Electronic supplementary information

A first glance on the micro-ZnO coating in maize (*Zea mays* L.) seeds: study of the elemental spatial distribution and Zn speciation analysis

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Daily monitoring of relative humidity and temperature during seed storage for 12 months

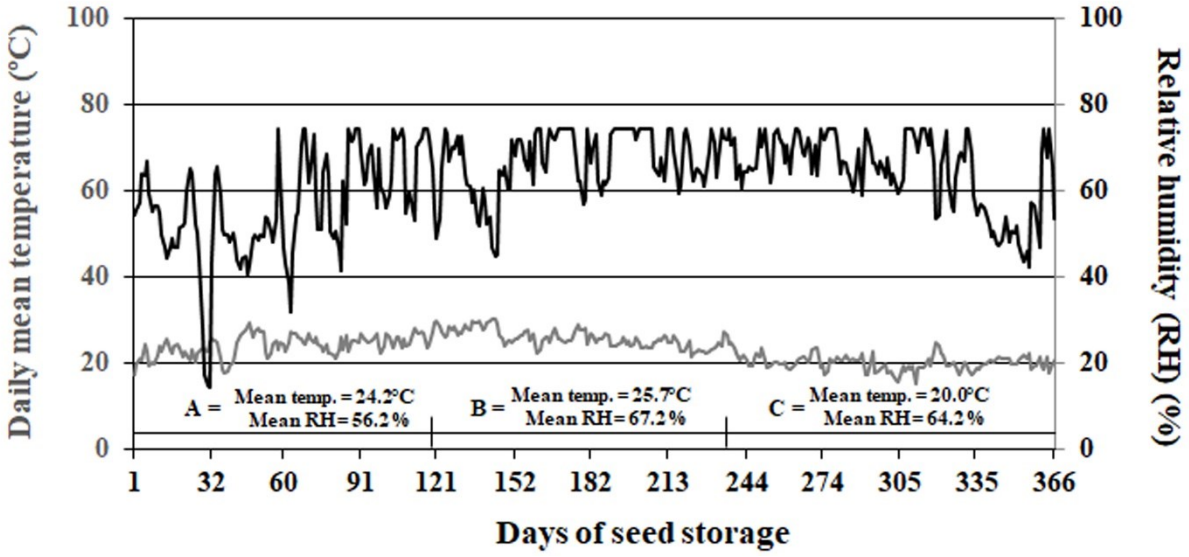

Fig. S1 Daily mean values of relative humidity and temperature during storage (in dark condition) for 12 months of ZnO-treated and untreated seeds. Values plotted in A, B, and C correspond to mean temperature and relative humidity in the period of 4, 8, and 12 months, respectively.

Table S1 Zn removal of untreated (control) and ZnO-treated seeds according to the number of washings (extractions)

	Untreated seeds			ZnO-treated seeds		
Washing cycle	Zn (µg)[1]	SD	Zn removed (%)[2]	Zn (µg)[1]	SD	Zn removed (%)[2]
1st	0.29	0.27	0.032	928	206	91.19
2nd	0.23	0.20	0.026	68	26	6.48
3rd	0,28	0,13	0.030	13	5	1.25
4th	0.41	0.55	0.034	4.0	1.6	0.43
5th	0.27	0.14	0.028	2.2	0.4	0.22
6th	0.31	0.17	0.029	2.4	0.7	0.25
7th	0.28	0.05	0.028	1.7	0.5	0.18
∑ Zn removed[2]	2.07			1019		

[1]Mean values and standard deviation correspond to 3 replicates.

^{2} Zn removed (%) = $\left(\frac{mass\ of\ Zn\ extracted\ in\ the\ washing\ cycle}{\sum mass\ of\ Zn\ removed\ in\ all\ cycles}\right)$ x 100

Calculation of limits of detection (LOD)

Limits of detection (LOD) were calculated according to the general expression for XRF analysis.[1] The following table summarizes the results:

Table S2 Limit of detection for XRF analysis.

Element	LOD (mg kg^{-1})
P	145
S	40
Zn	0.25

Zn speciation analysis

The excitation energy was selected using a Si(111) channel-cut crystal monochromator and calibrated by defining the first derivate peak of a Zn foil spectrum to be 9659.0 eV (Zn K-edge). Energy calibration was done in transmission mode. Spectra from samples and standards were recorded in fluorescence mode. The excitation energy was tuned across the Zn K-edge from 9579 to 9861 eV. Spectra were recorded from 9579 to 9639 eV using step sizes of 2 eV for the pre-edge region. 0.5 eV for the edge and 2 and 3 eV for the post-edge (9761 – 9861 eV). The count times and the number of scans were adjusted according to the counting statistics for each sample and standard. Given this conditions, the total number of point per scan was 314. The table 3 summarizes the main experimental conditions for XANES measurements:

Table S3 Experimental conditions for XANES measurements

Energy range	Energy step (eV)	Measuring time /point (s)		Number of points
		(Standards)	(Samples)	
9579 – 9639 eV	2	1	3	30
9639 – 9759 eV	0.5	2	6	241
9761 – 9829 eV	2	1	3	31
9831 – 9861 eV	3	1	3	12

References

1 R. E. Van Grieken and A. A. Markowicz. Eds.. *Handbook of X-Ray Spectrometry: methods and techniques*. Marcel Dekker. Inc. New York. v. 14. 1993.