

# **Responsive changes of DNA methylation in wheat (*Triticum aestivum*) under water deficit**

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## **Supplementary information**

Table S1 The sequence of adapters and primers in MSAP

Table S2 The cycling conditions in pre-amplification of MSAP

Table S3 The pattern of DNA methylation in wheat seedlings under water deficit

## Supplementary information

Table S1 The sequence of adapters and primers in MSAP

No.	Adapter and primer	Sequence
1	<i>EcoRI</i> adapter	5'-CTCGTAGACTGCGTACC CTGACGCATGGTTAA-5'
2	<i>MspI</i> and <i>HpaII</i> adapter	5'-GACGATGAGTCTAGAA CTACTCAGATCTTGC-5'
3	<i>MspI</i> and <i>HpaII</i> adapter	5'- GATCATGAGTCCTGCT GCTCGTCCTAGTACT-5'
4	<i>EcoRI</i> pre-amplification primer	5'-GACTGCCTACCAATTCA-3'
5	<i>MspI/HpaII</i> pre-amplification primer	5'-GATGAGTCTAGAACGGT-3'
6	Selective amplification primer	Eo: 5'-GACTGCCTACCAATTC MHo: 5'-GATGAGTCTAGAACGGT
7	<i>EcoRI</i> selective amplification primer	E1:Eo-AAC E2:Eo-AAG E3:Eo-ACA E4:Eo-ACT E5:Eo-AGG E6:Eo-ACC E7:Eo-ACG E8:Eo-AGC
8	<i>MspI/HpaII</i> selective amplification primer	MH4: MHo-ACA MH5: MHo-ACC MH6: MHo-CCG MH7: MHo-ACT MH8: MHo-TAA

Table S2 The cycling conditions in pre-amplification of MSAP

Cycle name	Temperature (°C)	Time	Number of cycles
Pre-PCR_1	72.0	1m	1
Pre-PCR_2	94.0	45s	1
	65.0	30s	
	72.0	1m	
Touch-down PCR	94.0	30s	12
	64.3	30s	
	72.0	1m	
PCR	94.0	30s	20
	55.9	30s	
	72.0	1m	
Elongation	72.0	30s	1
Store	4.0	-	-

The annealing temperature of Touch-down PCR was respectively 64.3°C, 63.6°C, 62.9°C, 62.2°C, 61.5°C, 60.8°C, 60.1°C, 59.4°C, 58.7°C, 58.0°C, 57.3°C and 56.6°C, respectively.

Table S3 The pattern of DNA methylation in wheat seedlings under water deficit

No.	Band				Change of methylation pattern		Type of methylation pattern	
	H1	M1	Hx	Mx	CK	Treatment		
1	1	1	1	1	CCGG GGCC	CCGG GGCC	A1	
2	1	0	1	0	CCGG CCGG GGCC GGCC	CCGG CCGG GGCC GGCC	A2	A
3	0	1	0	1	CCGG GGCC	CCGG GGCC	A3	
4	1	1	1	0	CCGG GGCC	CCGG CCGG GGCC GGCC	B1	
5	1	1	0	1	CCGG GGCC	CCGG GGCC	B2	
6	0	1	0	0	CCGG GGCC	CCGG GGCC	B3	B
7	1	0	0	0	CCGG CCGG GGCC GGCC	CCGG GGCC	B4	
8	1	1	0	0	CCGG GGCC	CCGG GGCC	B5	
9	0	1	1	1	CCGG GGCC	CCGG GGCC	C1	
10	1	0	1	1	CCGG CCGG GGCC GGCC	CCGG GGCC	C2	C
11	0	0	0	1	CCGG GGCC	CCGG GGCC	C3	
12	0	0	1	1	CCGG GGCC	CCGG GGCC	C4	

H and M represented *EcoRI/HpaII* (H) and *EcoRI/MspI* (M), H1 and M1: bands amplified from genome of wheat seedlings under the normal condition, Hx and Mx: bands amplified from genome of wheat seedling treated with PEG<sub>6000</sub> solution. Scores of 1 and 0 indicated the presence or absence of bands in MSAP, “1 1”, “1 0” and “0 1” was considered to be the pattern of non-methylation, hemi-methylation or full-methylation, respectively.