

SUPPLEMENTARY DATA

Table S1. Primers for real-time PCR and RNA probe synthesis (underlined sequences for T7 RNA polymerase site).

Gene		Primer sequence	PCR product size	
ω -2	Forward	TGGCCAATGAAATGAACACC	120bp	Real-time PCR
	Reverse	GGTCGGGGTTACACATTATGG		
	Forward	GAATTG <u>TAAATACGACTCACTATAGGG</u> CCATATGGGAGTAGTCTTAC	180bp	Sense probe
	Reverse	GAACTTATTTGTCTTTTATTCCCATGTTTGG		
	Forward	CCATATGGGAGTAGTCTTAC	180bp	Antisense probe
	Reverse	GAATTG <u>TAAATACGACTCACTATAGGG</u> GAACTTATTTGTCTTTTATTCCCATGTTTGG		
ω -5	Forward	CAAGAACCTTCCCCATACCA	117bp	Real-time PCR
	Reverse	CAACGATGATTCACCCGTCT		
	Forward	GAATTG <u>TAAATACGACTCACTATAGGG</u> GGCAAGAACCTTCCCCATACCA	217bp	Sense probe
	Reverse	GAACTTATTTGTCTTTTATTCCCATGTTTGG		
	Forward	GGCAAGAACCTTCCCCATACCA	217bp	Antisense probe
	Reverse	GAATTG <u>TAAATACGACTCACTATAGGG</u> GAACTTATTTGTCTTTTATTCCCATGTTTGG		

Fig. S1. Response of ω -gliadins to nitrogen application and N-terminal sequencing in (A) 'Hereward' (reported by Shewry *et al.*, 2009b) and (B) 'Cordiale'. Fractions from grain grown at N100, N200, and N350 kg N ha⁻¹ are shown in lanes 1, 2 and 3, respectively.

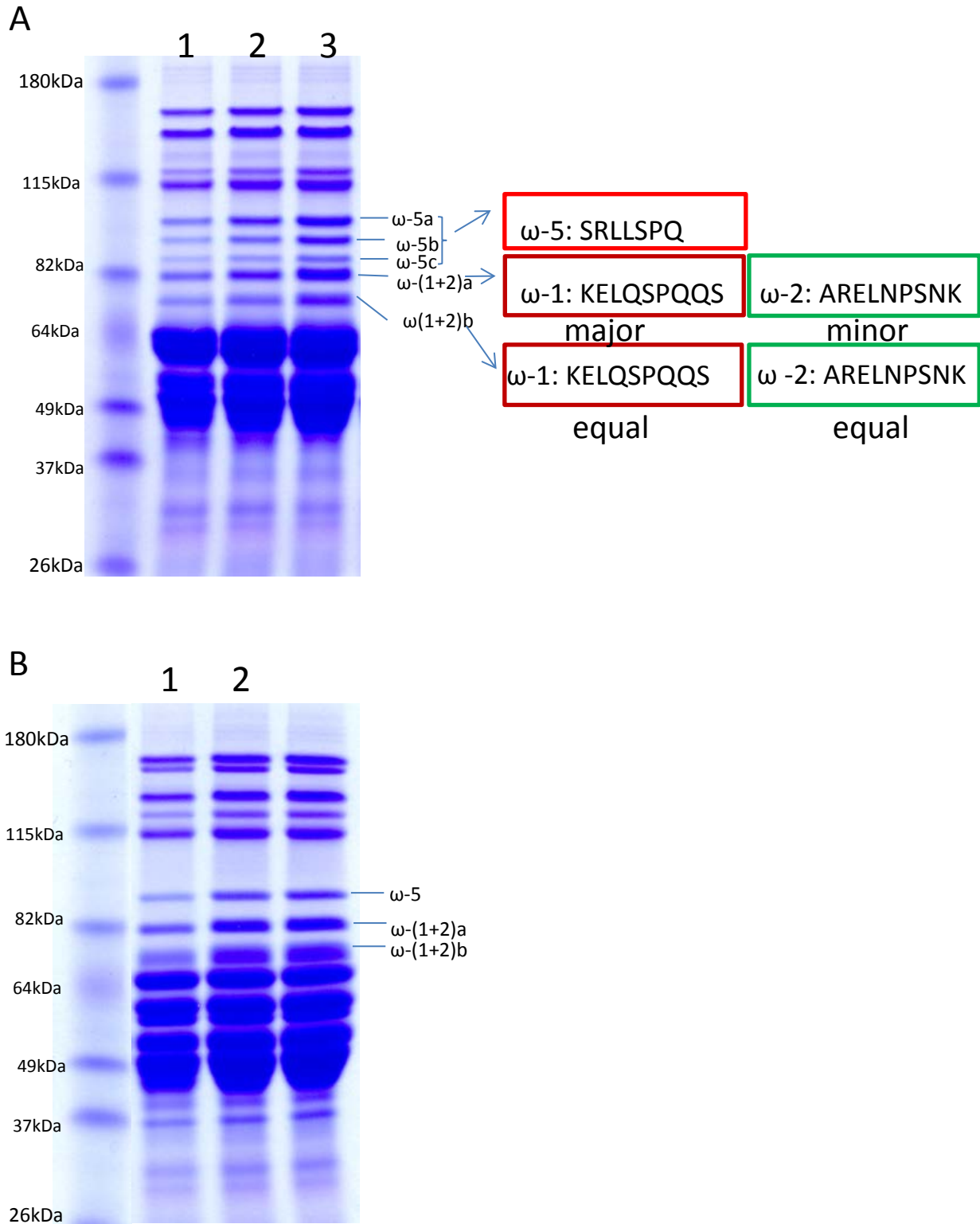


Fig. S2. Negative controls for *in situ* hybridisation using (A) the ω -2 gliadin sense probe and (B) the secondary antibody only for immunofluorescence analysis of ω -5 gliadin. Scale bars: 500 μ m.

