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Project Title: Construction and Utilization of a BAC Library of Sumai 3.

PROJECT 1 ABSTRACT (1 Page Limit)

Funded by USWBSI and NRI, we are using a map-based cloning approach to clone the major FHB resistance QTL for wheat on chromosome 3BS (*Fhb1*). Based on FHB phenotypes of recombinants and new DNA markers derived from 'Chinese Spring' BAC sequences, *Fhb1* was narrowed down to a 260 Kb region and seven candidate genes were identified. Five of the seven candidate genes have been tested via tranformation and none of them encode *Fhb1* based on the FHB phenotypes of the transgenic plants. The two remaining candidate genes will be tested in fall 2007 and spring 2008.

It is possible that Chinese Spring has a null *Fhb1* allele. In this case, it would be very difficult if not impossible to clone *Fhb1* based on the BAC sequence of Chinese Spring. Therefore, the availability of a BAC library of Sumai 3 is an essential resource in cloning *Fhb1* as well as other FHB QTLs. The specific objectives of this proposal are to:

1) Construct a BAC library of Sumai 3 for the wheat community.

2) Construct a Sumai 3 BAC contig spanning Fhb1.

3) Compare the sequences between Sumai 3 and Chinese Spring for the Fhb1 region to identify additional candidate genes, if any.

Considering the large genome size of common wheat (16,000 Mb) and the amount of funding required to construct a BAC library of Sumai 3, we propose to construct a five genome-equivalent BAC library. Our experiences with the construction and screening of a pooled cosmid library of Sumai 3 will be helpful for the proposed BAC library. We will use the Sumai 3-specific markers developed to screen our pooled cosmid library of Sumai 3 to screen 420 pools of BAC plate clones. After PCR-based identification of positive plate pools, single positive clones will be identified by DNA hybridization. We will sequence the genic regions of a BAC clone(s) of Sumai 3, and compare the sequences with the corresponding region of Chinese Spring to identify Sumai 3-specific nucleotides or sequences. We will compare the gene content and organization in the *Fhb1* region between Sumai 3 and Chinese Spring. We are especially interested in knowing if there are any gene insertions/deletions between these two genotypes. Any additional genes identified in Sumai 3 will be validated as a candidate for *Fhb1* by transformation and virus induced gene silencing.