

## Electronic Supplementary Material (ESM)

to XUE et al.: Characterizing Barley Seed Macro- and Micro-nutrients under Multiple Environmental Conditions

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*Table S1.* Average temperature, pH, organic matter content and element concentrations in the soil of three field locations in China

|                                      | Site 1-Y2011 | Site 1-Y2012 | Site 2-Y2013 |
|--------------------------------------|--------------|--------------|--------------|
| T <sub>april</sub> (°C) <sup>a</sup> | 11.7–18.6    | 13.5–22.6    | 12.0–19.9    |
| T <sub>may</sub> (°C) <sup>a</sup>   | 14.7–24.1    | 16.3–23.4    | 15.7–23.4    |
| pH                                   | 6.45         | 6.38         | 6.68         |
| Organic matter (%)                   | 6.49         | 5.37         | 4.53         |
| N (%)                                | 0.205        | 0.17         | 0.218        |
| P (%)                                | 0.12         | 0.12         | 0.13         |
| K (%)                                | 1.84         | 2.24         | 1.6          |
| Ca (%)                               | 0.43         | 0.51         | 0.32         |
| Mg (%)                               | 1.05         | 1.97         | 0.58         |
| Fe (%)                               | 6.78         | 8.13         | 8.86         |
| Cu (mg/kg)                           | 60.1         | 63.8         | 84.4         |
| Zn (mg/kg)                           | 134          | 145          | 191          |
| Mn (mg/kg)                           | 398          | 468          | 2370         |
| Se (mg/kg)                           | 2.06         | 1.46         | 0.64         |

<sup>a</sup>The values refer to average minimum and maximum temperature for whole month, respectively.

*Table S2.* Two-way ANOVA of seed components from 4 environments

|                | Environments    |                | Genotypes       |                | E × G           |                | Error           | Total           |
|----------------|-----------------|----------------|-----------------|----------------|-----------------|----------------|-----------------|-----------------|
|                | SS <sup>a</sup> | % <sup>b</sup> | SS <sup>a</sup> | % <sup>b</sup> | SS <sup>a</sup> | % <sup>b</sup> | SS <sup>a</sup> | SS <sup>a</sup> |
| DF             | 3               |                | 61              |                | 183             |                | 496             |                 |
| Sta            | 0.541**         | 47             | 0.061           | 5              | 0.148           | 13             | 0.408           | 1.158           |
| TN             | 2.847**         | 50             | 1.188**         | 21             | 0.645**         | 11             | 0.989           | 5.669           |
| TSP            | 1.569**         | 20             | 0.780           | 10             | 1.774           | 23             | 3.561           | 7.684           |
| Phy            | 1.968**         | 17             | 1.329           | 12             | 5.885**         | 51             | 2.262           | 11.444          |
| Phe            | 3.145**         | 24             | 2.308           | 18             | 5.472**         | 43             | 1.947           | 12.872          |
| Fla            | 4.860**         | 34             | 2.729*          | 19             | 4.710**         | 33             | 1.970           | 14.269          |
| P <sub>i</sub> | 3.396**         | 17             | 3.725           | 19             | 8.527**         | 43             | 4.404           | 20.052          |
| Zn             | 9.328**         | 47             | 1.967*          | 10             | 3.412**         | 17             | 4.992           | 19.699          |
| Fe             | 2.465**         | 27             | 1.573**         | 18             | 2.265**         | 25             | 2.673           | 8.976           |
| SKW            | 0.782**         | 21             | 0.653**         | 18             | 0.695           | 19             | 1.524           | 3.654           |
| HKW            | 0.685**         | 23             | 0.761**         | 26             | 0.488*          | 16             | 1.042           | 2.976           |
| CNR1           | 5.750**         | 60             | 1.419**         | 15             | 0.929**         | 10             | 1.560           | 9.658           |
| CNR2           | 3.369**         | 31             | 0.894           | 8              | 2.263*          | 21             | 4.213           | 10.739          |

<sup>a</sup>Significance of  $p < 0.05$  and  $p < 0.001$  marked with “\*” and “\*\*\*”, respectively.

<sup>b</sup>Percentage of SS (sum of squares) were calculated by dividing total SS.

Table S3. Nested ANOVA of seed components from 4 environments, genotype nested within haplotype *Hvm74*

|                | Environments    |                | Genotype nested <i>Hvm74</i> |                | <i>Hvm74</i>    |                | Error           | Total           |
|----------------|-----------------|----------------|------------------------------|----------------|-----------------|----------------|-----------------|-----------------|
|                | SS <sup>a</sup> | % <sup>b</sup> | SS <sup>a</sup>              | % <sup>b</sup> | SS <sup>a</sup> | % <sup>b</sup> | SS <sup>a</sup> | SS <sup>a</sup> |
| DF             | 3               |                | 60                           |                | 1               |                | 679             |                 |
| Sta            | 0.544**         | 47             | 0.046                        | 4              | 0.015**         | 1              | 0.557           | 1.162           |
| TN             | 2.868**         | 50             | 0.818**                      | 14             | 0.394**         | 7              | 1.634           | 5.714           |
| TSP            | 1.617**         | 21             | 0.804*                       | 10             | 0.001           | 0              | 5.335           | 7.757           |
| Phy            | 1.952**         | 17             | 1.241*                       | 11             | 0.116*          | 1              | 8.147           | 11.456          |
| Phe            | 3.252**         | 25             | 2.396**                      | 19             | 0.027           | <1             | 7.149           | 12.824          |
| Fla            | 4.986**         | 35             | 2.670**                      | 19             | 0.061*          | <1             | 6.681           | 14.398          |
| P <sub>i</sub> | 3.303**         | 16             | 3.789**                      | 19             | 0.022           | <1             | 12.931          | 20.045          |
| Zn             | 9.338**         | 47             | 1.935**                      | 10             | 0.028           | <1             | 8.404           | 19.705          |
| Fe             | 2.464**         | 27             | 1.298**                      | 14             | 0.272**         | 3              | 4.938           | 8.972           |
| SKW            | 0.793**         | 22             | 0.651**                      | 18             | 0.000           | 0              | 2.219           | 3.663           |
| HKW            | 0.695**         | 23             | 0.765                        | 26             | 0.001           | 0              | 1.530           | 2.991           |
| CNR1           | 5.867**         | 56             | 0.935**                      | 11             | 0.544**         | 6              | 2.489           | 9.835           |
| CNR2           | 3.556**         | 32             | 0.954*                       | 9              | 0.015           | <1             | 6.476           | 11.001          |

<sup>a</sup>Significance of  $p < 0.05$  and  $p < 0.001$  marked with “\*” and “\*\*\*”, respectively.

<sup>b</sup>Percentage of SS (sum of squares) were calculated by dividing total SS.

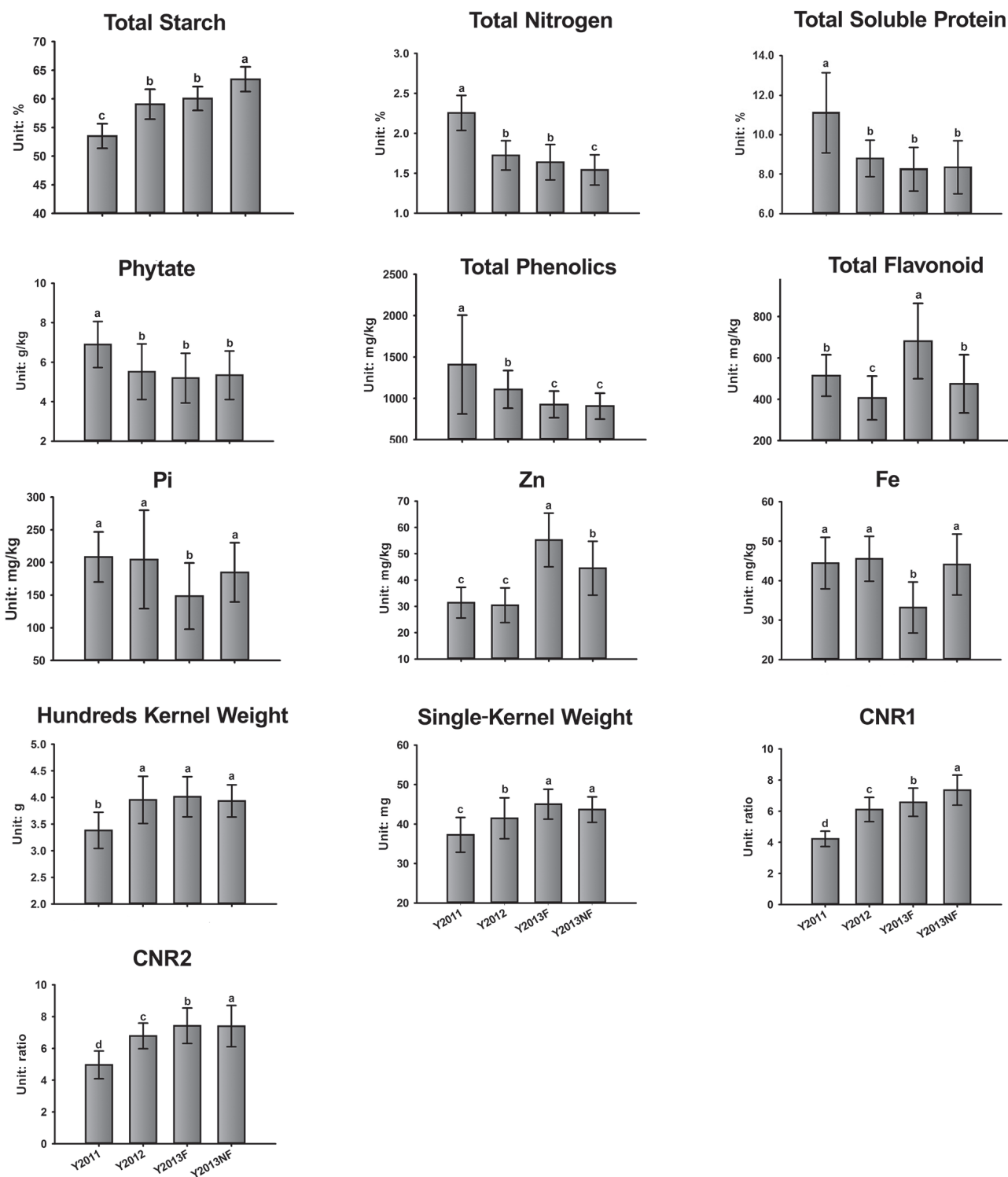


Figure S1. Comparative average of barley seed components, carbon/nitrogen ratio, HKW and SKW in 4 environments

For CNR1 and CNR2, Sta regarded as carbon (C), TN (C/N ratio 1) and TSP (C/N ratio 2) taken as nitrogen (N), respectively. The bars within graphs represent the mean  $\pm$  S.D. Different alphabets refer to significant differences with  $p < 0.05$  (Tukey HSD). Sta = starch, TN = total nitrogen, TSP = total soluble protein, Phy = phytate, Phe = total phenolics, Fla = total flavonoids,  $P_i$  = inorganic phosphorus, Zn = zinc, Fe = iron, HKW = hundreds kernel weight, SKW = single-kernel weight, CNR1 = starch/total nitrogen ratio, CNR2 = starch/total soluble protein