Towards the Development of a Chromosome Segment Substitution Library (CSSL) for Avalon x Cadenza (AxC):

High-Density Maps of 18 AxC NILs generated on the Axiom® HD Wheat Genotyping Array

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The Avalon x Cadenza Doubled Haploid Population (AxC) was one of several developed to represent a broad spectrum of the variation present in the UK elite winter germplasm pool. The population is a Defra UK reference population and was developed as part of WGIN1. The AxC population has been extensively phenotyped in field trials in Norfolk from 2005-2008 and a number of QTL's affecting ear emergence, crop height and yield have been identified (Griffiths *et al* 2009, Gegas *et al* 2010, Griffiths *et al* 2012, Ma *et al* 2015). 552 individual streams from DH lines carrying a single QTL affecting plant heading date, height or yield were generated; 250 lines in an Avalon background and 302 lines in a Cadenza background. Eighteen of these NILs representing most of the identified QTLs were selected for this preliminary study (Table 1).

Table 1

NIL	QTL	Background
AC33_E11_5_10	1D ear emergence	Cadenza
AC34_E61_4_13	3B yield	Cadenza
AC43_E55_6_62	6A height	Cadenza
AC49_E14_1_11	5A yield	Avalon
AC49_E16_2_19	1D ear emergence	Avalon
AC51_E43_9_17	2D yield	Avalon
AC69_E44_6_67	3A height	Avalon
AC69_E70_5_18	7D yield	Avalon
AC75_E101_2_10	6B ear emergence / height	Cadenza
AC89_E5_1_37	6A height	Cadenza
AC101_E81_10_55	6B height	Cadenza
AC104_E96_3_13	1B ear emergence	Cadenza
AC105_E92_2_16	7B yield	Cadenza
AC113_E113_10_72	3A height	Cadenza
AC148_E47_6_47	2D height	Avalon
AC160_E28_1_71	3B height	Cadenza
AC162_E21_8_71	2D height	Cadenza
AC195_E13_4_21	1B ear emergence	Avalon

The NILs are in the Avalon or Cadenza backgrounds respectively, with the QTL region coming from Cadenza or Avalon donors', respectively. In addition other chromosomal segments from the donor parents are likely to be randomly distributed across the genome. Figure 1 shows a *hypothetical* example of this.

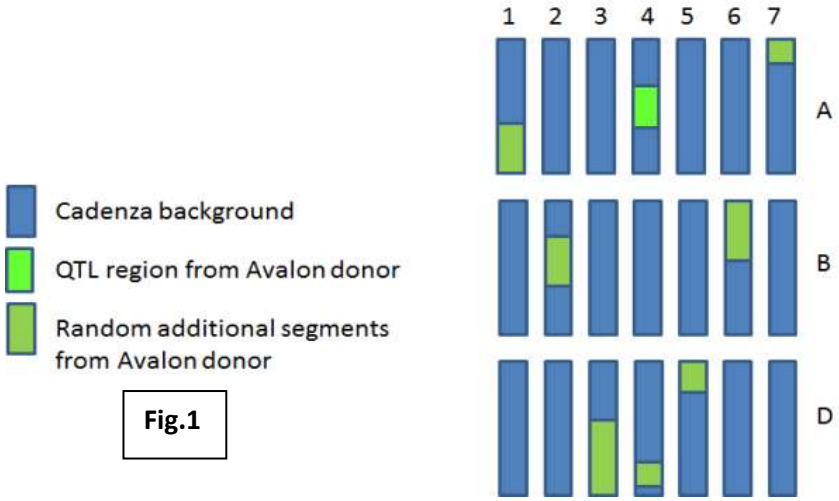

Fig.1

The **QTL region** (from Avalon on 4A in this *hypothetical* NIL) had been selected for using flanking molecular markers, however additional **chromosomal segments** will also have been randomly introduced into the Cadenza background in each NIL. Only by whole genome genotyping is it possible to determine the location and extent of these regions.

The University of Bristol Genomics Facility have developed a high-density wheat genotyping array using the Affymetrix Axiom® platform, known as the Axiom® HD Wheat Genotyping Array. The array, (also known as the 820K Axiom Array) contains nearly 820,000 exome-captured SNP sequences derived from hexaploid wheat accessions, as well as diploid and tetraploid progenitor accessions and wheat relatives. The 18 NILs were genotyped at the UoB Genomics Facility on the 820K Axiom® Array and the segregation data analysed (Figure 2). This suggested that the **NILs have an average of 12.5% donor background**, which includes both the selected QTL region and the random segments**.**

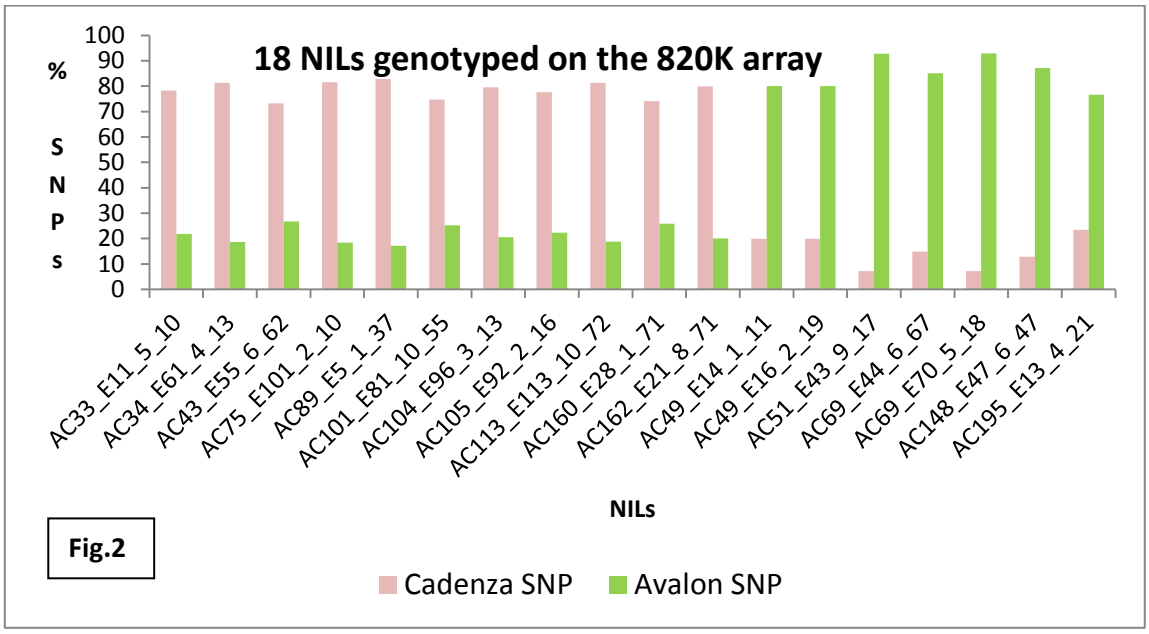

Fig.2

The UoB Genomics Facility has recently published maps from the Avalon x Cadenza mapping population using the 820K Axiom® Array data, Wingfield *et al* (2015). The **high-density map** consists of **180942 markers** but a chromosome **'frame map'** was also constructed, with a single marker representing a bin of markers at each chromosomal position, consisting of **1286 markers**.

We used the 820K Axiom® Array genotyping data from the 18 NILs and produced maps of each NIL using the frame map generated by Wingfield *et al* (2015) to determine the order of the markers. Where possible we have used the same markers as the frame map but if these are not found in the Breeders' 35K Axiom® Array* we have selected another marker at the same position. Additional criteria for the selection of markers were whether a marker was associated with a KASP marker from the UoB Genomics Facility and, where possible, these markers to be co-dominant. This map consists of 1260 markers. The markers on the frame map, and those selected on the 820K and 35K maps are compared [**PDF: Frame v 820K v 35K**].

The genotyping data from the 18 NILs are presented in two formats: *all* the chromosomes of an individual NIL [**PDF: NILS**] and the *individual chromosomes*, for each genome, from all 18 NILs, [**PDFs: A genome, B genome and D genome**]. Markers where the chromosome is Avalon are **black**, Cadenza **red**, heterozygous markers are **blue** and markers which are not present or un-scorable are **grey**. The positions of the KASP markers relative to the Axiom markers are also shown on maps [**PDF: 820K and BS markers**].

*The next stage of this project is to genotype another 94 NILs, this time using the Breeders' 35K Axiom® Array. These data, along with that from the original 18 NILs, should allow the generation of a large number of lines each containing a defined chromosomal segment from Avalon or Cadenza, ideally in an otherwise clean Cadenza or Avalon background, respectively. The whole collection of lines should form a 'Tiling path' across the whole genome so breeders will be able to select the line containing their region of interest to make crosses.

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