Supporting Information for

What's the difference? 2D DIGE image analysis by DeCyder™ versus SameSpots™

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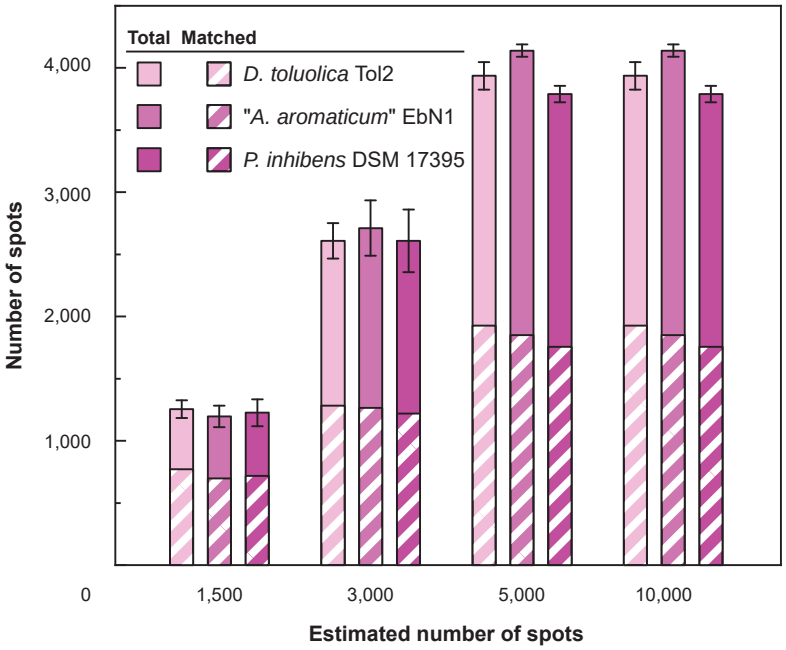


Fig. S1: Effect of the estimated number of spots setting in the DeCyder™ Batch processor module. Average number of detected spots per gel applying different settings for the estimated number of spots within the DeCyder™ software for the three model organisms: *D. toluolica* Tol2 (light pink), "*A. aromaticum*" EbN1 (pink) and *P. inhibens* DSM 17395 (purple). The number of spots matched in all gels of the respective gel-set is indicated by hatching. Standard deviation is indicated.

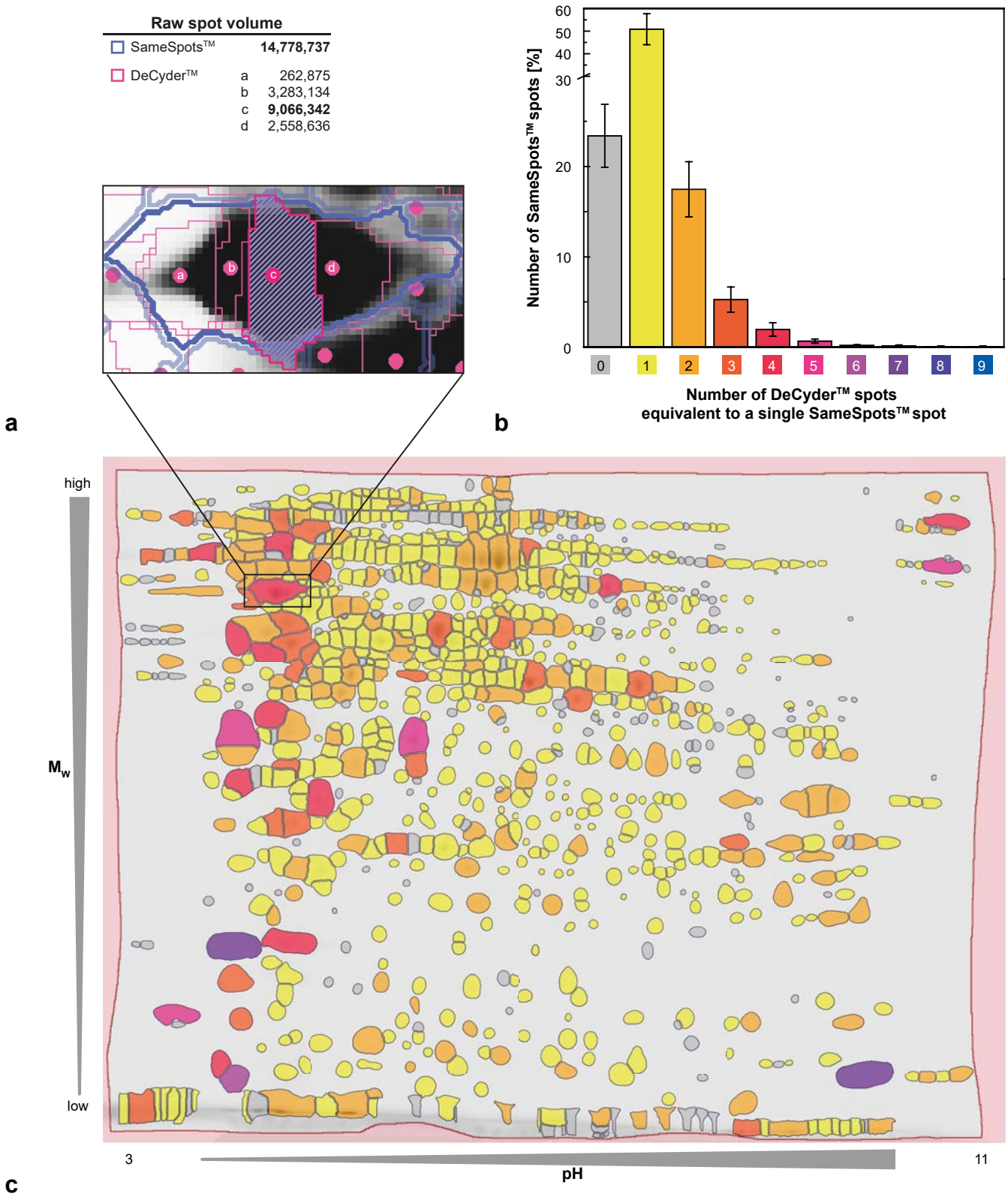
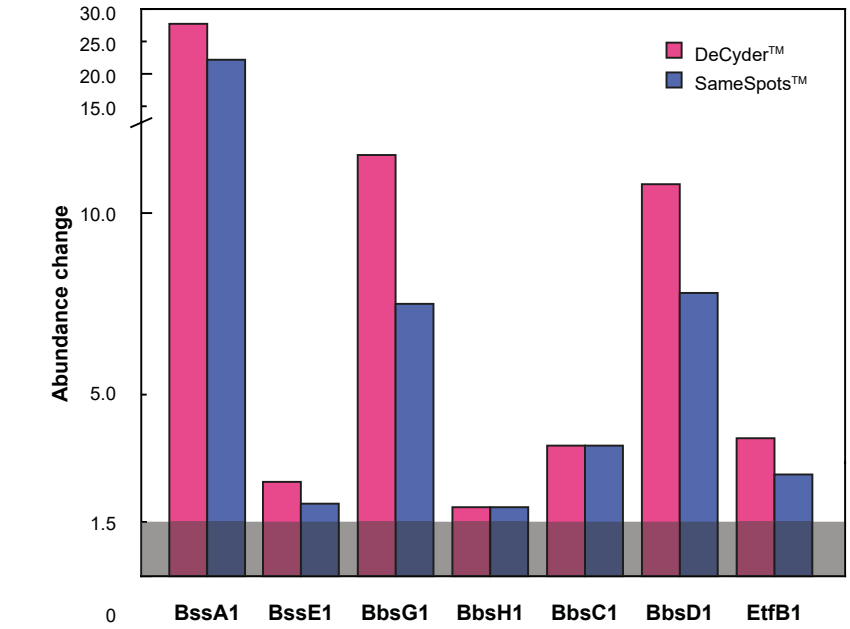
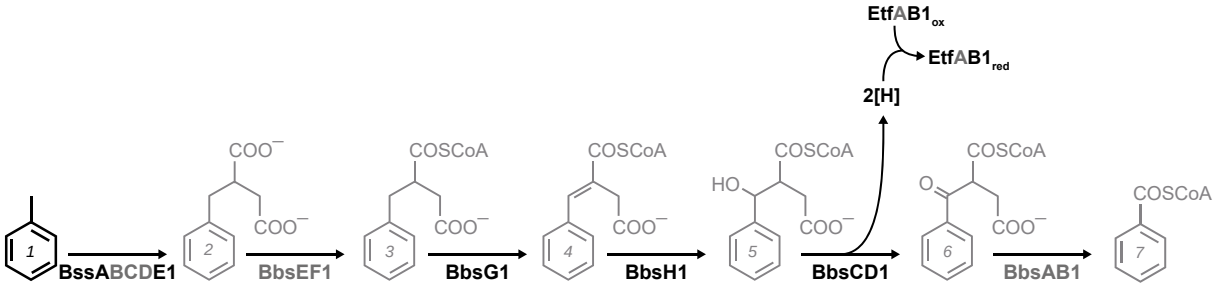


Fig. S2: Spot fractionation encountered with DeCyder™. Image detail of an overlay of DeCyder™ (pink) and SameSpots™ (blue) spot maps (**a**). Multiple DeCyder™ spots could be assigned to one SameSpots™ spot (dark blue boundary), denoted by their center of mass (pink circles, marked a-d). For inter-software matching, the DeCyder™ spots with largest raw volume within the internal standard (here spot c) was assigned to the corresponding SameSpots™ spot (respective volumes indicated). Average share of SameSpots™ spots with none, single, or multiple DeCyder™ spot equivalents of all studied gel-sets (**b**; standard deviation indicated). SameSpots™ spot map of *D. toluolica* Tol2 indicating the number of DeCyder™ spot equivalents per spot (**c**). Color coding according to bar chart b.



a



b

Fig. S3: Abundance changes of proteins involved in anaerobic degradation of toluene by *D. toluolica* Tol2 calculated with DeCyder™ and SampeSpots™ (**a**). Proteins are given in consecutive order of the pathway (**b**). The threshold of significance is indicated by grey shading. Enzyme names are as follows: BssABCDE1, benzylsuccinate synthase; BbsEF1, succinyl-CoA:(*R*)-benzylsuccinate CoA-transferase; BbsG1, (*R*)-benzylsuccinyl-CoA dehydrogenase; BbsH1, phenylitaconyl-CoA hydratase; BbsCD1, 2-[hydroxy(phenyl)methyl]-succinyl-CoA dehydrogenase; BbsAB1, benzoylsuccinyl-CoA thiolase; EftAB1, electron transfer flavoprotein. Compound names: 1, toluene; 2, (*R*)-benzylsuccinate; 3, (*R*)-benzylsuccinyl-CoA; 4, (*E*)-phenylitaconyl-CoA; 5, 2-[hydroxyl(phenyl)methyl]succinyl-CoA; 6, benzoylsuccinyl-CoA; 7, benzoyl-CoA.