

Supplemental Information

Table S1. Analytical methods used to determine selected characteristics of the three reference soils.

| Property | Detail | Reference |
|----------------------------|---|-----------|
| pH | 1:5 (w/v) soil suspension using 0.01 M CaCl ₂ . | [1] |
| CEC | Cobalt hexamine method, using 0.0166 M cobalt(III) hexamine chloride ([Co(NH ₃) ₆]Cl ₃) as extracting solution at a 1:20 soil:liquid ratio (w/v). | [2] |
| Fe-ox | Oxalic acid-oxalate extracts of 1:50 (w/v) extraction systems, at buffered pH ≈ 3. | [3] |
| TC | Total carbon content of the soils was determined by dry combustion using a CN analyzer (VarioMax). | [4] |
| Inorganic carbon | CO ₂ preasure after the addition of a HCl:FeSO ₄ mixture to the soils sample in a sealed container | [5] |
| Total metal concentrations | Hot-acid digestion of 50 mg (dry mass) of each sample using a mixture of 1:3 HNO ₃ to HCl concentrated acids | [1] |

Table S2. MixTox–model parameters corresponding to different deviation patterns. Associated deviation function and practical meaning of their values are included. Adapted from Jonker et al., 2005.

* $x=1$ for CA; $x=2$ for IA.

| Deviations | Parameter | CA G function | IA G function | Value | Interpretation |
|-------------------------|-----------|---------------------------------|------------------------------------|---------|--|
| Synergism or Antagonism | a^* | $G = e^{az_iz_j}$ | $G = az_iz_j$ | >0 | Antagonism |
| | | | | <0 | Synergism |
| Dose Ratio related (DR) | b_i | $G_i = e^{(a+bz_i)z_iz_j}$ | $G_i = (a + bz_i)z_iz_j$ | >0 | Antagonism with effects mainly caused by i |
| | | | | <0 | Synergism with effects mainly caused by i |
| Dose Level related (DL) | b_j | $G_i = e^{(a+bz_j)z_iz_j}$ | $G_j = (a + bz_j)z_iz_j$ | >0 | Antagonism with effects mainly caused by j |
| | | | | <0 | Synergism with effects mainly caused by j |
| | a | $G = e^{az_iz_j}$ | $G = az_iz_j$ | >0 | Low dose level: Antagonism |
| | | | | <0 | Low dose level: Synergism |
| | b^* | $G = e^{a(1-b(q_i+q_j))z_iz_j}$ | $G = a(1 - b(1 - q_i q_j)) z_iz_j$ | >x* | Inflexion point < EC50 |
| | | | | =x | Inflexion point = EC50 |
| | | | | x>b>x-1 | Inflexion point > EC50 |
| | | | | <x-1 | Magnitude of deviation is dose or effect level dependent |

* z_i : relative toxic unit of component i (Equation (6)); * q_i : individual toxic effect of component i in the mixture (Equation (7)).

Table S3. Zinc and arsenic averaged total concentrations (mg kg^{-1}) for all treatments and all soils. Standard errors (SE) are given between brackets. Bold values correspond to the variable component and treatment standard error (SE, mg kg^{-1}) are between brackets. Dose levels are those of Figure 1.

| | | Single Zn | | Single As | | Fixed Zn + Increasing As | | Fixed As + Increasing Zn | | Ray | |
|------|------------|---------------------|---------------------|---------------------|---------------------|--------------------------|---------------------|--------------------------|---------------------|---------------------|---------------------|
| Soil | Dose Level | Zn | As | Zn | As | Zn | As | Zn | As | Zn | As |
| | | mg kg^{-1} | mg kg^{-1} | mg kg^{-1} | mg kg^{-1} | mg kg^{-1} | mg kg^{-1} |
| S1 | 0 | 1400 (6) | 240 (23) | 1400(6) | 240 (23) | 1600 (31) | 300 (5) | 1400 (40) | 530 (15) | 1400 (6) | 240 (23) |
| | 1 | 1400 (37) | 170 (25) | 1400 (13) | 380 (14) | 1600 (29) | 350 (21) | 1300 (29) | 420 (47) | 1400 (7) | 370 (2) |
| | 2 | 1500 (28) | 300 (28) | 1300 (35) | 400 (17) | 1600 (50) | 400 (18) | 1400 (65) | 360 (21) | 1400 (18) | 420 (2) |
| | 3 | 1600 (31) | 300 (5) | 1400 (40) | 530 (15) | 1800 (158) | 430 (23) | 1800 (158) | 430 (29) | 1800 (160) | 430 (23) |
| | 4 | 2100 (28) | 250 (35) | 1500 (57) | 604 (4) | 1600 (21) | 620 (68) | 2200 (83) | 400 (29) | 2600 (29) | 670 (150) |
| | 5 | 3700 (3) | 300 (51) | 1400 (30) | 850 (41) | 1700 (14) | 1000 (13) | 3600(25) | 400 (54) | 4400 (129) | 1500 (240) |
| S2 | 6 | 8700 (130) | 300 (19) | 1400 (61) | 1300(200) | 1700 (120) | 1700 (10) | 8500 (76) | 430 (200) | 9700 (0.5) | 3300 (270) |
| | 0 | 87(3) | 22 (2) | 87 (3) | 22 (2) | 360 (22) | 39 (4) | 87 (2) | 80 (5) | 87 (3) | 22 (2) |
| | 1 | 130 (10) | 45 (8) | 87 (2) | 52 (0.5) | 290 (5) | 50 (1) | 110 (0.1) | 80 (3) | 150 (9) | 100 (3) |
| | 2 | 170 (2) | 48 (5) | 85 (1) | 55 (0.4) | 280 (2) | 64 (6) | 170 (6) | 47 (21) | 230 (16) | 200 (13) |
| | 3 | 360 (22) | 39 (12) | 87 (2) | 80 (5) | n.d. | n.d. | n.d. | n.d. | 500 (12) | 510 (30) |
| | 4 | 670 (14) | 60 (7) | 96 (12) | 150 (8) | 270 (3) | 100 (4) | 1000 (122) | 51 (16) | 1300 (111) | 1500 (120) |
| S3 | 5 | 2100 (99) | 40 (4) | 95 (1) | 250 (6) | 360 (15) | 120 (1) | 1700 (31) | 46 (4) | 3800 (3) | 4600 (75) |
| | 6 | 6800 (39) | 59 (3) | 98 (3) | 470 (3) | 320 (27) | 370 (47) | 6200 (95) | 52 (10) | 9600 (44) | 13000 (400) |
| | 0 | 350 (8) | 72 (2) | 350 (8) | 72 (2) | 700 (46) | 73 (3) | 320 (2) | 130 (0.1) | 350 (8) | 72 (2) |
| | 1 | 420 (25) | 97 (22) | 300 (3) | 81 (21) | 630 (23) | 61 (2) | 380 (5) | 130 (7) | 390 (1) | 130 (3) |
| | 2 | 450 (26) | 83 (15) | 300 (21) | 95 (6) | 710 (38) | 82 (10) | 420 (3) | 140 (1) | 450 (48) | 130 (6) |
| | 3 | 700 (46) | 73 (3) | 320 (2) | 130 (0.1) | n.d. | n.d. | n.d. | n.d. | 730 (21) | 360 (27) |
| S4 | 4 | 1300 (2) | 86 (4) | 280 (9) | 180 (7) | 670 (31) | 98 (2) | 1200 (30) | 120 (3) | 1500 (25) | 810 (5) |
| | 5 | 3400 (7) | 81 (5) | 340 (18) | 470 (35) | 610 (19) | 200 (2) | 3100 (85) | 140 (1) | 3500 (70) | 2200 (80) |
| | 6 | 8300 (730) | 130 (53) | 320 (13) | 1100 (25) | 640 (31) | 1100 (25) | 7900 (44) | 130 (3) | 9800 (82) | 7400 (170) |

Table S4. Total Zn and As measured concentrations (mg kg^{-1}) and corresponding Net Root Elongation (NRE, cm) response for all treatments. Reported NRE values are treatment averaged values with associated standard deviations (cm) between brackets.

| SOIL | Single Zn | | Single As | | Fixed Zn + Increasing As | | | Zn + Fixed As | | | Ray | | |
|------|-------------------------------|-------------|-------------------------------|-------------|-------------------------------|-------------------------------|-------------|-------------------------------|-------------------------------|-------------|-------------------------------|-------------------------------|-------------|
| | Zn (mg kg^{-1}) | NRE (cm) | As (mg kg^{-1}) | NRE (cm) | Zn (mg kg^{-1}) | As (mg kg^{-1}) | NRE (cm) | Zn (mg kg^{-1}) | As (mg kg^{-1}) | NRE (cm) | Zn (mg kg^{-1}) | As (mg kg^{-1}) | NRE (cm) |
| S1 | 1400 | 9.6 (1) | 240 | 9.6 (1) | 1600 | 300 | 8.6 (1) | 1400 | 530 | 9.1 (1) | 1400 | 240 | 9.6 (1) |
| | 1400 | 10 (1) | 380 | 9.3 (1) | 1600 | 350 | 9.1 (1) | 1300 | 420 | 9.9 (1) | 1400 | 370 | 9.1 (1) |
| | 1500 | 9.6 (1) | 400 | 9.1 (1) | 1600 | 400 | 9.4 (1) | 1400 | 360 | 9.4 (1) | 1400 | 420 | 9.0 (1) |
| | 1600 | 8.6 (1) | 530 | 9.1 (1) | | | | | | | 1800 | 430 | 8.1 (2) |
| | 2100 | 7.6 (1) | 604 | 5.4 (2) | 1600 | 620 | 7.2 (1) | 2200 | 400 | 9.6 (1) | 2600 | 670 | 1.9 (2) |
| | 3700 | 5.7 (2) | 850 | 1.4 (0.2) | 1700 | 1000 | 1.8 (1) | 3600 | 400 | 5.8 (1) | 4400 | 1500 | 1.2 (1) |
| | 8700 | 1.1 (0.2) | 1300 | 1.0 (0.1) | 1700 | 1700 | 0.9 (0.1) | 8500 | 430 | 0.9 (0.1) | 9700 | 3300 | 0.7 (0.1) |
| S2 | 87 | 11 (1) | 22 | 11 (1) | 360 | 39 | 10 (0.4) | 87 | 80 | 8.6 (1) | 87 | 22 | 11 (1) |
| | 130 | 11 (1) | 52 | 9.6 (1) | 290 | 50 | 9.7 (1) | 110 | 80 | 4.7 (1) | 150 | 100 | 6.7 (1) |
| | 170 | 9.8 (1) | 55 | 9.5 (1) | 280 | 64 | 9.4 (0.1) | 170 | 47 | 8.3 (0.3) | 230 | 200 | 3.9 (0.4) |
| | 360 | 10 (0.4) | 80 | 8.6 (1) | | | | | | | 500 | 510 | 0.9 (0.1) |
| | 670 | 10 (1) | 150 | 2.5 (0.4) | 270 | 100 | 4.9 (0.3) | 1000 | 51 | 8.6 (0.1) | 1300 | 1500 | 0.9 (0.1) |
| | 2100 | 8.9 (1) | 250 | 0.8 (0.4) | 360 | 120 | 2.0 (0.3) | 1700 | 46 | 8.6 (1) | 3800 | 4600 | 0.7 (0.1) |
| | 6800 | 0.9 (0.1) | 470 | 0.9 (0.1) | 320 | 370 | 0.9 (0.1) | 6200 | 52 | 0.8 (0.3) | 9600 | 13000 | 0.9 (0.1) |
| S3 | 350 | 9.1 (2) | 72 | 9.1 (2) | 700 | 73 | 8.3 (1) | 320 | 130 | 1.0 (0.3) | 350 | 72 | 9.1 (2) |
| | 420 | 8.4 (1) | 81 | 7.3 (1) | 630 | 61 | 7.3 (1) | 380 | 130 | 8.5 (1) | 390 | 130 | 5.7 (1) |
| | 450 | 7.9 (1) | 95 | 4.3 (2) | 710 | 82 | 5.2 (1) | 420 | 140 | 7.4 (1) | 450 | 130 | 6.1 (1) |
| | 700 | 8.3 (1) | 130 | 1.0 (0.3) | | | | | | | 730 | 360 | 6.6 (0.3) |
| | 1300 | 6.8 (1) | 180 | 0.8 (0.3) | 670 | 98 | 2.2 (1) | 1200 | 120 | 9.1 (2) | 1500 | 810 | 3.1 (2) |
| | 3400 | 3.0 (1) | 470 | 1.0 (0.2) | 610 | 200 | 0.6 (0.4) | 3100 | 140 | 5.5 (2) | 3500 | 2200 | 1.3 (0.5) |
| | 8300 | 0.8 (0.1) | 1100 | 0.8 (0.1) | 640 | 1100 | 0.6 (0.3) | 7900 | 130 | 0.4 (0.5) | 9800 | 7400 | 0.8 (0.1) |

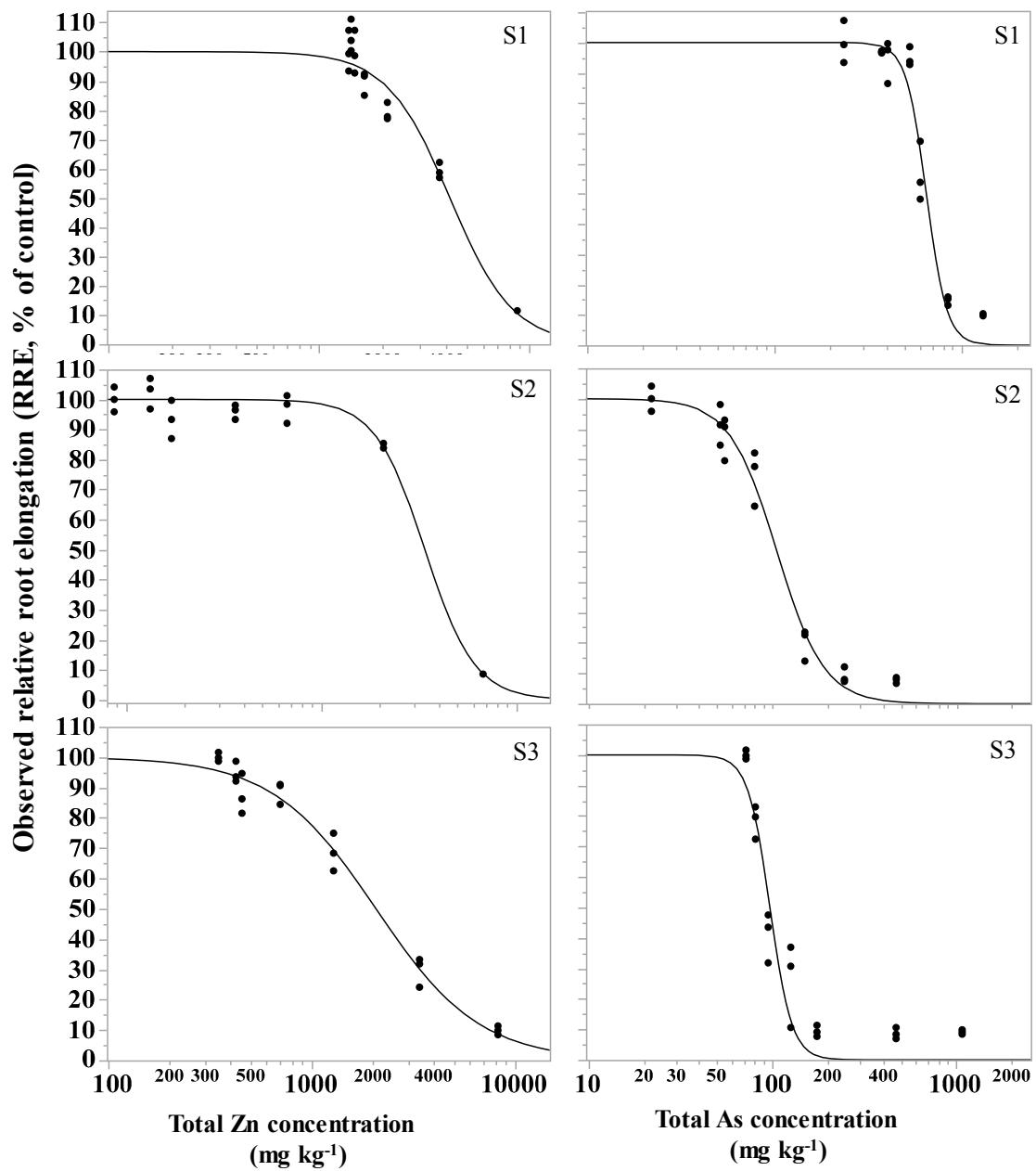


Figure S1. Dose–response curves for total zinc (left) and total arsenate (right) when applied singly to 3 different Mexican soils: S1, Zimapán; S2, Pozos; S3, Taxco. RRE, relative root elongation.

Table S5. Zinc and arsenic EC50 values using different concentration expressions.

| Zn EC50 | | | | | | As EC50 | | | | | | |
|-----------------------------|-----------------------|--------------------------------|----------------------|--|-----------------------|-----------------------------|-----------------------|--------------------------------|----------------------|--|-----------------------|-------|
| Total Zn Soil Concentration | | Soil Solution Zn Concentration | | Soil Solution Zn EC50 Referred to Soil Dry Mass ^a | | Total As Soil Concentration | | Soil Solution As Concentration | | Soil Solution As EC50 Referred to Soil Dry Mass ^a | | |
| mg kg ⁻¹ | mmol kg ⁻¹ | mg L ⁻¹ | mmol L ⁻¹ | mg kg ⁻¹ | mmol kg ⁻¹ | mg kg ⁻¹ | mmol kg ⁻¹ | mg L ⁻¹ | mmol L ⁻¹ | mg kg ⁻¹ | mmol kg ⁻¹ | |
| S1 | 4200 | 64 | 12 | 0.18 | 4 | 0.06 | 650 | 9 | 8 | 0.11 | 2.64 | 0.04 |
| S2 | 3400 | 52 | 18 | 0.28 | 6 | 0.09 | 110 | 1 | 3 | 0.04 | 0.96 | 0.01 |
| S3 | 2100 | 32 | 2 | 0.03 | 1 | 0.01 | 100 | 1 | 0.26 | 4E-03 | 0.08 | 1E-03 |

^a Considering a solid: liquid ratio of 25 g: 8mL.

Table S6. Zinc and arsenic concentrations ($\mu\text{mol L}^{-1}$) in the pore waters of selected single treatments for each experimental soil, dose levels are given in Table S2 and correspond to those of Figure 1. Standard errors (SE) are given in brackets.

| Soil | Single Zn | | Single As | | |
|------|------------|------------------------|------------------------|------------------------|------------------------|
| | Dose level | Zn | As | Zn | As |
| | | $\mu\text{mol L}^{-1}$ | $\mu\text{mol L}^{-1}$ | $\mu\text{mol L}^{-1}$ | $\mu\text{mol L}^{-1}$ |
| S1 | 0 | 3 (1) | 1.8 (0.2) | 3 (1) | 1.8 (0.2) |
| | 2 | 5 (0.1) | 1.2 (0.1) | 5 (0.2) | 5.8 (0.1) |
| | 4 | 17 (0.4) | 1.0 (0.1) | 8 (1) | 75 (4) |
| | 6 | 3500 (140) | 0.5 (<0.1) | 11 (0.2) | 4200 (120) |
| S2 | 0 | 3.1 (0.1) | 0.6 (0.1) | 3 (0.1) | 0.5 (0.1) |
| | 2 | 6.8 (0.1) | 0.5 (0.1) | 3 (<0.1) | 8 (1) |
| | 4 | 12 (1) | 0.3 (<0.1) | 2 (0.3) | 123 (2) |
| | 6 | 3000 (16) | 0.1 (<0.1) | 1 (0.3) | 2400 (7) |
| S3 | 0 | 1.4 (0.1) | 0.3 (<0.1) | 1.4 (0.1) | 0.3 (<0.1) |
| | 2 | 6.7 (0.3) | 0.3 (<0.1) | 2.0 (0.2) | 4.3 (0.1) |
| | 4 | 16 (0.1) | 0.3 (0.1) | 1.4 (0.2) | 79 (4) |
| | 6 | 2400 (80) | 0.4 (0.1) | 0.8 (0.1) | 5600 (80) |

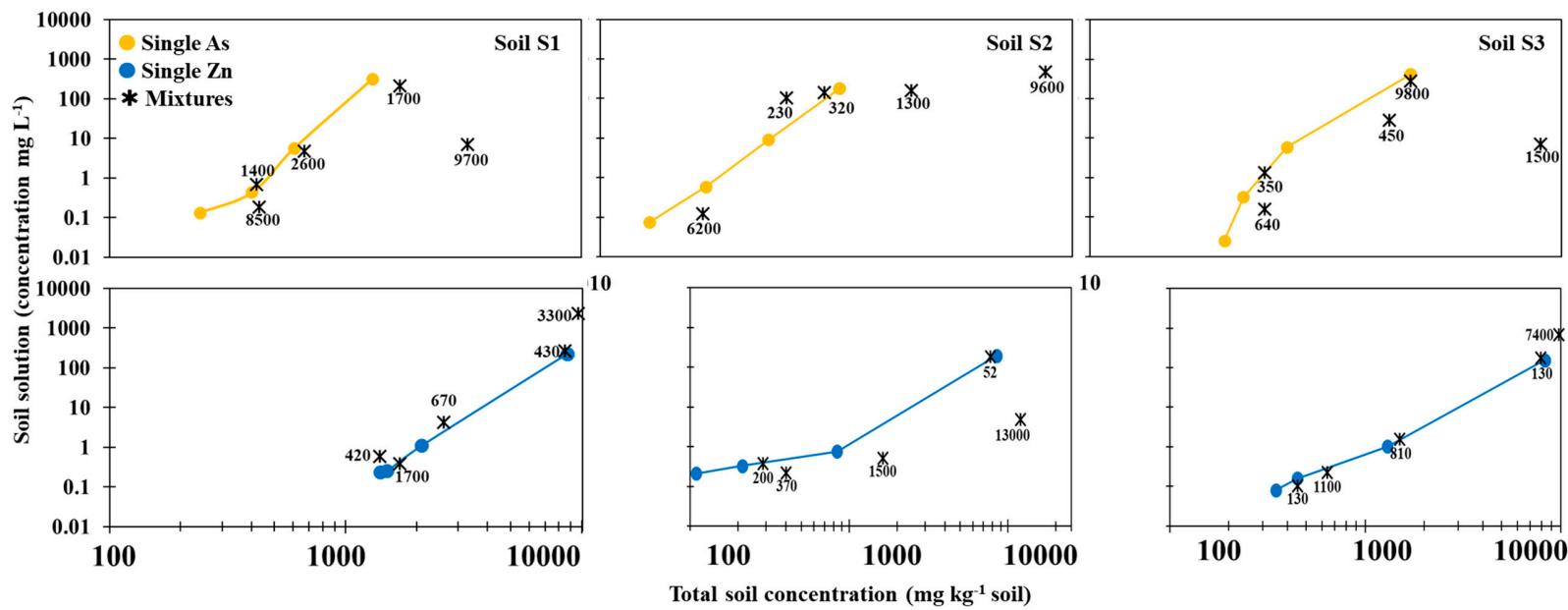


Figure S2. Soil solution concentration as a function of total soil concentration for As (top) and Zn (bottom) in single and mixture treatments. Data labels in the top panel present the Zn total concentration (mg Zn kg^{-1} soil) in each mixture. Data labels in the bottom panel present the As total concentration (mg As kg^{-1} soil) in each soil. Total and soil solution concentrations can be consulted in Table S7.

Table S7. Total (mg kg^{-1}) and soil solution (mg L^{-1}) concentrations of As and Zn for selected single (-) and mixture (+) treatments.

| Soil | Dose level | As Concentration (Total: mg kg^{-1} ; Dissolved: mg L^{-1}) | | | | Zn Concentration (Total: mg kg^{-1} ; Dissolved: mg L^{-1}) | | | |
|-----------|------------|---|-----------|-----------------------------|-----------|---|-----------|-----------------------------|-----------|
| | | Single Treatments (-Zn) | | Mixture Treatments (+Zn) | | Single Treatments (-As) | | Mixture Treatments (+As) | |
| | | Total | Dissolved | Total | Dissolved | Total | Dissolved | Total | Dissolved |
| S1 | 0 | 240 | 0.1 | -- | -- | 1400 | 0.2 | -- | -- |
| | 2 | 400 | 0.4 | 420 | 0.7 | 1500 | 0.3 | 1400 | 0.6 |
| | 4 | 604 | 5.6 | 670 | 5 | 2100 | 1 | 2600 | 4 |
| | 6 | 1300 | 311.8 | 3300 | 7 | 8700 | 226 | 9700 | 2293 |
| | 6 | 1300 | 311.8 | 1700 | 210 | 8700 | 226 | 1700 | 0.4 |
| | 3 | -- | -- | 430 | 0.2 | -- | -- | 8500 | 265 |
| S2 | 0 | 22 | 0.1 | -- | -- | 87 | 0.2 | -- | -- |
| | 2 | 55 | 0.6 | 200 | 102 | 170 | 0.3 | 230 | 0.4 |
| | 4 | 150 | 9.2 | 1500 | 158 | 670 | 0.8 | 1300 | 0.5 |
| | 6 | 470 | 180.9 | 13000 | 462 | 6800 | 199 | 9600 | 5 |
| | 6 | 470 | 180.9 | 370 | 143 | 6800 | 199 | 320 | 0.2 |
| | 3 | -- | -- | 52 | 0.1 | -- | -- | 6200 | 185 |
| S3 | 0 | 72 | 0.0 | -- | -- | 350 | 0.1 | -- | -- |
| | 2 | 95 | 0.3 | 130 | 1 | 450 | 0.2 | 450 | 0.1 |
| | 4 | 180 | 5.9 | 810 | 29 | 1300 | 1 | 1500 | 2 |
| | 6 | 1100 | 416.6 | 7400 | 7 | 8300 | 155 | 9800 | 689 |
| | 6 | 1100 | 416.6 | 1100 | 281 | 8300 | 155 | 640 | 0.2 |
| | 3 | -- | -- | 130 | 0.2 | -- | -- | 7900 | 175 |

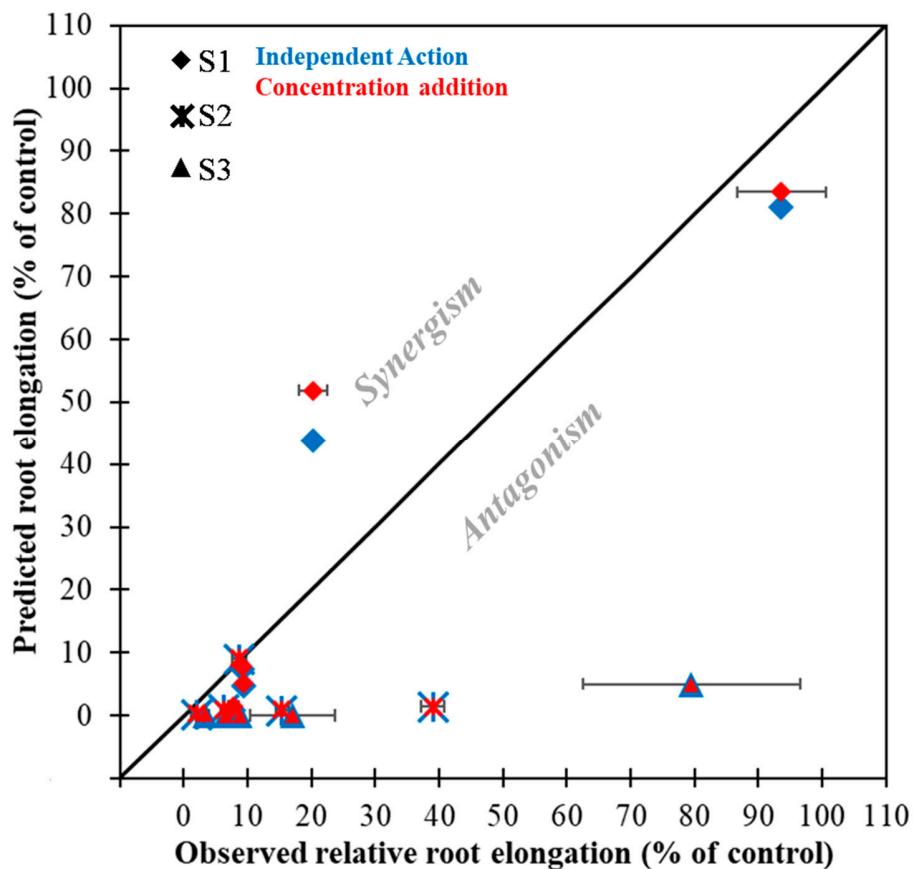


Figure S3. Predicted vs observed root relative elongation using soil solution Zn and Arsenic concentrations as dose expression. Predicted responses were obtained from CA (red) and IA (blue) reference models. Errors bars correspond to the 95% confidence interval (CI) for each treatment observation.

Table S8. Log transformed ion activity products (log IAP) of the solid species that could precipitate from those components with high soil solution concentration at highly dosed treatments. The IAP given in bold suggest saturation and potential precipitation. The solubility products (log K_{sp}) are given as reference.

| Soil | Dose Level | | Zn ($\mu\text{mol L}^{-1}$) | AsO ₄ ($\mu\text{mol L}^{-1}$) | Ca (mmol L^{-1}) | Soil Solution pH | log IAP | | |
|------|------------|----|----------------------------------|--|--------------------------------|------------------------|-----------------------------------|--|--|
| | Zn | As | | | | | As ₂ O _{5(s)} | Ca ₃ (AsO ₄) ₂ 4H ₂ O _(s) | Zn ₃ (AsO ₄) ₂ 2.5H ₂ O _(s) |
| S1 | 0 | 0 | 3.6 | 1.8 | 7.7 | 6.9 | -64 | -29 | -39 |
| | 2 | 0 | 3.9 | 1.2 | 8.9 | 7.5 | -64 | -29 | -39 |
| | 4 | 0 | 17 | 1 | 12 | | -65 | -29 | -38 |
| | 6 | 0 | 3449 | 0.5 | 137 | 7 | -66 | -28 | -33 |
| | 0 | 2 | 4.5 | 6 | 6.5 | 7.6 | -63 | -29 | -39 |
| | 0 | 4 | 0.6 | 75 | 6.6 | | -61 | -26 | -41 |
| | 0 | 6 | 9 | 4163 | 5.3 | 7.6 | -57 | -23 | -31 |
| | 2 | 2 | 9 | 9 | 8.3 | 7.5 | -63 | -28 | -36 |
| S2 | 4 | 4 | 64 | 63 | 27 | | -61 | -25 | -33 |
| | 6 | 6 | 35056 | 93 | 111 | 6 | -63 | -26 | -28 |
| | 0 | 0 | 3.3 | 1 | 4.4 | 6.8 | -64 | -30 | -40 |
| | 2 | 0 | 5 | 0.5 | 4.3 | 7.5 | -65 | -31 | -39 |
| S3 | 4 | 0 | 12 | 0.5 | 8.9 | | -65 | -30 | -39 |
| | 6 | 0 | 3043 | 0.1 | 50 | 7.6 | -67 | -30 | -34 |

| | | | | | | | | | |
|-----------------|---|---|-------|------|-----|------------|------------|------------|-----|
| S3 | 0 | 2 | 7.7 | 8 | 4.7 | 7.6 | -63 | -28 | -37 |
| | 0 | 4 | 1.7 | 123 | 6.1 | 7.7 | -60 | -26 | -36 |
| | 0 | 6 | 0.7 | 2415 | 4 | 7.8 | -58 | -24 | -35 |
| | 2 | 2 | 6 | 1364 | 8.2 | 7.7 | -58 | -23 | -33 |
| | 4 | 4 | 8 | 2103 | 10 | 7.4 | -58 | -23 | -32 |
| | 6 | 6 | 75 | 6166 | 36 | 6.3 | -57 | -21 | -29 |
| | 0 | 0 | 1.3 | 0.3 | 7.4 | 6.9 | -66 | -31 | -42 |
| | 2 | 0 | 4.7 | 0.3 | 4.1 | 7.4 | -65 | -31 | -40 |
| | 4 | 0 | 16 | 0.2 | 16 | | -66 | -30 | -39 |
| | 6 | 0 | 2362 | 0.4 | 103 | 7 | -66 | -28 | -33 |
| | 0 | 2 | 1.2 | 4.2 | 5.8 | 7.6 | -63 | -28 | -40 |
| | 0 | 4 | 1.2 | 79 | 4.9 | | -61 | -26 | -37 |
| | 0 | 6 | 1 | 5562 | 4.5 | 7.6 | -57 | -23 | -34 |
| | 2 | 2 | 1.6 | 18 | 7.3 | 7.6 | -62 | -27 | -38 |
| | 4 | 4 | 24 | 385 | 19 | | -59 | -24 | -32 |
| | 6 | 6 | 10530 | 4332 | 99 | 6.2 | -61 | -23 | -28 |
| log Ksp: | | | | | | -35 | -19 | -28 | |

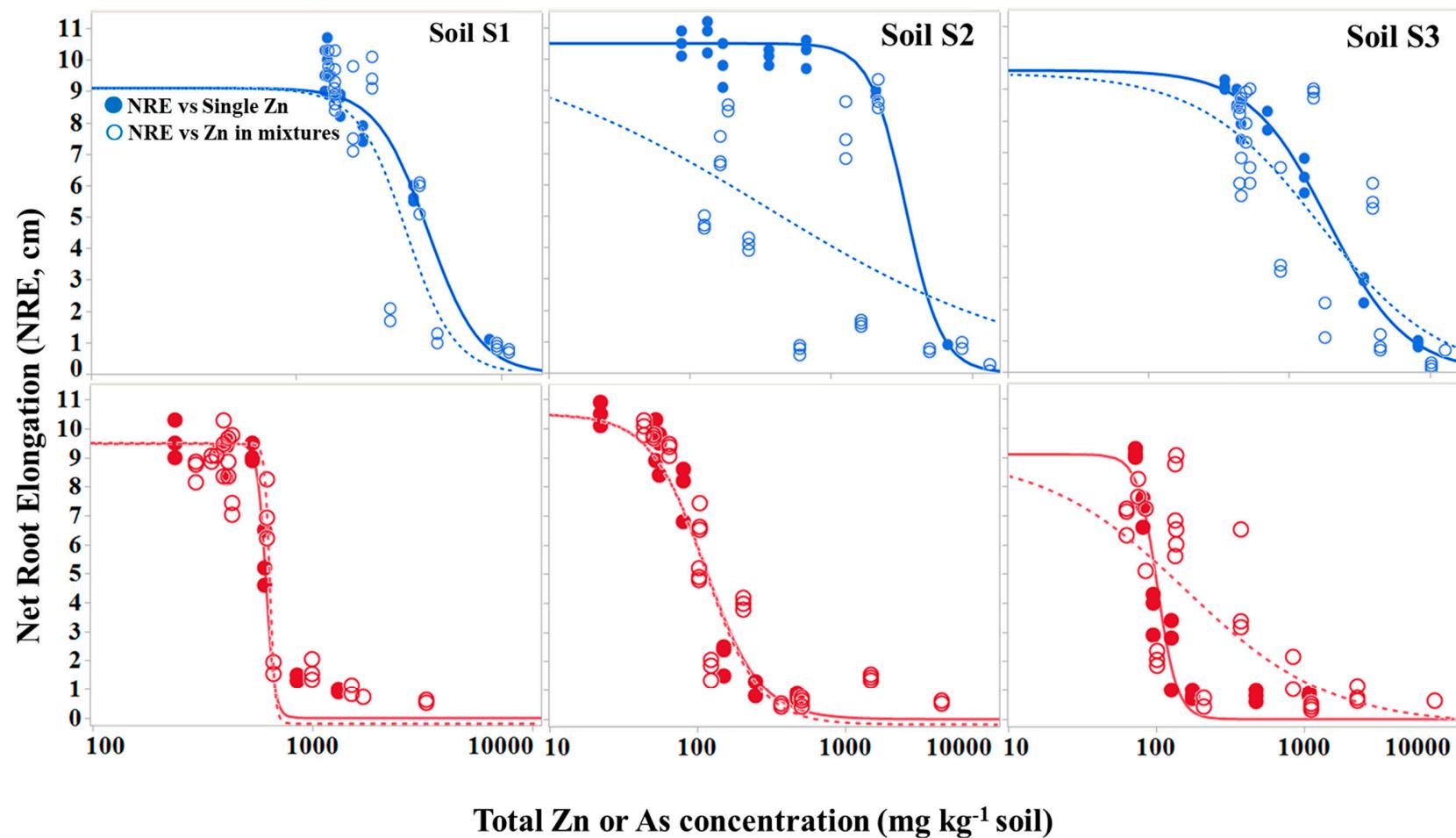


Figure S4. Barley net root elongation (NRE, cm) response to total Zn (top; in blue) or As (bottom, red) soil concentrations (mg kg^{-1}) in single and mixture treatments.

References

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