

Fig. S1. Hydrolysate coloration before (A) and after (B) charcoal filtration. The hydrolysate was generated by butylamine pretreatment followed by enzymatic hydrolysis and conditioning. For detoxification, the hydrolysate was treated with 1/10 (v/v) of 40 - 60 mesh charcoal and sonicated for 10 min. After allowing the charcoal to settle at room temperature, the clarified supernatant was collected and passed through 0.2 μm filters.

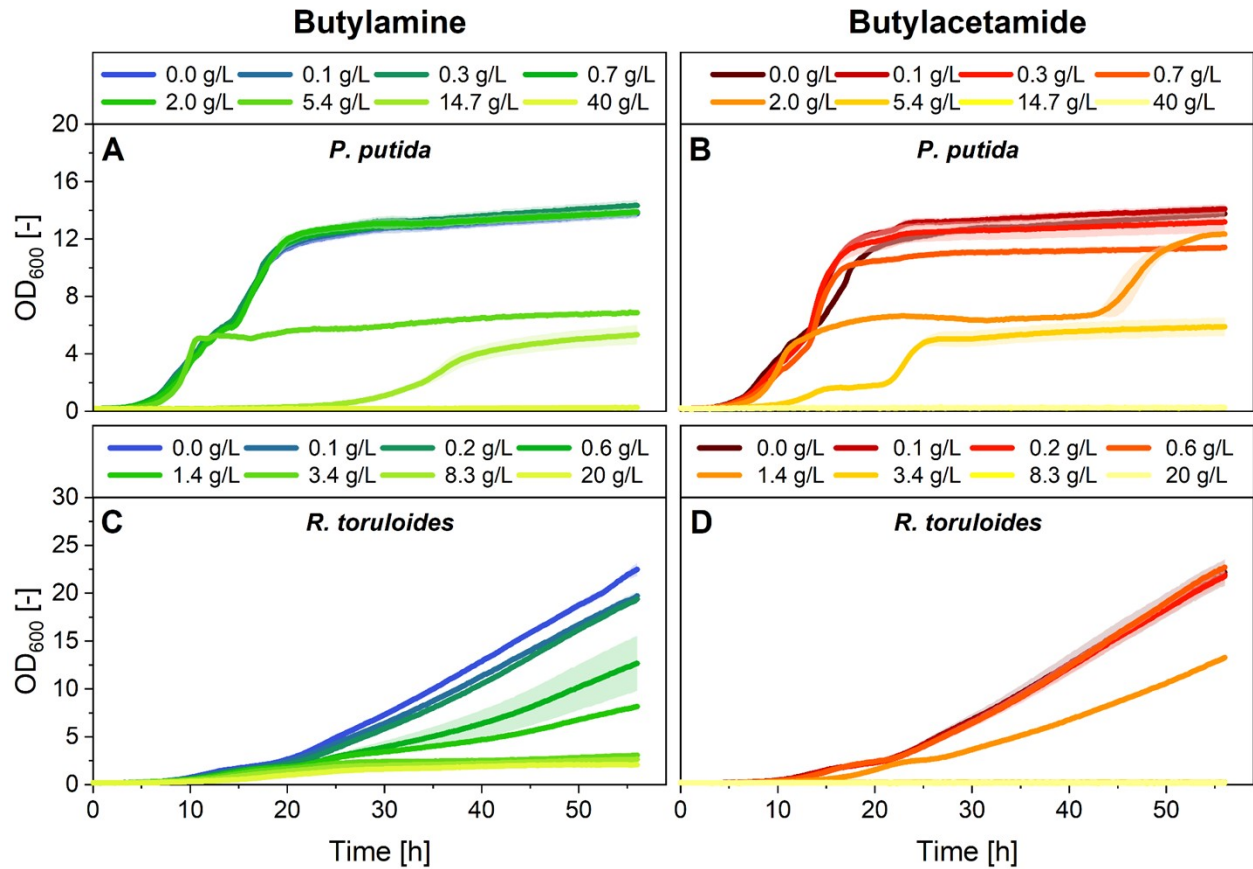


Fig. S2. Growth profiles of *P. putida* (A, B) and *R. toruloides* (C, D) from dose–response assays used for IC₅₀ determination. Cultures were exposed to a log-spaced series of butylamine (blue/green) and butylacetamide (red/yellow) concentrations in chemically defined media. Assays were performed for 56 hours in 48-well FlowerPlates using a microbioreactor system, with each condition tested in triplicate.

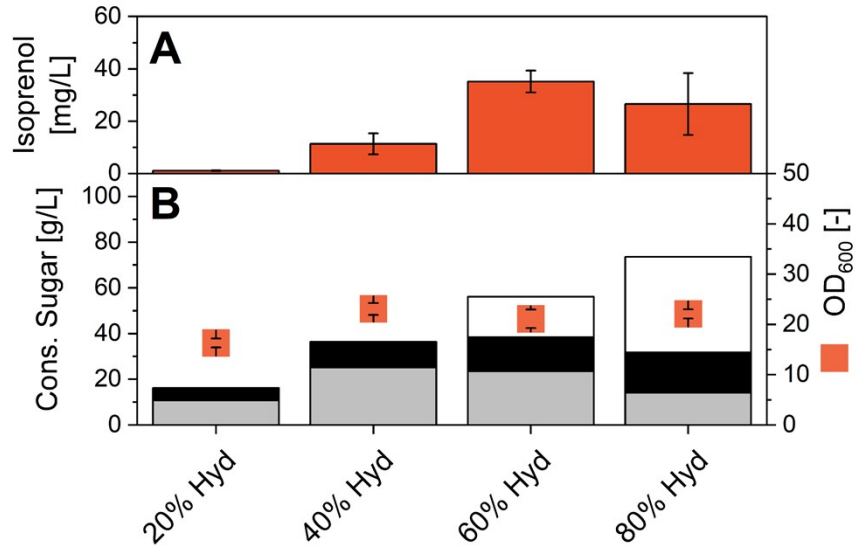


Fig. S3. Hydrolysate concentration test for isoprenol production by *Pseudomonas putida*. Cultures were grown in hydrolysate diluted to different concentrations with water and supplemented with a standard nutrient mix (1× M9 salts, 2 mM MgSO₄, 0.1 mM CaCl₂, 1× trace metal solution, and 5 g/L (NH₄)₂SO₄). (A) Isoprenol titers; (B) Sugar utilization, with total initial sugar (white bars), glucose consumed (grey), and xylose consumed (black); Final cell density (OD₆₀₀, orange) after 72 h.

Table S1. LC-MS analysis of the pretreated hydrolysate. Concentration of sugars, solvent residues, and phenolic compounds in hydrolysate before and after charcoal treatment. Quantified concentrations (g/L) of glucose, xylose, solvent residues (n-butylamine, n-butylacetamide), and identified phenolic compounds in untreated hydrolysate and charcoal-filtered hydrolysate. ND = not detected. The values correspond to the data presented in Figure 2.

Compound	Concentration [g/L]	
	Hydrolysate	Hydrolysate, charcoal-filtered
Glucose	67.46436	68.2709
Xylose	29.02059	29.15766
Benzoic acid	0.57514	ND
4-Hydroxybenzoic acid	0.09801	ND
N-butyl-4-hydroxybenzamide	0.0715	ND
4-Hydroxybenzaldehyde	0.00115	ND
Ferulic acid	0.06423	ND
Syringic acid	0.02462	ND
Vanillic acid	0.02935	ND
Vanillin	0.00955	ND
Protocatechuic acid	0.00141	ND
Salicylic acid	0.00202	ND
Catechol	0.00128	ND
Syringaldehyde	0.00559	ND
Butylamine	0.82502	0.61438

Butylacetamide	7.04495	0.41692
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Table S2. *A. niger* cell biomass concentrations from the butylamine toxicity assay. Biomass was quantified after 144 h as dry cell weight (DCW). Assays were performed in shake flasks with chemically defined medium (30 °C, 200 rpm), following pre-culture from 1×10^8 spores in CM overnight.

Condition	Butylamine [g/L]	Biomass [g/L]
Ctrl	0	10.75 ± 1.07
Lvl. 1	0.1	10.75 ± 1.07
Lvl. 2	0.3	9.5 ± 0.71
Lvl. 3	0.7	10.25 ± 1.07
Lvl. 4	2.0	9.75 ± 0.35
Lvl. 5	5.4	1.00 ± 0.71
Lvl. 6	14.7	0.00 ± 0.00
Lvl. 7	40.0	0.00 ± 0.00

Table S3. *A. niger* cell biomass concentrations from the butylacetamide toxicity assay. Biomass was quantified after 144 h as dry cell weight (DCW). Assays were performed in shake flasks with chemically defined medium (30 °C, 200 rpm), following pre-culture from 1×10^8 spores in CM overnight.

Condition	Butylacetamide [g/L]	Biomass [g/L]
Ctrl	0	9.75 ± 1.77
Lvl. 1	0.1	10.50 ± 0.71
Lvl. 2	0.3	10.75 ± 2.48
Lvl. 3	0.7	8.00 ± 1.41
Lvl. 4	2.0	3.50 ± 0.71
Lvl. 5	5.4	0.00 ± 0.00
Lvl. 6	14.7	0.00 ± 0.00
Lvl. 7	40.0	0.00 ± 0.00

Table S4. Genotypes and features of the microbial strains used in this work.

Genotype/features	Name	Registry ID
<i>A. niger</i> ATCC 11414/ <i>PgpdA</i> - C4t318- <i>TtrpC</i>	JJ1	JBx_275419
<i>P. putida</i> KT2440 Δ phaABC Δ mvaB Δ hbdH Δ ldhA Δ PP2675 Δ gcd::xylABE talBtkA pRK2-PBAD-mvaS-mvaE-Ptrc-MKMM-PMDHKQ-aphA	IY1449SOT	JBx_257853
<i>R. toruloides</i> IFO0880/ <i>PGAPDH</i> -BIS-TNOS, PANT-BIS-TNOS	GB2	ABFPUB_000311

Table S5. Logarithmically spaced concentration ranges of butylamine and butylacetamide used for IC₅₀ determination with the three organisms. Each organism was exposed to a log-spaced series of concentrations to assess the toxicity of each compound. Concentrations were selected to span sub-inhibitory to fully inhibitory levels, enabling robust curve fitting for each species.

	Conc. [g/L]							
Organism	Ctrl	Lvl. 1	Lvl. 2	Lvl. 3	Lvl. 4	Lvl. 5	Lvl. 6	Lvl.7
<i>A. niger</i>	0	0.1	0.3	0.7	2.0	5.4	14.7	40.0
<i>P. putida</i>	0	0.1	0.3	0.7	2.0	5.4	14.7	40.0
<i>R. toruloides</i>	0	0.1	0.2	0.6	1.4	3.4	8.3	20.0