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Introduction



- Grain texture is the single most important trait that determines end-use quality of wheat. The Hardness (Ha) locus on chromosome 5D is the main determinant of grain texture in hexaploid wheat. Three structurally related genes have been identified that are closely linked at this locus: Puroindoline-a (Pina-D1), Puroindoline-b (Pinb-D1) and Grain Softness Protein (Gsp-D1). Mutations in the Pina-D1 and Pinb-D1 genes are associated with increased grain hardness.
- There is currently a need to increase the range of texture available in bread and pasta wheat, to allow the range of end uses to be expanded. Such variation exists in wild and primitive species, in terms of grain hardness and puroindoline sequences. As wild relatives of wheat, *Aegilops* species contain genomes called D, S, G, U, C and M, constituting a valuable resource and an extensive reservoir of genetic variability for improving cultivated bread wheat and pasta wheat by wide crossing.

Table 1. Ten accessions of *Aegilops* from China studied.

Accessions	Taxonomic species	Genome
Q03-011	<i>Ae. kotschyi</i>	UUSS
Q03-038	<i>Ae. kotschyi</i>	UUSS
Q03-039	<i>Ae. kotschyi</i>	UUSS
Q03-012	<i>Ae. triuncialis</i>	UUCU
Q03-023	<i>Ae. longissima</i>	SS
Q03-026	<i>Ae. sharonensis</i>	SS
Q03-021	<i>Ae. bicornis</i>	SS
Q03-022	<i>Ae. bicornis</i>	SS
Q03-002	<i>Ae. tauschii</i>	DD
Q03-013	<i>Ae. juvenalis</i>	UUCU
Q03-011	<i>Ae. kotschyi</i>	UUSS

Figure 1: Ten accessions of *Aegilops* from China studied.

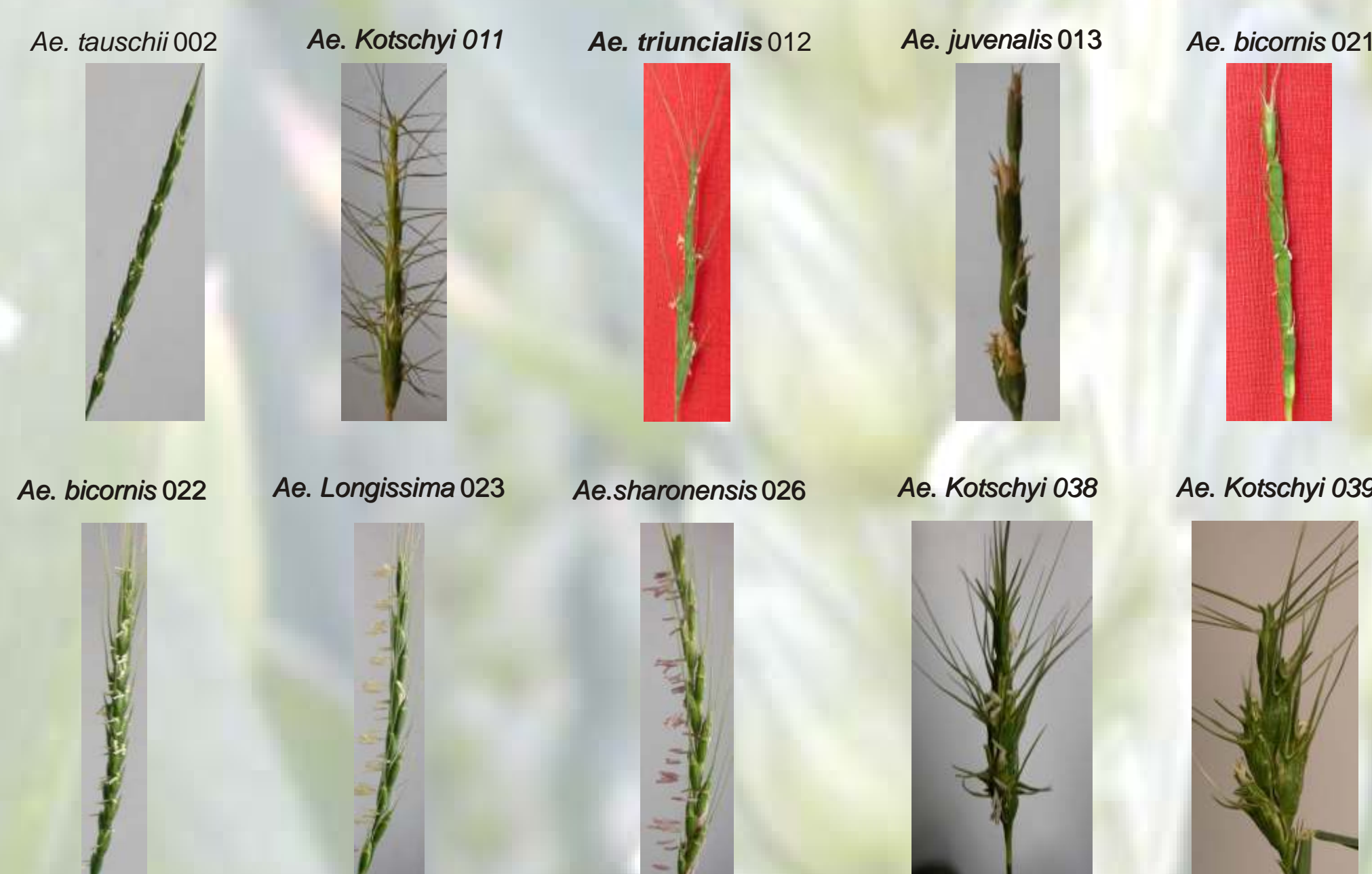


Figure 2. Puroindoline-a Amino acid sequence alignment of ten *Aegilops* accessions and *T. aestivum* HiLine (hard bread wheat, used as control)

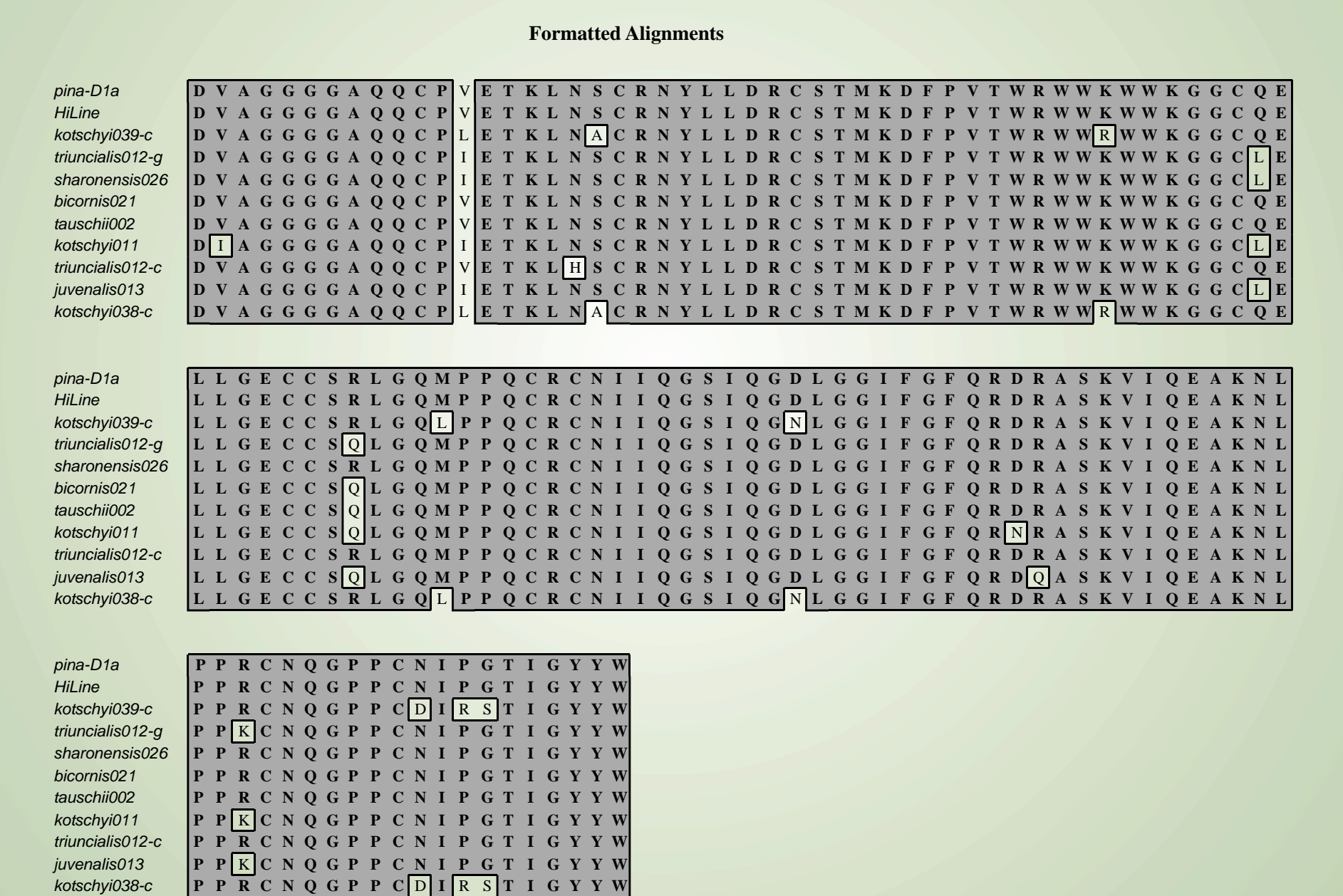
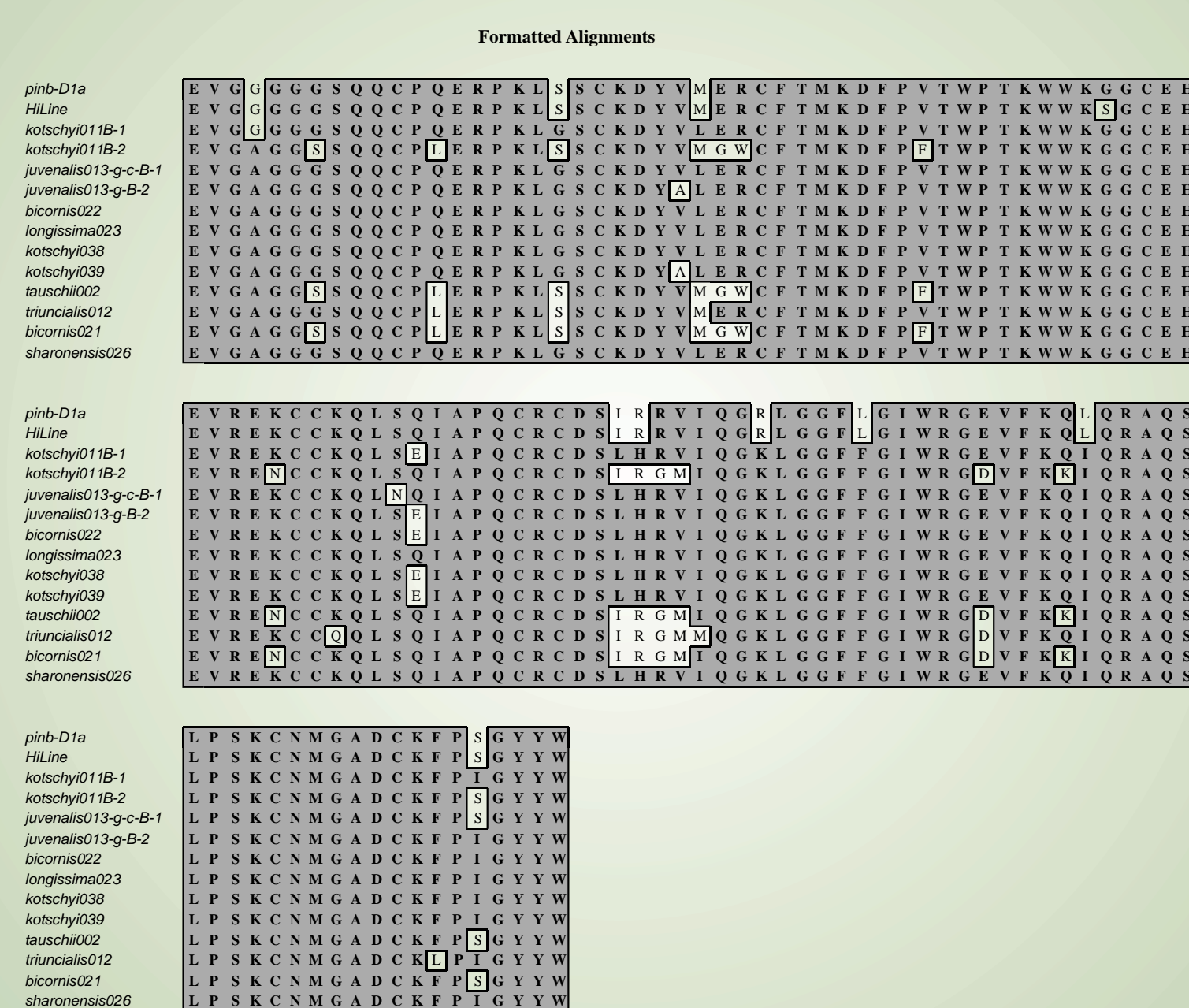


Figure 3. Puroindoline-b Amino acid sequence alignment of ten *Aegilops* accessions and *T. aestivum* HiLine



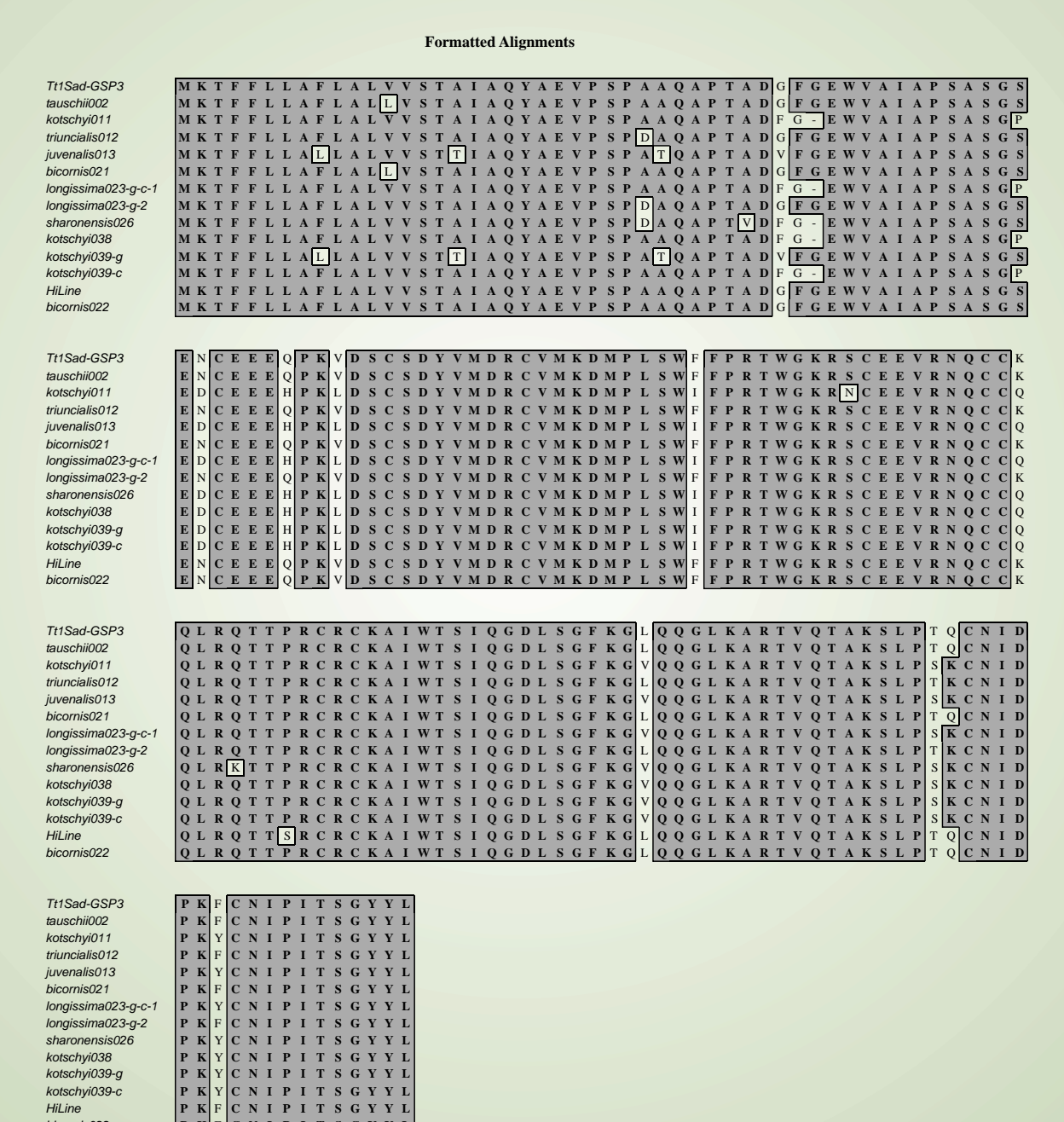
Results

- Eight *pina* and seven *pinb* mutant forms have been identified in *Aegilops* accessions by PCR amplification. This is in contrast with previous studies indicating far less variability for *Pina* with respect to *Pinb*. All the mutations are different from those reported so far.
- *GSP* sequences were also amplified from *Aegilops* accessions and 6 new mutants have been identified.
- Southern Blot analysis showed that most of the accessions have more than one copy of each of the three genes.
- Expression of *Friabilin* was confirmed by Western Blot analysis of grain total protein extracts.

Conclusion

Our study show that wild relatives of wheat represent a rich source of genetic variability for the improvement of hexaploid and pasta wheat. All the accessions examined contained in fact *pina*, *pinb* and *GSP* genes and several new mutant forms of the genes have been identified.

Figure 4. *GSP* Amino acid sequence alignment of ten *Aegilops* accessions and *T. aestivum* HiLine



Reference:

Craig F. Morris (2002) Puroindolines: the molecular genetic basis of wheat grain hardness *Plant Molecular Biology* 48: 633–647.

Acknowledgements:

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