

## **Supplemental Material to:**

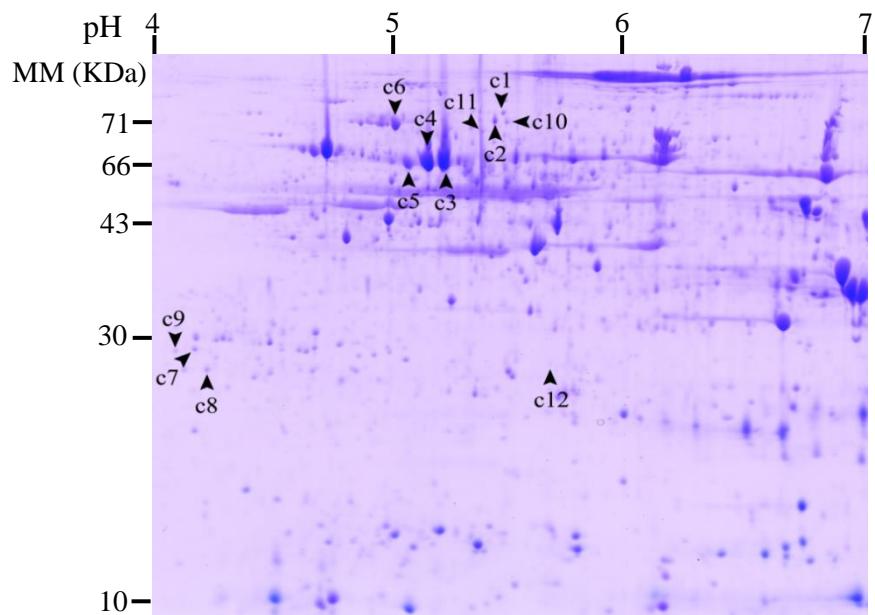
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and Belay T Ayele\***

**Wheat seed proteins regulated by imbibition independent  
of dormancy status**

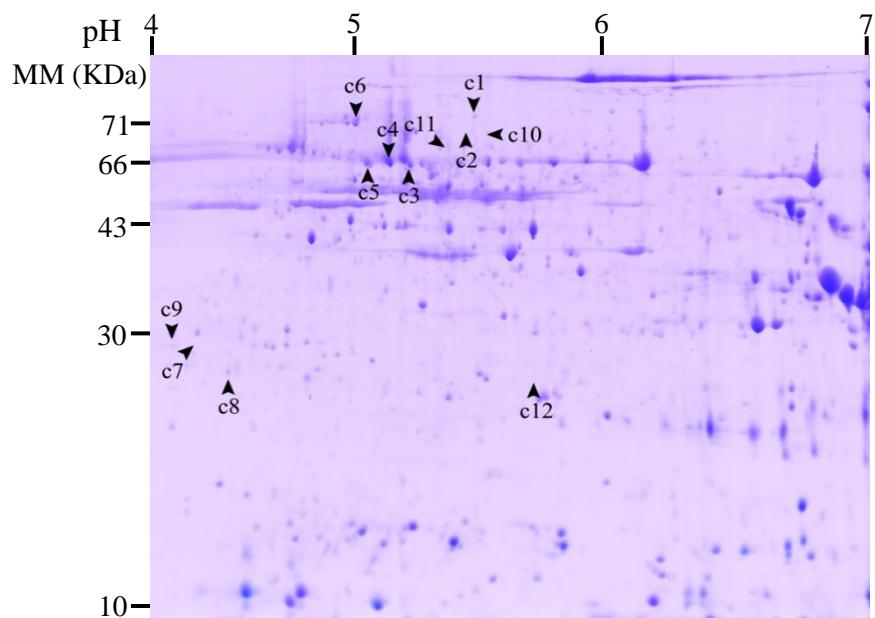
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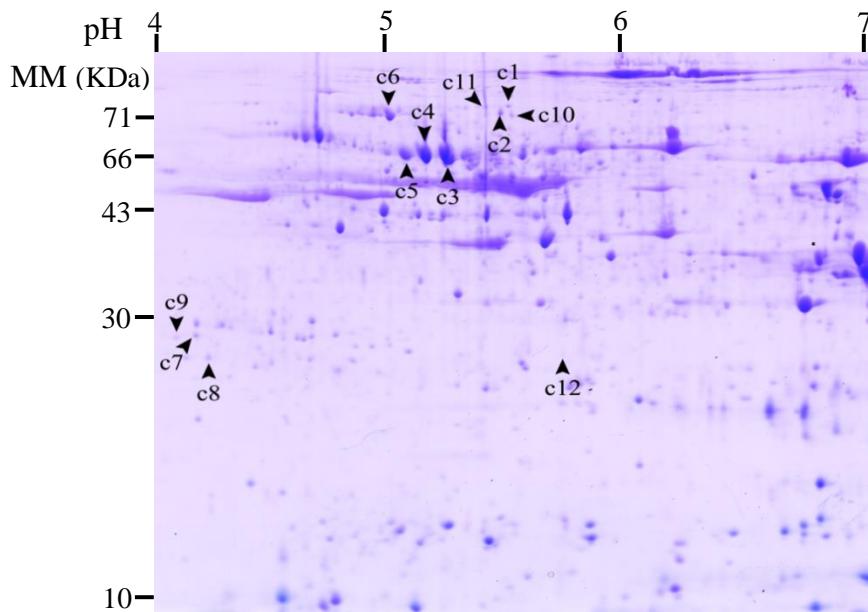
**A)**



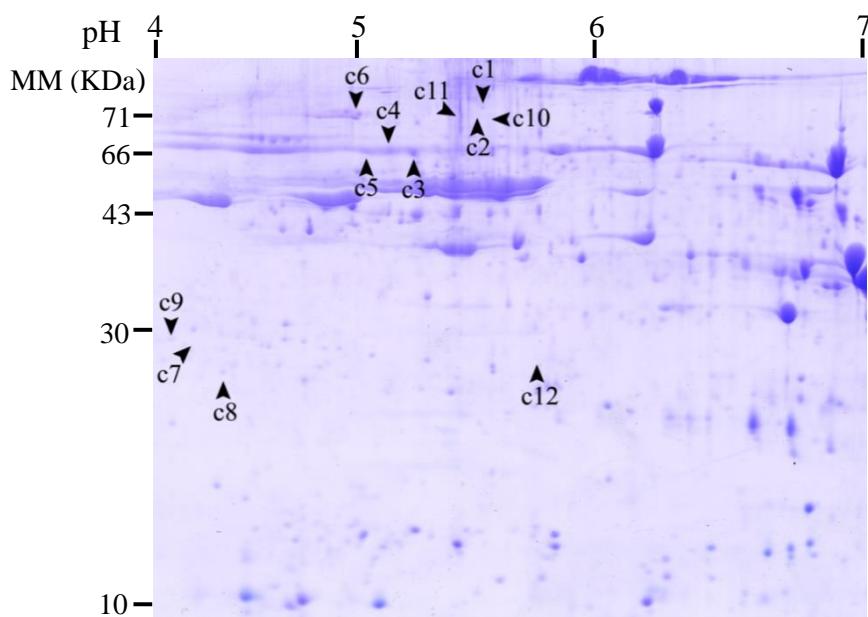
**B)**



C)



D)



**Figure S1.** Two-dimensional gel of dry dormant (A), imbibed dormant (B), dry after-ripened (C) and imbibed after-ripened (D) seeds of wheat using a linear pH gradient. Protein extraction and two-dimensional gel analysis were performed as described before<sup>3</sup>. All experiments were performed with three independent biological replicates.

**Table S1. Fold changes in the expression of probesets corresponding to proteins regulated by imbibition in both dormant and after-ripened seeds.**

Protein spot	Probeset ID	D-24/D-0	AR-24/AR-C	UniProt Desc
c1	Ta.1137.1.S1_at	-1.03	-1.32	Soluble starch synthase 1, chloroplastic/amyloplastic n=7 Tax=Oryza sativa RepID=SSY1_ORYSJ
c1	Ta.55.1.S1_x_at	1.06	1.08	Starch synthase II n=8 Tax=Triticeae RepID=Q8H1Y7_HORVD
c1	Ta.55.2.S1_x_at	1.26	-1.04	Starch synthase (Fragment) n=1 Tax=Triticum aestivum RepID=O24398_WHEAT
c1	Ta.55.2.S1_a_at	1.33	1.16	Starch synthase II n=8 Tax=Triticeae RepID=Q8H1Y7_HORVD
c1	Ta.55.2.S1_at	1.36	-1.00	Starch synthase (Fragment) n=1 Tax=Triticum aestivum RepID=O24398_WHEAT
c1	Ta.24114.6.S1_s_at	-1.48	-1.43	Granule-bound starch synthase 1, chloroplastic/amyloplastic n=29 Tax=Triticeae RepID=SSG1_WHEAT
c1	Ta.24114.3.S1_a_at	-2.15	-2.06	Granule-bound starch synthase 1, chloroplastic/amyloplastic n=29 Tax=Triticeae RepID=SSG1_WHEAT
c1	Ta.24114.3.S1_at	-1.05	-1.01	Granule-bound starch synthase 1, chloroplastic/amyloplastic n=29 Tax=Triticeae RepID=SSG1_WHEAT
c1	Ta.24114.2.S1_x_at	-2.59	-2.26	Granule-bound starch synthase 1, chloroplastic/amyloplastic n=29 Tax=Triticeae RepID=SSG1_WHEAT
c1	Ta.238.1.S1_at	-2.70	-5.74	Granule-bound starch synthase 1b, chloroplastic/amyloplastic (Fragment) n=2 Tax=Triticeae RepID=SSG1B_HORVU
c1	Ta.2795.1.S1_a_at	-1.59	-1.55	Granule-bound starch synthase n=2 Tax=Triticum aestivum RepID=Q9XEN9_WHEAT
c2	Ta.24114.6.S1_s_at	-1.48	-1.43	Granule-bound starch synthase 1, chloroplastic/amyloplastic n=29 Tax=Triticeae RepID=SSG1_WHEAT
c2	Ta.24114.3.S1_a_at	-2.15	-2.06	Granule-bound starch synthase 1, chloroplastic/amyloplastic n=29 Tax=Triticeae RepID=SSG1_WHEAT
c2	Ta.24114.3.S1_at	-1.05	-1.01	Granule-bound starch synthase 1, chloroplastic/amyloplastic n=29 Tax=Triticeae RepID=SSG1_WHEAT
c2	Ta.24114.2.S1_x_at	-2.59	-2.26	Granule-bound starch synthase 1, chloroplastic/amyloplastic n=29 Tax=Triticeae RepID=SSG1_WHEAT
c2	Ta.2795.1.S1_a_at	-1.59	-1.55	Granule-bound starch synthase n=2 Tax=Triticum aestivum RepID=Q9XEN9_WHEAT
c2	Ta.238.1.S1_at	-2.70	-5.74	Granule-bound starch synthase 1b, chloroplastic/amyloplastic (Fragment) n=2 Tax=Triticeae RepID=SSG1B_HORVU
c2	Ta.55.1.S1_x_at	1.06	1.08	Starch synthase II n=8 Tax=Triticeae RepID=Q8H1Y7_HORVD
c2	Ta.55.2.S1_x_at	1.26	-1.04	Starch synthase (Fragment) n=1 Tax=Triticum aestivum RepID=O24398_WHEAT
c2	Ta.55.2.S1_a_at	1.33	1.16	Starch synthase II n=8 Tax=Triticeae RepID=Q8H1Y7_HORVD
c2	Ta.55.2.S1_at	1.36	-1.00	Starch synthase (Fragment) n=1 Tax=Triticum aestivum RepID=O24398_WHEAT
c2	Ta.1137.1.S1_at	-1.03	-1.32	Soluble starch synthase 1, chloroplastic/amyloplastic n=7 Tax=Oryza sativa RepID=SSY1_ORYSJ
c3	Ta.3.1.S1_at	-1.14	-1.26	Beta-amylase n=2 Tax=Triticeae RepID=AMYB_WHEAT
c3	Ta.27780.1.S1_at	-7.97	-6.90	Beta-amylase n=12 Tax=Hordeum vulgare RepID=AMYB_HORVU
c3	Ta.27780.1.S1_x_at	-6.27	-5.52	Beta-amylase n=12 Tax=Hordeum vulgare RepID=AMYB_HORVU
c3	Ta.137.1.S1_at	-1.10	-1.23	Beta-amylase (Fragment) n=1 Tax=Triticum aestivum RepID=Q9ZR48_WHEAT
c3	Ta.4494.1.S1_x_at	-3.71	-8.39	Beta-amylase n=3 Tax=Oryza sativa RepID=Q10RZ1_ORYSJ
c3	Ta.4494.1.S1_at	-4.04	-8.61	Beta-amylase n=3 Tax=Oryza sativa RepID=Q10RZ1_ORYSJ
c3	Ta.1062.1.S1_at	1.10	-1.66	Beta-amylase n=1 Tax=Zea mays RepID=B6SVZ0_MAIZE
c3	TaAффx.68872.1.S1_at	-1.39	-1.61	Putative uncharacterized protein Sb01g028700 n=1 Tax=Sorghum bicolor RepID=C5WRG3_SORBI
c3	Ta.27780.2.A1_x_at	-10.60	-10.47	Beta-amylase (Fragment) n=1 Tax=Secale cereale RepID=AMYB_SECCE
c3	Ta.27780.2.A1_a_at	-10.48	-10.72	Beta-amylase n=12 Tax=Hordeum vulgare RepID=AMYB_HORVU
c4	Ta.137.1.S1_at	-1.10	-1.23	Beta-amylase (Fragment) n=1 Tax=Triticum aestivum RepID=Q9ZR48_WHEAT
c4	Ta.4494.1.S1_x_at	-3.71	-8.39	Beta-amylase n=3 Tax=Oryza sativa RepID=Q10RZ1_ORYSJ
c4	Ta.4494.1.S1_at	-4.04	-8.61	Beta-amylase n=3 Tax=Oryza sativa RepID=Q10RZ1_ORYSJ
c4	Ta.1062.1.S1_at	1.10	-1.66	Beta-amylase n=1 Tax=Zea mays RepID=B6SVZ0_MAIZE
c4	TaAффx.68872.1.S1_at	-1.39	-1.61	Putative uncharacterized protein Sb01g028700 n=1 Tax=Sorghum bicolor RepID=C5WRG3_SORBI
c4	Ta.3.1.S1_at	-1.14	-1.26	Beta-amylase n=2 Tax=Triticeae RepID=AMYB_WHEAT
c4	Ta.27780.1.S1_x_at	-6.27	-5.52	Beta-amylase n=12 Tax=Hordeum vulgare RepID=AMYB_HORVU
c4	Ta.27780.1.S1_at	-7.97	-6.90	Beta-amylase n=12 Tax=Hordeum vulgare RepID=AMYB_HORVU
c5	Ta.3.1.S1_at	-1.14	-1.26	Beta-amylase n=2 Tax=Triticeae RepID=AMYB_WHEAT

c5	Ta.27780.1.S1_at	-7.97	-6.90	Beta-amylase n=12 Tax=Hordeum vulgare RepID=AMYB_HORVU
c5	Ta.27780.1.S1_x_at	-6.27	-5.52	Beta-amylase n=12 Tax=Hordeum vulgare RepID=AMYB_HORVU
c5	Ta.137.1.S1_at	-1.10	-1.23	Beta-amylase (Fragment) n=1 Tax=Triticum aestivum RepID=Q9ZR48_WHEAT
c5	Ta.4494.1.S1_x_at	-3.71	-8.39	Beta-amylase n=3 Tax=Oryza sativa RepID=Q10RZ1_ORYSJ
c5	Ta.4494.1.S1_at	-4.04	-8.61	Beta-amylase n=3 Tax=Oryza sativa RepID=Q10RZ1_ORYSJ
c5	Ta.1062.1.S1_at	1.10	-1.66	Beta-amylase n=1 Tax=Zea mays RepID=B6SVZ0_MAIZE
c5	TaAffx.68872.1.S1_at	-1.39	-1.61	Putative uncharacterized protein Sb01g028700 n=1 Tax=Sorghum bicolor RepID=C5WRG3_SORBI
c5	Ta.27780.2.A1_x_at	-10.60	-10.47	Beta-amylase (Fragment) n=1 Tax=Secale cereale RepID=AMYB_SECCE
c5	Ta.27780.2.A1_a_at	-10.48	-10.72	Beta-amylase n=12 Tax=Hordeum vulgare RepID=AMYB_HORVU
c6	Ta.23807.4.S1_x_at	1.56	1.52	70 kDa heat shock protein n=16 Tax=Embryophyta RepID=Q9LHA8_ARATH
c6	Ta.23807.4.S1_s_at	1.60	1.46	70 kDa heat shock protein n=16 Tax=Embryophyta RepID=Q9LHA8_ARATH
c6	Ta.24515.1.S1_at	-2.04	-3.33	70 kDa heat shock protein n=16 Tax=Embryophyta RepID=Q9LHA8_ARATH
c6	Ta.23807.7.S1_x_at	1.50	1.34	70 kDa heat shock protein n=16 Tax=Embryophyta RepID=Q9LHA8_ARATH
c6	Ta.23807.7.S1_at	1.47	1.21	70 kDa heat shock protein n=16 Tax=Embryophyta RepID=Q9LHA8_ARATH
c6	Ta.10259.1.S1_at	-1.53	-1.11	70 kDa heat shock protein n=16 Tax=Embryophyta RepID=Q9LHA8_ARATH
c6	Ta.23807.6.A1_x_at	1.42	1.52	70 kDa heat shock protein n=16 Tax=Embryophyta RepID=Q9LHA8_ARATH
c6	Ta.23807.6.A1_at	1.37	1.38	70 kDa heat shock protein n=16 Tax=Embryophyta RepID=Q9LHA8_ARATH
c6	Ta.21335.1.S1_a_at	-1.16	2.09	Luminal-binding protein 2 n=10 Tax=Magnoliophyta RepID=BIP2_MAIZE
c6	Ta.21335.1.S1_at	-1.20	2.83	Luminal-binding protein 2 n=10 Tax=Magnoliophyta RepID=BIP2_MAIZE
c6	Ta.28220.1.S1_at	1.52	1.12	Predicted protein n=4 Tax=core eudicotyledons RepID=B9N9W5_POPTR
c6	TaAffx.112076.1.S1_at	-1.09	-1.52	Heat shock protein 70 (Fragment) n=3 Tax=Magnoliophyta RepID=Q5EBY7_MAIZE
c6	Ta.15522.1.S1_at	1.04	1.01	Putative uncharacterized protein n=1 Tax=Oryza sativa Indica Group RepID=A2Y5F9_ORYSI
c6	Ta.21335.2.A1_x_at	-1.66	3.16	Luminal-binding protein 2 n=10 Tax=Magnoliophyta RepID=BIP2_MAIZE
c6	Ta.522.1.S1_at	-2.11	-1.49	Putative uncharacterized protein n=1 Tax=Oryza sativa Japonica Group RepID=B9FK56_ORYSJ
c6	Ta.2706.3.S1_at	1.46	1.76	Os01g0180800 protein n=1 Tax=Oryza sativa Japonica Group RepID=Q9AQZ5_ORYSJ
c6	Ta.15522.2.S1_x_at	-1.31	-1.51	Putative uncharacterized protein Sb06g014400 n=1 Tax=Sorghum bicolor RepID=C5YEW8_SORBI
c7	Ta.1548.2.S1_x_at	1.06	1.08	Putative elongation factor 1 beta n=1 Tax=Hordeum vulgare RepID=Q9M3U8_HORVU
c8	Ta.1548.2.S1_x_at	1.06	1.08	Putative elongation factor 1 beta n=1 Tax=Hordeum vulgare RepID=Q9M3U8_HORVU
c8	Ta.26753.1.A1_at	-1.23	-1.12	Elongation factor 1-delta 1 n=4 Tax=Andropogoneae RepID=B4FNT1_MAIZE
c9	Ta.1548.2.S1_x_at	1.06	1.08	Putative elongation factor 1 beta n=1 Tax=Hordeum vulgare RepID=Q9M3U8_HORVU
c10	no hit			
c11	Ta.1314.1.S1_s_at	-4.59	-4.83	Serpin-Z1B n=5 Tax=Triticum aestivum RepID=SPZ1B_WHEAT
c11	Ta.1314.1.S1_at	-4.61	-6.31	Serpin-Z1B n=5 Tax=Triticum aestivum RepID=SPZ1B_WHEAT
c11	Ta.117.1.S1_at	-8.65	-5.43	Serpin-Z2B n=1 Tax=Triticum aestivum RepID=SPZ2B_WHEAT
c11	Ta.118.1.S1_at	-4.43	-4.87	Serpin-Z1B n=5 Tax=Triticum aestivum RepID=SPZ1B_WHEAT
c11	Ta.9259.1.S1_at	-1.45	-1.15	Serpin-ZX n=1 Tax=Hordeum vulgare RepID=SPZX_HORVU
c11	Ta.9999.1.S1_at	-7.35	-5.71	Serpin 2 n=2 Tax=Triticum aestivum RepID=COLF31_WHEAT
c11	Ta.119.1.S1_x_at	-1.80	-1.55	Serpin-Z2A n=1 Tax=Triticum aestivum RepID=SPZ2A_WHEAT
c11	Ta.26273.1.A1_at	-2.13	-5.51	Serpin-ZX n=1 Tax=Hordeum vulgare RepID=SPZX_HORVU
c12	Ta.7031.3.S1_a_at	-2.68	-4.13	Os03g0332400 protein n=2 Tax=Oryza sativa RepID=Q10LW8_ORYSJ
c12	TaAffx.112982.1.S1_at	-1.11	-1.34	Hydroxyacylglutathione hydrolase n=3 Tax=Andropogoneae RepID=B6T2Y2_MAIZE

**Table S1.** Relative changes in expression of probesets representing proteins downregulated during imbibition in both dormant and after-ripened seeds. Conversion of the proteins into their corresponding probesets was performed as described previously<sup>3</sup>. Expression data of the probesets was obtained from our previously reported transcriptomic data (GEO accession number GSE32409) analysed as described before<sup>2</sup>. Probesets with  $\geq 2$ -fold change ( $P \leq 0.05$ ) were considered as differentially expressed.