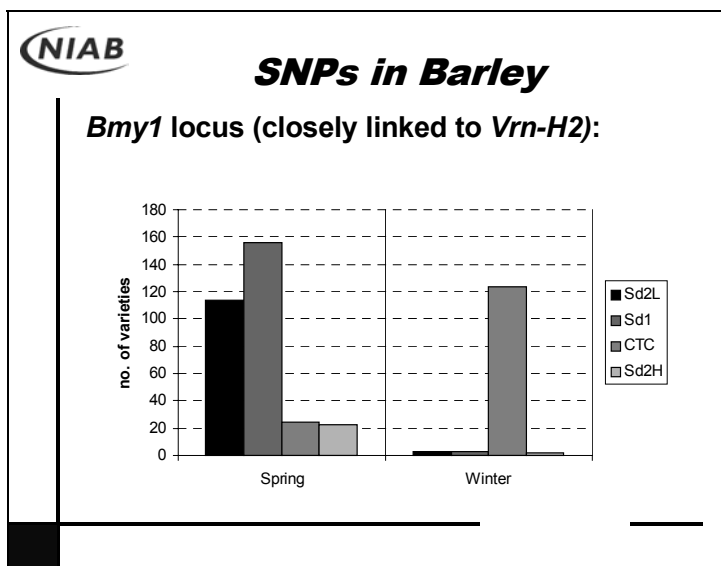
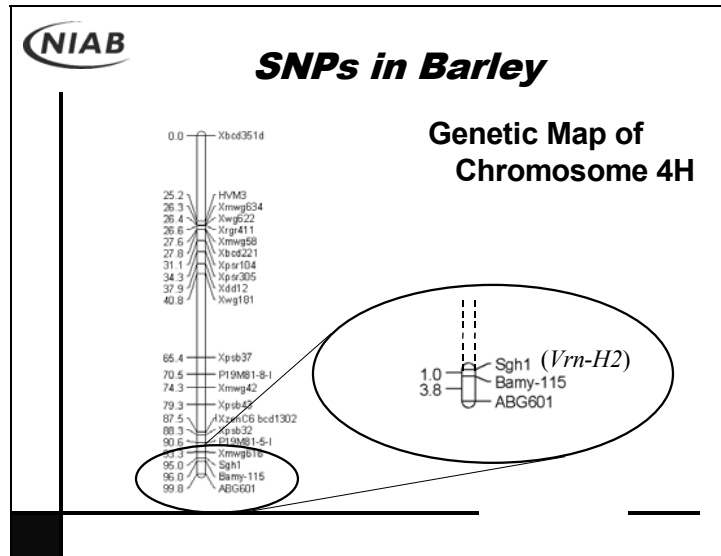
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## ***SNPs in Barley***

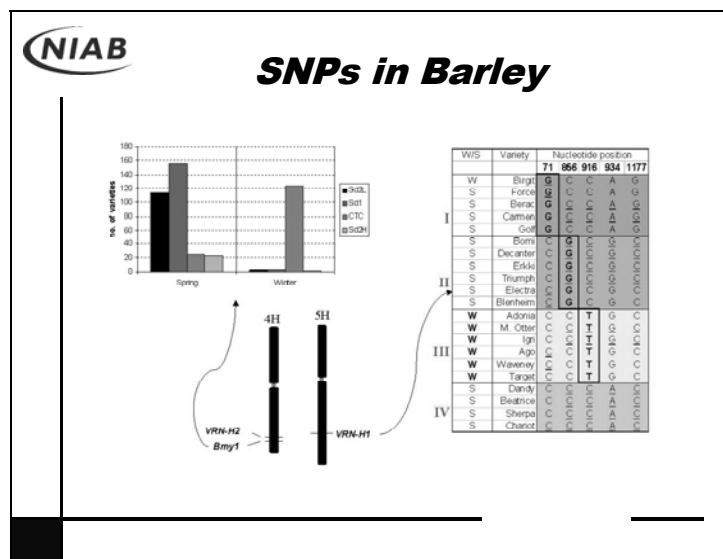
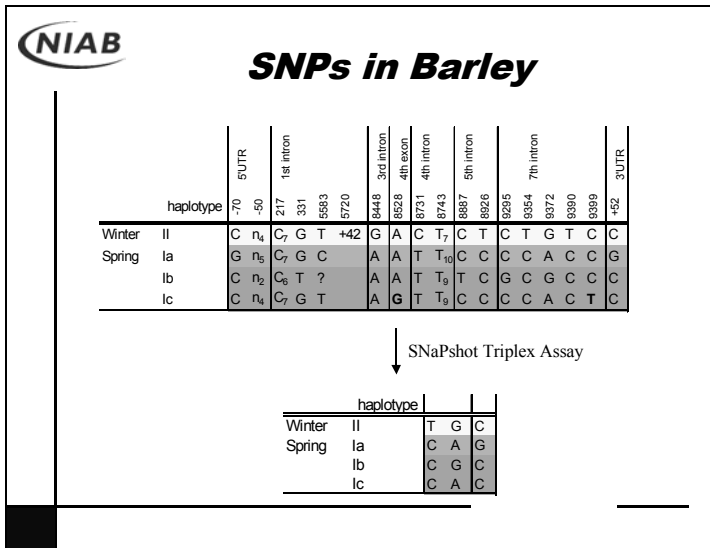
**EU Gediflux Project:**

For the *Bmy1* locus (linked to ABG601 & closely linked to *Vrn-H2* on chromosome 4H), a triplex SNP assay was scored on 467 varieties, giving four haplotypes designated Sd2L, Sd1, CTC and Sd2H.

Portions of the *Vrn-H1* gene were sequenced from a panel of 21 varieties, leading to the identification of 5 SNPs which split the Gediflux varieties into 4 haplotypes, one of which contained only winter varieties.



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- ### SNPs in Barley
- *Vrn H1* - candidate gene (BM5) identified from database, and nucleotide diversity examined in a small number of varieties
  - Specific genotyping assay developed:



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### SNPs in Barley

**Current Project:-**

**Hypothesis:** Characteristic haplotype combinations at the *Vrn-H1* and *Vrn-H2* loci, which can be rapidly identified via a SNP assay, will be diagnostic for seasonal growth habit.

The significance of this would be the opportunity to develop an Option 1- type approach to the use of molecular markers in DUS testing.

**defra**  
Department for Environment  
Food and Rural Affairs

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### ***SNPs in Barley***

In barley, the current UK protocol for evaluating seasonal growth habit stipulates that – *“In the case of winter varieties an additional 50 ear-rows are sown in late April during the first year of tests to examine the uniformity of the vernalisation response of the variety under test.”*

The scoring of this characteristic therefore entails a dedicated field trial. A quick & cost-effective molecular test for vernalisation has attractions, as a direct replacement for an existing field-based characteristic.

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### ***SNPs in Barley***

**Objectives:**

- more comprehensive haplotyping of H1
- analysis of H2 (rather than linked gene) + haplotyping

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### ***SNPs in Barley***

**H1 - BM5 haplotype distribution in the Gediflux barley set (467 varieties):**

Season	Ia	Ib	Ic	II
Spring	~150	~150	~10	~15
Winter	~5	~5	~5	~130

**‘Spring’ varieties being investigated.**

**Are they really winter types?**

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### ***SNPs in Barley***

- H2 - candidate gene identified (ZCCT-type)
- Appear to be 3 of these in barley
- Sequence data available
- Haplotyping underway
- Analyse 100 varieties of known seasonal type (H1 and H2)

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### ***SNPs in Barley***

- Design of SNP-based assay for winter/spring type determination in barley will require information from both H1 and H2 genes
- Cost-effective assay should become possible in due course.

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### ***DUS: Molecular Markers***

**Molecular Markers in DUS Testing : SNP Haplotyping for Assessment of Seasonal Type in Barley**

Use of molecular markers to predict winter/spring characteristic.

