1 Three-chain simulations with alpha = 0.1, beta = 0

Three-chain model with artificial data (noise-free kinetic data, noise-free state data) - alpha = 0.1, beta = 0

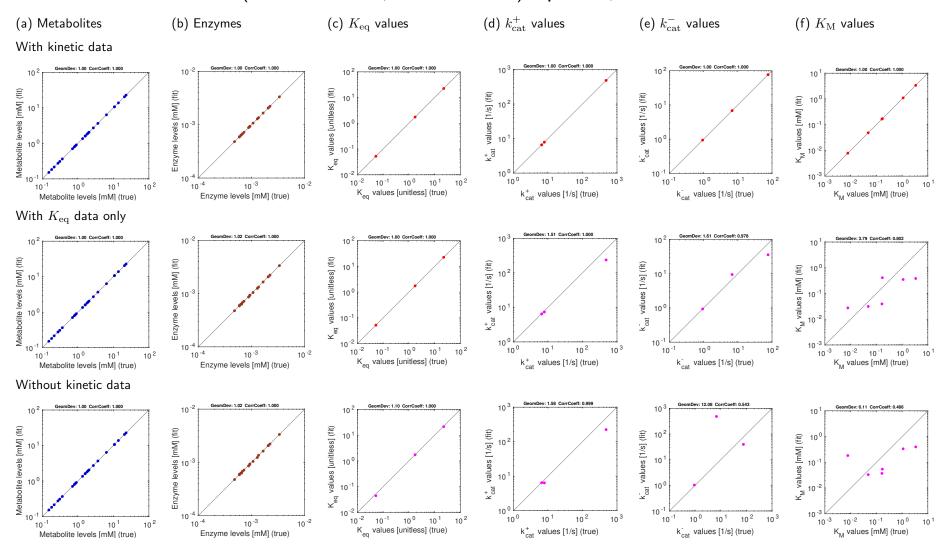


Figure 3: Model balancing results for Three-chain model with artificial data. The model structure is shown in Figure ??. Each subfigure shows "true" values (x-axis) versus reconstructed values (y-axis). Similarities are quntified by geometric standard deviations ("GeomDev") and Pearson correlation coefficients ("CorrCoeff"). (a) Metabolite levels. (b) Enzyme levels. (c)-(f) Different types of kinetic constants. Rows show different estimation scenarios (see Figure) Upper row: simple scenario S1 (noise-free artificial data, data for kinetic constants). Centre row: scenario S2 (noise-free artificial data, no data for kinetic constants). Depending on the scenario, kinetic constants are either fitted (red dots) or predicted (magenta dots).

Three-chain model with artificial data (noisy kinetic data, noise-free state data) - alpha = 0.1, beta = 0

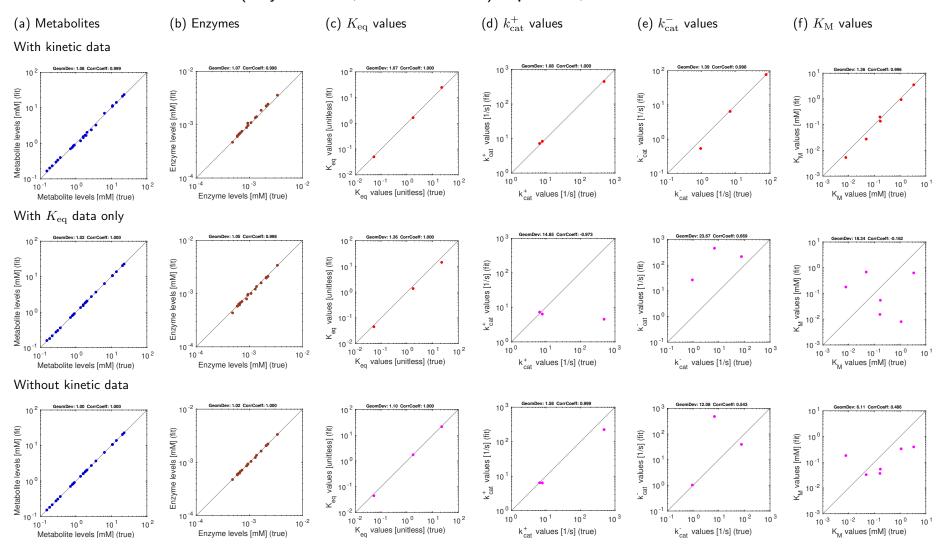


Figure 4: Same as Figure 8, with noisy kinetic data

Three-chain model with artificial data (noise-free kinetic data, noisy state data) - alpha = 0.1, beta = 0

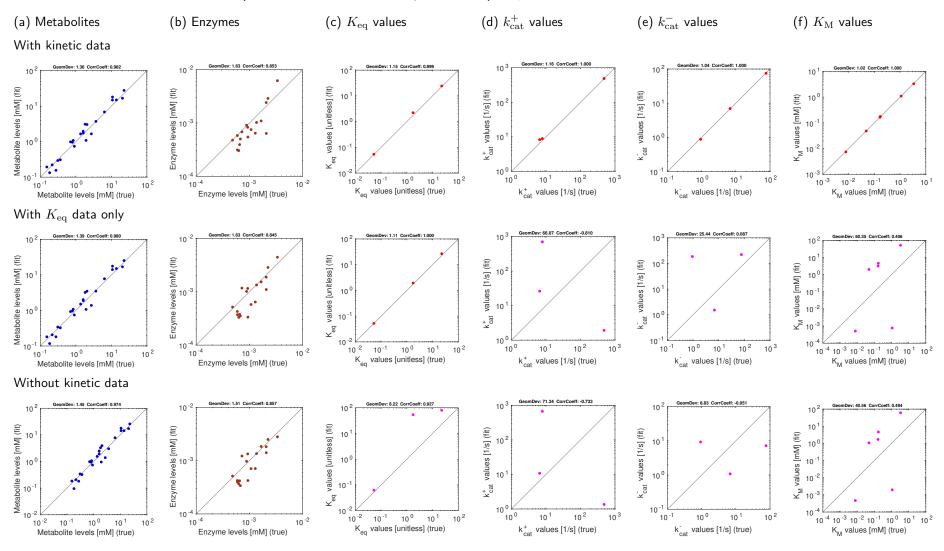


Figure 5: Results for Three-chain with noisy artificial data. Top row: estimation scenario S3 (noisy artificial data, data used for kinetic constants). Centre row: estimation scenario S3K (noisy artificial data, data for equilibrium constants only). Bottom row: estimation scenario S4 (noisy artificial data, no data for kinetic constants).

Three-chain model with artificial data (noisy kinetic data, noisy state data) - alpha = 0.1, beta = 0(c) $K_{ m eq}$ values (d) $k_{\rm cat}^+$ values (e) $k_{\rm cat}^-$ values (a) Metabolites (b) Enzymes (f) $K_{ m M}$ values With kinetic data GeomDev: 1.34 CorrCoeff: 0.998 GeomDev: 1.19 CorrCoeff: 0.999 10 102 GeomDev: 1.47 CorrCoeff: 0.989 10² 10² 10 Metabolite levels [mM] (fit) values [unitless] (fit) values [1/s] (fit) Enzyme levels [mM] (fit) 10 values [1/s] (fit) values [mM] (fit) 10¹ 10⁰ 10 -10⁰ 10 + cat , cat ⊼_@ 10² 10⁰ 10¹ 10³ 10¹ 10² 10⁻² 10 ¹ 10 ⁻¹ 10 ⁻³ 10 ¹ 10² 10⁻⁴ K_{eq} values [unitless] (true) k_{cat} values [1/s] (true) k+ values [1/s] (true) K_M values [mM] (true) Metabolite levels [mM] (true) Enzyme levels [mM] (true) With K_{eq} data only GeomDev: 17.90 CorrCoeff: 0.163 10 10³ GeomDev: 54.66 CorrCoeff: 0.406 10-2 10 Metabolite levels [mM] (fit) Enzyme levels [mM] (fit) values [unitless] (fit) values [1/s] (fit) 10 10 values [1/s] (fit) 10² values [mM] (fit) 10⁰ 10 ¹ 10⁰ 10 -10 -2 10⁰ 10 k+ cat , cat ⊼ 8 10 " 10² 10 ¹ 10² 10¹ 10³ 10⁻¹ 10³ 10⁻² 10⁻¹ 10 ⁰ 10 ¹ 10⁻⁴ 10⁻³ 10⁻² 10⁻¹ 10⁰ 10¹ 10² 10 ⁻³ 10 ¹ 10² 10⁻⁴ 10 ⁻² K_{eq} values [unitless] (true) k at values [1/s] (true) k_{cat} values [1/s] (true) K_M values [mM] (true) Metabolite levels [mM] (true) Enzyme levels [mM] (true) Without kinetic data nDev: 1.51 CorrCoeff: 0.857 GeomDev: 8.22 CorrCoeff: 0.927 GeomDev: 8.83 CorrCoeff: -0.051 GeomDev: 40.56 CorrCoeff: 0.494 102 102 10 -2 10 Metabolite levels [mM] (fit) values [1/s] (fit) 10 values [unitless] (fit) Enzyme levels [mM] (fit) values [1/s] (fit) values [mM] (fit) 10 ¹ 10² 10¹ 10 ⁰ 10⁰ 10⁻¹ , cat κ cat 10 -10² 10⁰ 10¹ 10³ 10⁻¹ 10 ⁰ 10 ¹ 10² 10 ⁰ 10 ¹ 10 -4 10 -3 10 -2 10 -1 10 0 10 1 10 2 10⁻² 10 ⁰ 10⁻⁴ 10 ⁻³ 10 ⁻² 10 ⁻¹ 10 ¹ 10² K_{eq} values [unitless] (true)

Figure 6: Same as Figure 10, with noisy kinetic data

Enzyme levels [mM] (true)

Metabolite levels [mM] (true)

k+ values [1/s] (true)

k-at values [1/s] (true)

K_M values [mM] (true)

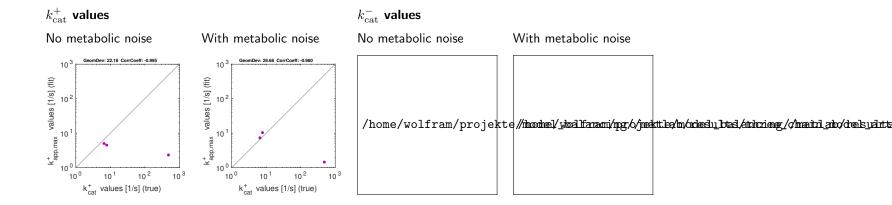


Figure 7: Catalytic constants in Three-chain (artificial data), estimated by kinetic profiling. Note that k_{cat} values can only be estimated in the direction of fluxes (e.g. k_{cat}^+ for reactions with forward fluxe).

2 Three-chain simulations with alpha = 1, beta = 0

Three-chain model with artificial data (noise-free kinetic data, noise-free state data) - alpha = 1, beta = 0

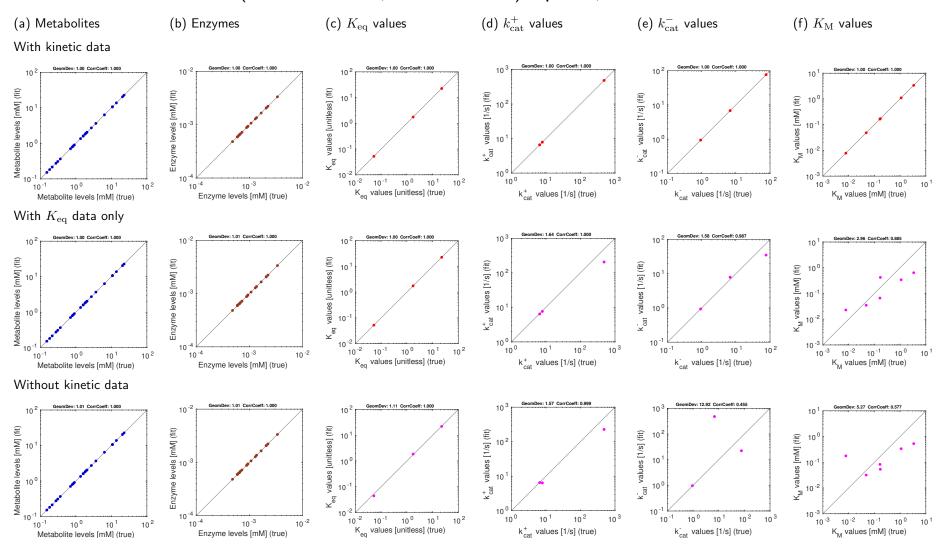


Figure 8: Model balancing results for Three-chain model with artificial data. The model structure is shown in Figure ??. Each subfigure shows "true" values (x-axis) versus reconstructed values (y-axis). Similarities are quntified by geometric standard deviations ("GeomDev") and Pearson correlation coefficients ("CorrCoeff"). (a) Metabolite levels. (b) Enzyme levels. (c)-(f) Different types of kinetic constants. Rows show different estimation scenarios (see Figure) Upper row: simple scenario S1 (noise-free artificial data, data for kinetic constants). Centre row: scenario S2 (noise-free artificial data, no data for kinetic constants). Depending on the scenario, kinetic constants are either fitted (red dots) or predicted (magenta dots).

Three-chain model with artificial data (noisy kinetic data, noise-free state data) - alpha = 1, beta = 0

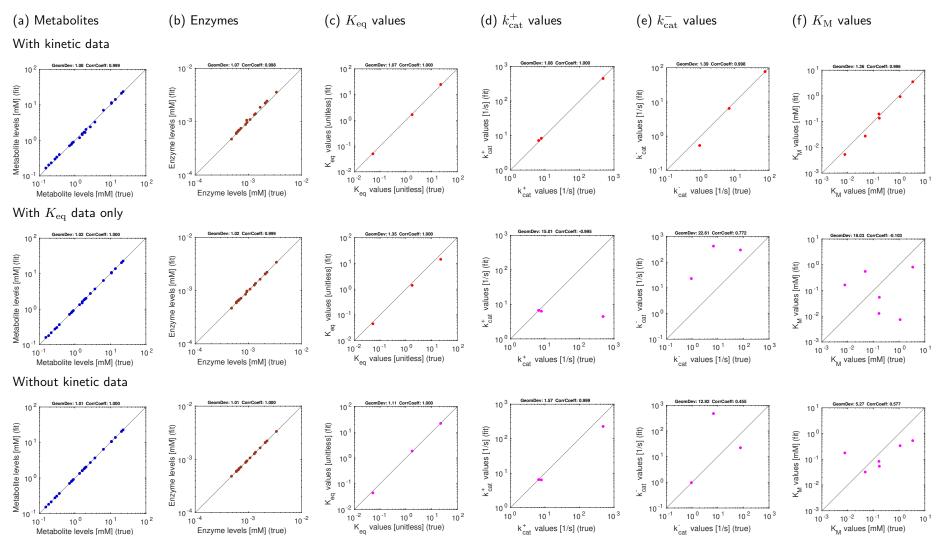


Figure 9: Same as Figure 8, with noisy kinetic data

Three-chain model with artificial data (noise-free kinetic data, noisy state data) - alpha = 1, beta = 0

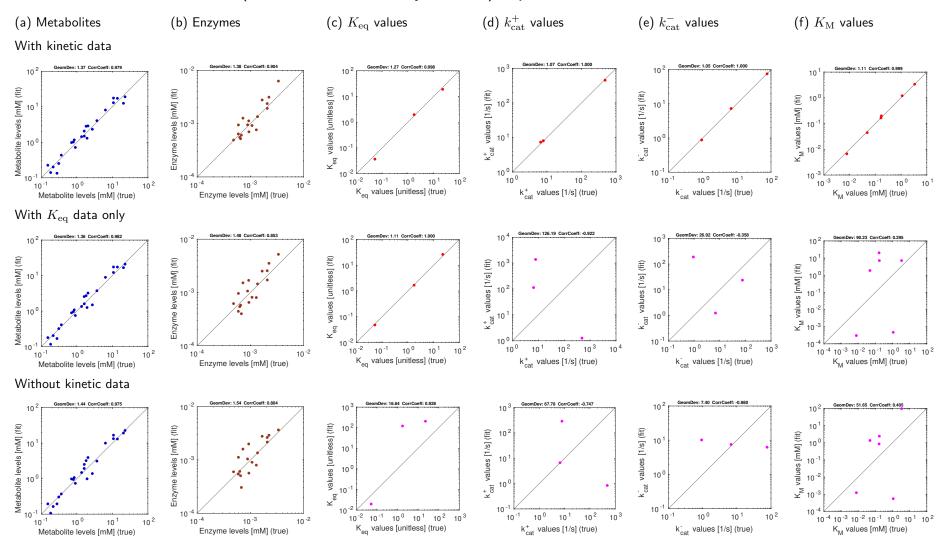


Figure 10: Results for Three-chain with noisy artificial data. Top row: estimation scenario S3 (noisy artificial data, data used for kinetic constants). Centre row: estimation scenario S3K (noisy artificial data, data for equilibrium constants only). Bottom row: estimation scenario S4 (noisy artificial data, no data for kinetic constants).

10⁻¹

10 ¹

Metabolite levels [mM] (true)

10²

Three-chain model with artificial data (noisy kinetic data, noisy state data) - alpha = 1, beta = 0(c) $K_{ m eq}$ values (d) $k_{\rm cat}^+$ values (e) $k_{\rm cat}^-$ values (a) Metabolites (b) Enzymes (f) $K_{ m M}$ values With kinetic data GeomDev: 1.23 CorrCoeff: 0.999 GeomDev: 1.20 CorrCoeff: 0.999 10 102 GeomBey: 1.49 CorrCoeff: 0.986 10² 10² Metabolite levels [mM] (fit) values [unitless] (fit) values [1/s] (fit) Enzyme levels [mM] (fit) 10 values [1/s] (fit) values [mM] (fit) 10¹ 10⁰ 10 -10⁰ 10 + cat , cat ⊼_@ 10² 10⁰ 10¹ 10³ 10 ¹ 10² 10⁻² 10⁰ 10 ¹ 10 ⁻¹ 10 ⁻³ 10 ⁻² 10 ¹ 10² 10⁻⁴ K_{eq} values [unitless] (true) k_{cat} values [1/s] (true) K_M values [mM] (true) k+ values [1/s] (true) Metabolite levels [mM] (true) Enzyme levels [mM] (true) With K_{eq} data only GeomDev: 84.67 CorrCoeff: -0.747 GeomDev: 18.49 CorrCoeff: 0.019 10 10³ GeomDev: 62.84 CorrCoeff: 0.366 10-2 10 vels [mM] (fit) values [1/s] (fit) Enzyme levels [mM] (fit) values [unitless] (fit) 10 10 values [1/s] (fit) 10² values [mM] (fit) 10⁰ 10 ¹ 10⁰ 10 Metabolite lev 10 -2 10⁰ 10 k+ cat , cat ⊼ 8 10² 10 ¹ 10² 10¹ 10³ 10⁻¹ 10³ 10⁻² 10⁻¹ 10 ⁰ 10 ¹ 10⁻⁴ 10⁻³ 10⁻² 10⁻¹ 10⁰ 10¹ 10² 10 ⁻³ 10⁰ 10 ¹ 10² 10⁻⁴ 10 ⁻² K_{eq} values [unitless] (true) k at values [1/s] (true) K_M values [mM] (true) k_{cat} values [1/s] (true) Metabolite levels [mM] (true) Enzyme levels [mM] (true) Without kinetic data nDev: 1.54 CorrCoeff: 0.804 GeomDev: 67.78 CorrCoeff: -0.747 GeomDev: 51.65 CorrCoeff: 0.405 10² 10-2 10 10² Metabolite levels [mM] (fit) 10¹ values [unitless] (fit) Enzyme levels [mM] (fit) 10² values [mM] (fit) values [1/s] (fit) values [1/s] (fit) 10² 10 ¹ 10 10 ¹ 10⁰ 10 ⁰ te 10 0 } k cat 10

Figure 11: Same as Figure 10, with noisy kinetic data

10⁻¹ 10⁰ 10¹ 10² 10³

K_{eq} values [unitless] (true)

10⁻⁴

10 ⁻³

Enzyme levels [mM] (true)

10 ⁻²

10²

10⁰ 10¹

k+ values [1/s] (true)

10 ⁻¹

10 ¹

k cat values [1/s] (true)

10⁻⁴ 10⁻³ 10⁻² 10⁻¹ 10⁰ 10¹ 10²

K_M values [mM] (true)

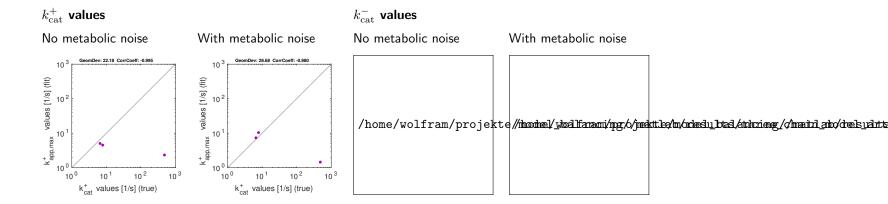


Figure 12: Catalytic constants in Three-chain (artificial data), estimated by kinetic profiling. Note that $k_{\rm cat}$ values can only be estimated in the direction of fluxes (e.g. $k_{\rm cat}^+$ for reactions with forward fluxe).