

Supplementary material

Micronutrient nanoprotectants curtail arsenic-induced physio-oxidative damages by differentially regulating antioxidant and metabolic mechanisms across *Brassica napus* genotypes

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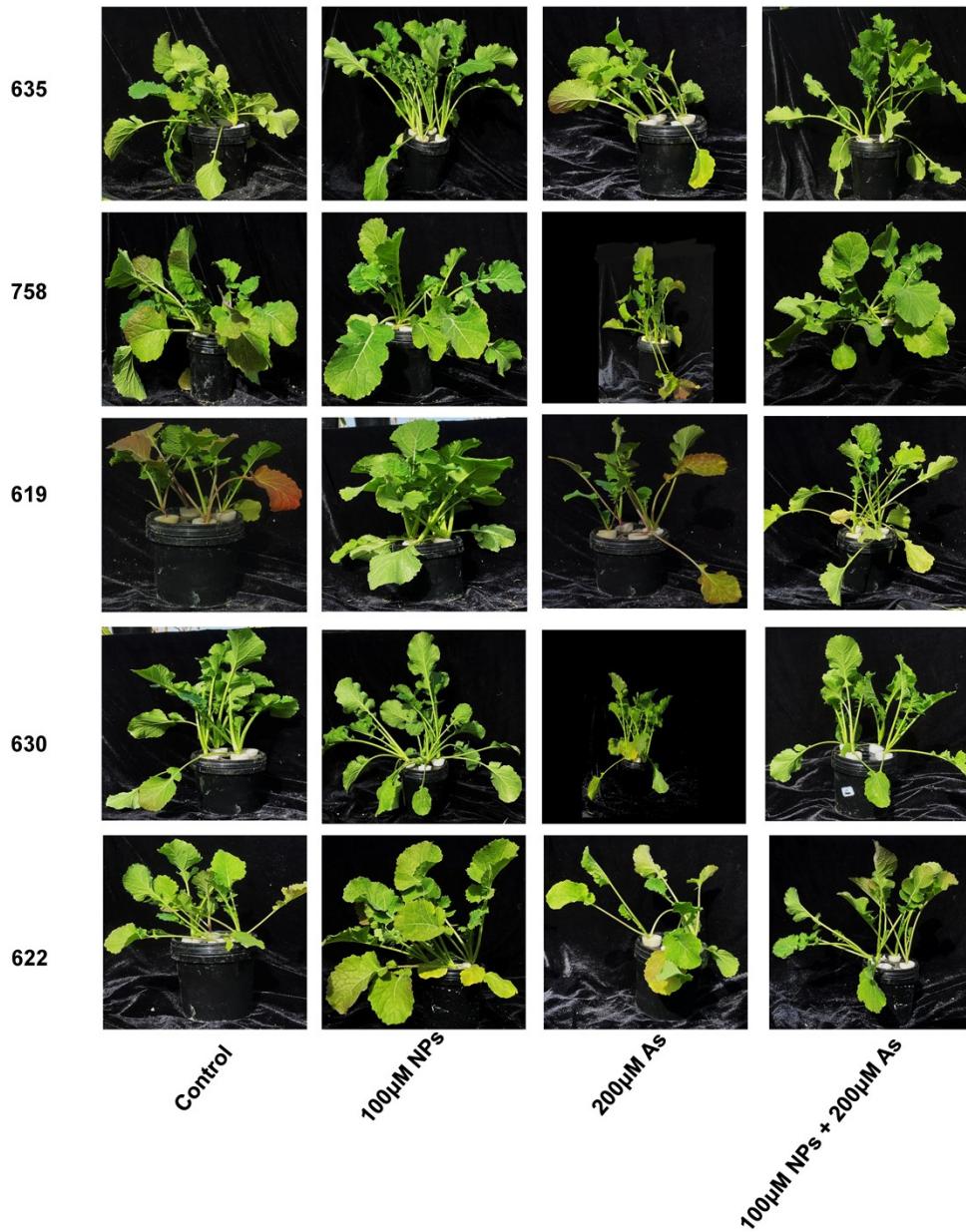


Fig. S1. Visual representation of the effects of different treatments on the growth of *Brassica napus* cultivars (635, 758, 619, 630, and 622). The images show the plant responses to four treatment conditions: Control, 100µM MnNPs, 200µM As, and 100µM MnNPs + 200µM As. The photographs illustrate the varying degrees of As-induced toxicity and the protective effects of MnNPs across different cultivars, highlighting the genotypic differences in stress tolerance.

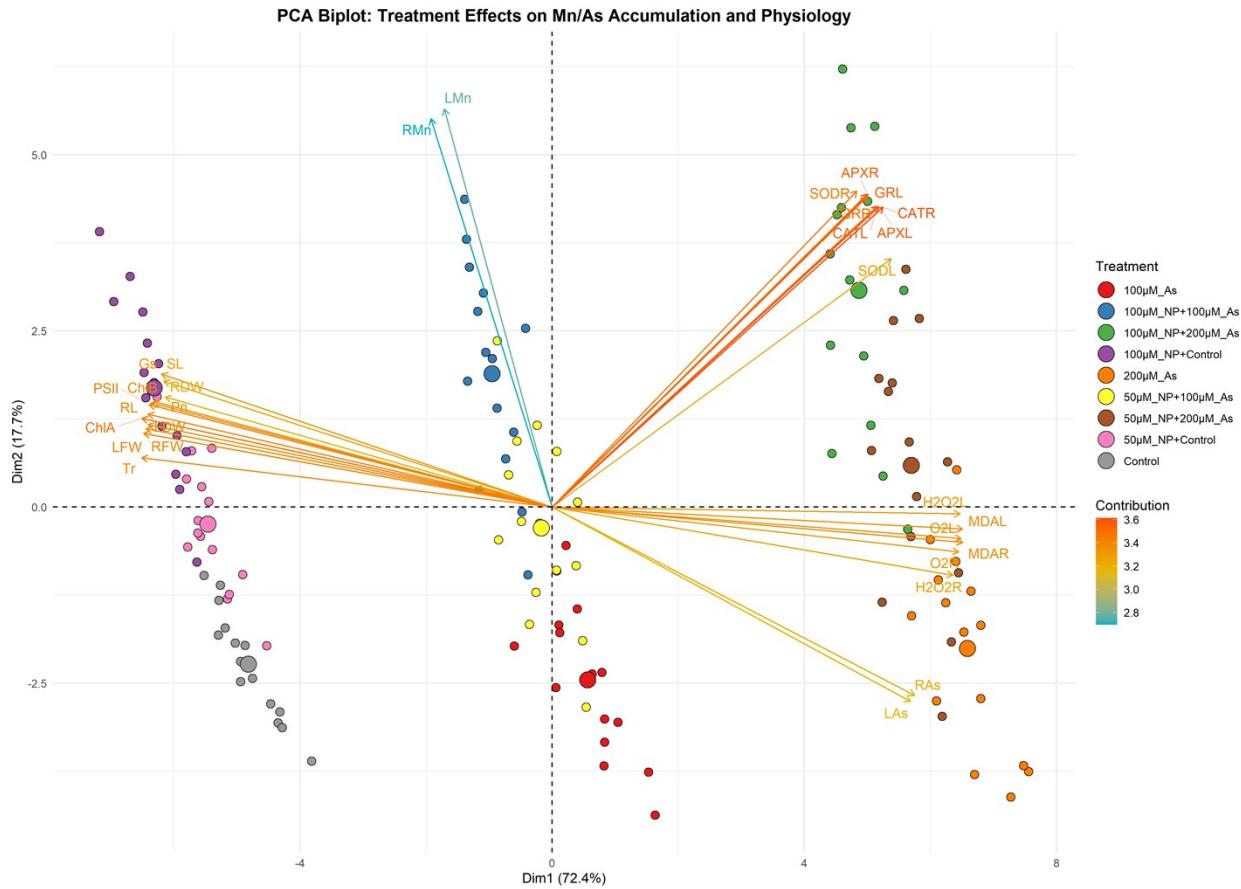


Fig. S2. PCA Biplot showing treatment effects on Mn/As accumulation and physiological responses. The biplot displays the relationship between Mn/As accumulation and various physiological and biochemical traits, such as growth, photosynthesis, and antioxidant enzyme activity. Treatments are color-coded as follows: 100µM As (red), 100µM NP + 100µM As (green), 100µM NP + 200µM As (yellow), 200µM As (blue), 50µM NP + 200µM As (purple), 50µM NP + 100µM As (cyan), 50µM NP + Control (pink), and Control (grey). The arrows represent the contribution of various traits (e.g., SOD, GRL, APX) to the principal components (PC1 and PC2).

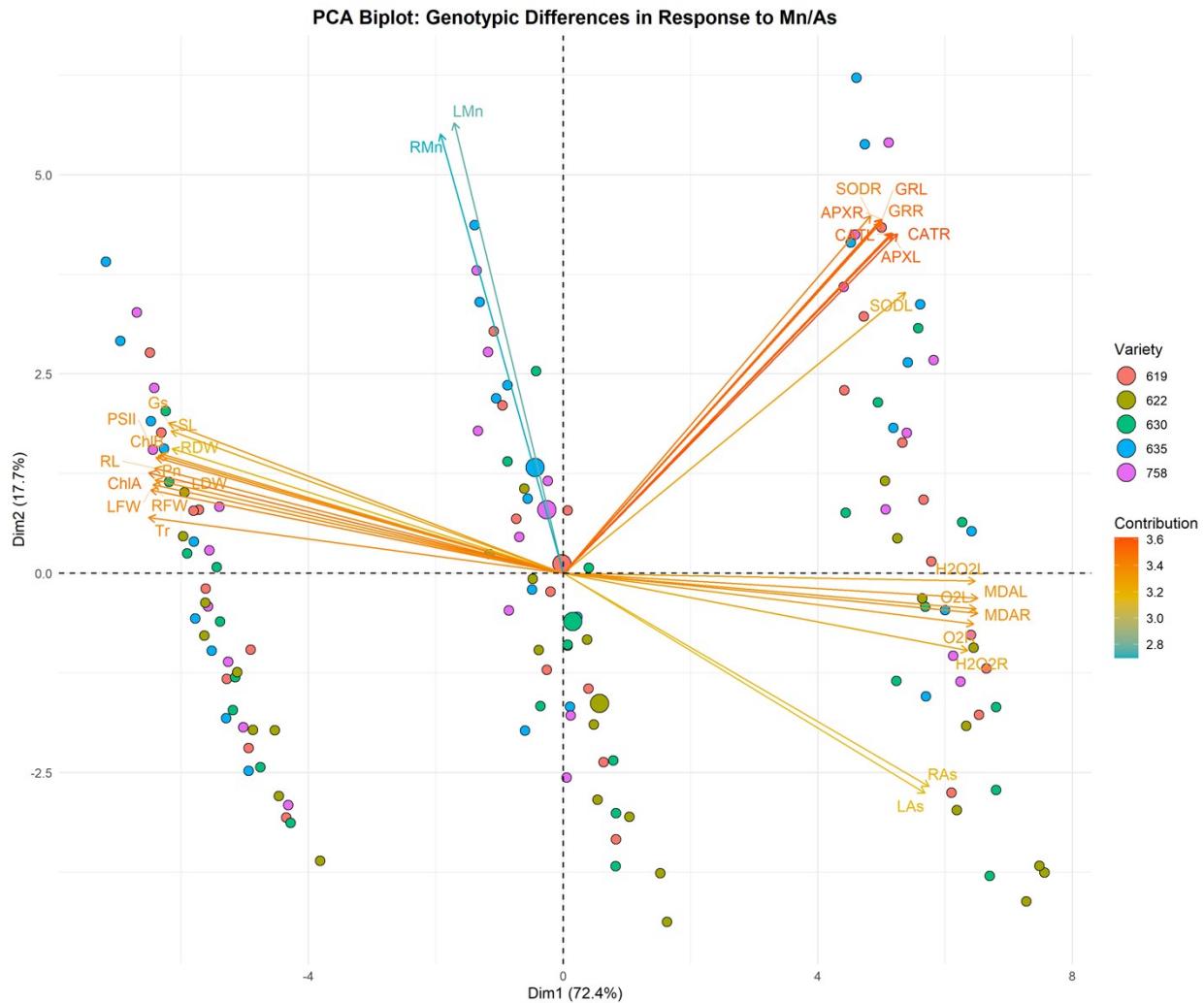


Fig. S3. PCA Biplot showing genotypic differences in response to Mn/As accumulation and physiological traits. The plot illustrates how five *Brassica napus* cultivars (619, 622, 630, 635, and 758) respond to varying treatments, with each cultivar represented by differently colored points. The arrows indicate the contribution of physiological and biochemical traits (e.g., growth, photosynthesis, antioxidant enzyme activity) to the principal components (PC1 and PC2). The percentage of variance explained by each component is indicated, with PC1 explaining 72.4% and PC2 explaining 17.7% of the variance. The color gradient represents the contribution of each trait to the principal components, with darker shades indicating higher contributions.

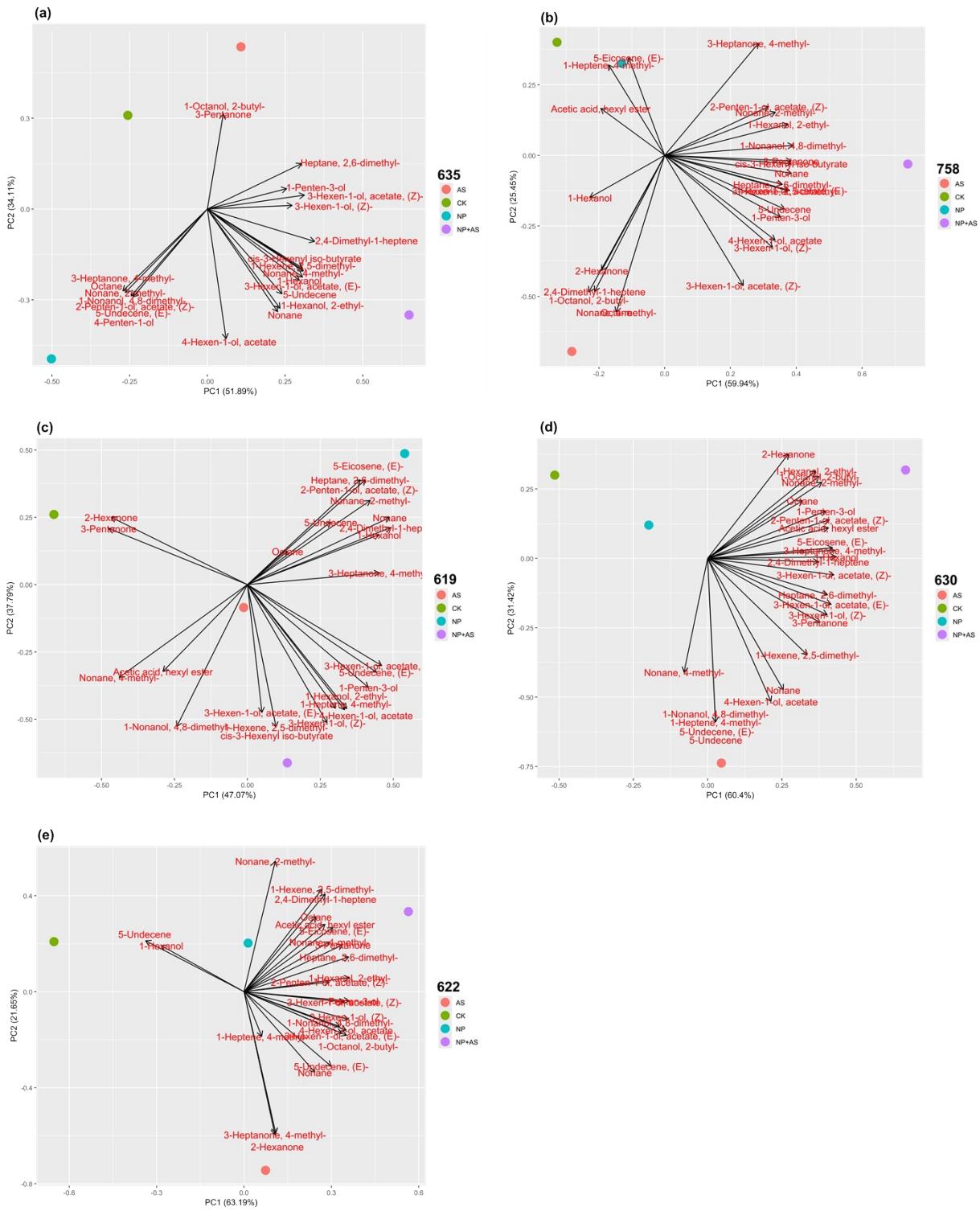


Fig. S4. PCA analysis of treatment effects within individual varieties. PCA plots for each of the five varieties, showing metabolic variations induced by treatments. The plots illustrate varietal-specific responses, with some varieties exhibiting distinct treatment clustering and others showing minimal separation.

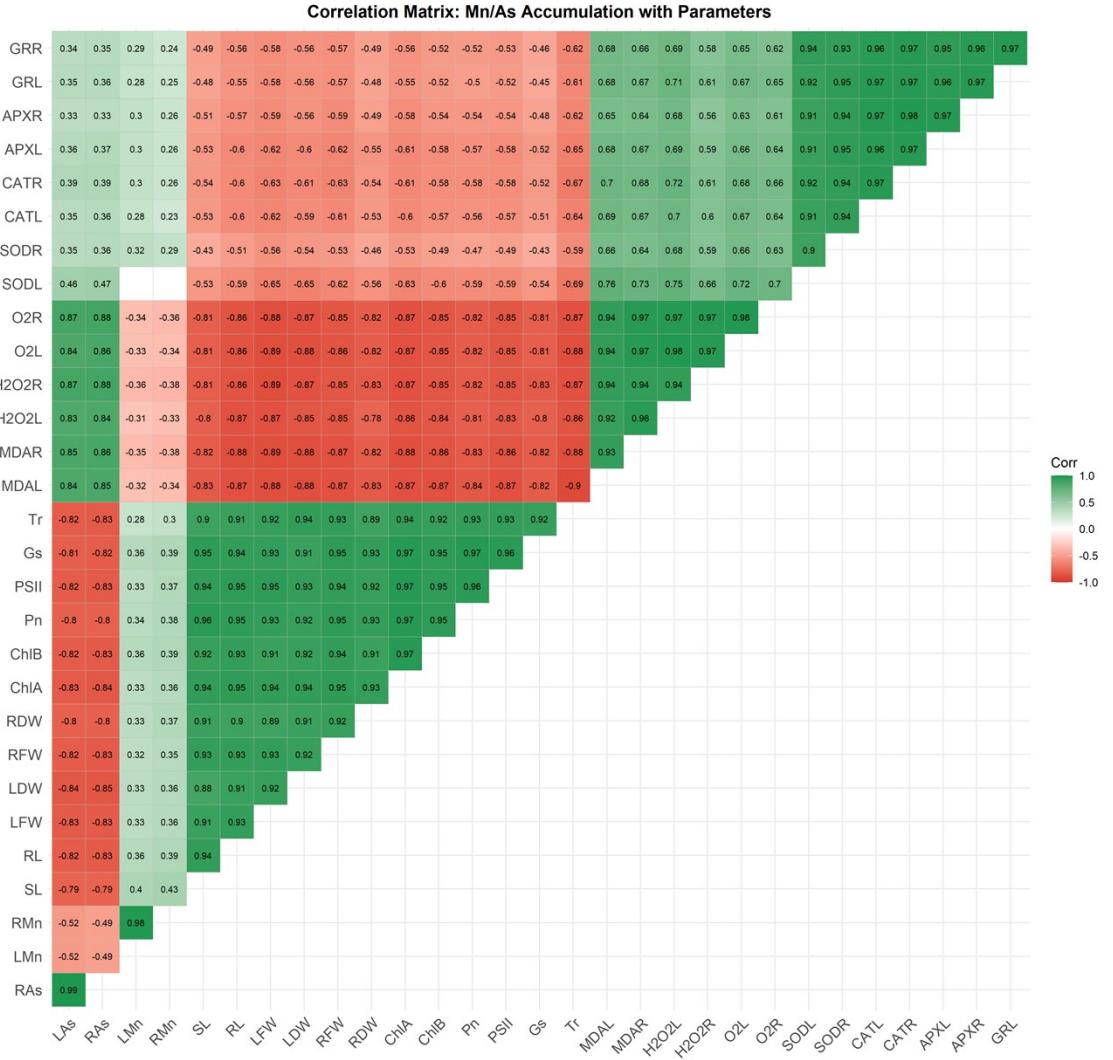


Fig. S5. Correlation matrix showing the relationship between Mn/As accumulation and various physiological and biochemical parameters in *Brassica napus*. The matrix presents Pearson correlation coefficients (r) between Mn (RMn, LMn), As (RAs, LAs) accumulation, and various plant traits, including growth parameters, photosynthesis, antioxidant enzymes, and oxidative stress markers. Positive correlations are indicated by shades of green, while negative correlations are shown in red. High positive correlations ($r > 0.9$) are observed between Mn/As accumulation and physiological traits like growth (RDW, RFW, SL) and photosynthetic efficiency (Pn, PSII), while antioxidant markers such as SOD, CAT, and GR show varying degrees of correlation with both Mn and As accumulation.

Table S1. Physicochemical characteristics of MnNPs used in this study.

Parameter	Description / Value	Analytical Method
Particle size (diameter)	~33 nm (average)	SEM, TEM
Particle morphology	Predominantly spherical	SEM, TEM
Aggregation state	Low agglomeration; well-dispersed	SEM
Elemental composition	Mn: 75.02%, O: 24.98%	EDX

Table S2. Oligonucleotide primer sequences used for qRT-PCR analysis

Gene name	Forward	Reverse
Super oxide dismutase(SOD)	ACGGTGTGACCACTGTGACT	GCACCGTGTGTTACCATC
Catalase (CAT)	TCGCCATGCTGAGAAGTATC	TCTCCAGGCTCCTGAAGTT
Ascorbate peroxidases(APX)	ATGAGGTTGACGGTGAGC	CAGCATGGGAGATGGTAGG
Glutathione reductase (GR)	AAGCTGGAGCTGTGAAGGTT	AGACAGTGGTCGCAAAGCAG
<i>Actin</i>	TTGGGATGGACCAGAAGG	TCAGGAGCAATACGGAGC