ELECTRONIC SUPPLEMENTARY INFORMATION (E. S. I.)

Pinch-Valve Interface for Automated Sampling and Monitoring of Dynamic Processes by Gas Chromatography – Mass Spectrometry

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ADDITIONAL TABLES

Tab. 1 Calibration equations (*cf.* **Fig. S3**) and limits of detection (LODs) for the four analytes discussed in this report. The LODs were calculated using the $S/N = 3$ criterion, where S is the peak amplitude and N is the RMS noise of baseline. The injection volume resulting from the estimation described in section 3.2 was used to calculate $LOD_{tot.}$. Note well, the conditions used to analyse the first (1-butyl acetate) and the other three (limonene, pinene, terpinene) compounds are different (*cf.* section 2.4).

Compound	EIC (*m/z*, range) / u e^{-1}	Ret. time / min	Internal standard (IS)	EIC of IS (*m/z*, range) / u e^{-1}	Ret. time of IS / min	Calibration equation *A* – area ratio (analyte/IS) *C* – concentration / mM	Coefficient of determination (R^2)	Linear range / mM	$LOD_{conc.}$ / mM (± SD, *n* = 3)	$LOD_{tot.}$ / mmol (± SD, *n* = 3)
1-Butyl acetate	43 ± 0.5	7.9	10^{-5} M limonene	93 ± 0.5	10.7	$A = (240.3 \pm 7.8)C - (0.0550 \pm 0.5481)$	0.989	0.0005-0.0400	$1.97 \times 10^{-4} \pm 2.48 \times 10^{-5}$	$3.34 \times 10^{-10} \pm 4.21 \times 10^{-11}$
Limonene	93 ± 0.5	9.3	10^{-5} M thymol	135 ± 0.5	12.4	$A = (27.8 \pm 2.9)C + (0.0373 \pm 0.0198)$	0.944	0.0001-0.0600	$2.96 \times 10^{-5} \pm 6.89 \times 10^{-6}$	$5.03 \times 10^{-11} \pm 1.17 \times 10^{-11}$
Pinene	93 ± 0.5	8.9	10^{-5} M thymol	135 ± 0.5	12.4	$A = (67.3 \pm 4.4)C + (0.00830 \pm 0.00510)$	0.965	0.0001-0.0600	$1.50 \times 10^{-4} \pm 5.93 \times 10^{-5}$	$2.55 \times 10^{-10} \pm 1.01 \times 10^{-10}$
Terpinene	93 ± 0.5	9.6	10^{-5} M thymol	135 ± 0.5	12.4	$A = (70.6 \pm 5.7)C + (0.00660 \pm 0.00490)$	0.949	0.0001-0.0600	$9.66 \times 10^{-5} \pm 1.92 \times 10^{-5}$	$1.64 \times 10^{-10} \pm 3.26 \times 10^{-11}$

Tab. 2 Calculation of the lipase-catalysed transesterification velocities obtained during the single-microbead transesterification experiment (**Fig. 6**) facilitated by the proposed automated system (**Figs. 1** and **2**).

Microbead symbol (*cf.* Fig. 6)	**Fitted line *A* – area ratio *t* – time / min**	**Reaction velocity / nmol L^{-1} min^{-1}**
1, □	$A = (0.0190 \pm 0.0022)t$	308
2, ○	$A = (0.0208 \pm 0.0014)t$	315
3, □	$A = (0.0241 \pm 0.0012)t$	329
4, ⬩	$A = (0.0293 \pm 0.0030)t$	351
5, ◆	$A = (0.0392 \pm 0.0007)t$	392
6, ❒	$A = (0.0343 \pm 0.0020)t$	372
7, ❋	$A = (0.0378 \pm 0.0023)t$	386
8, ❖	$A = (0.0487 \pm 0.0029)t$	432

ADDITIONAL FIGURES

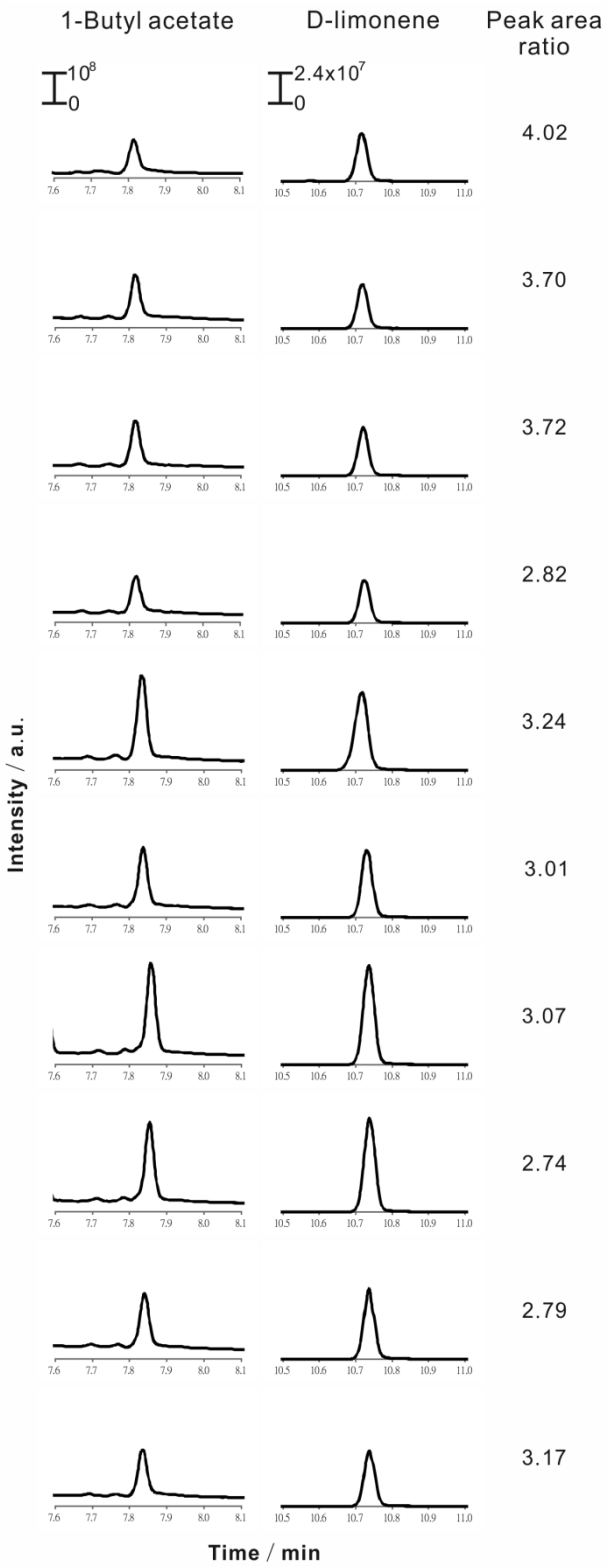

Fig. S1 Injection repeatability test. EICs (analyte *m/z* 43 ± 0.5 u e^{-1}, internal standard *m/z*, 93 ± 0.5 u e^{-1}) from 10 consecutive analyses carried out using the automated sampling system (**Figs. 1** and **2**) combined with GC-MS instrument. Sample: 86.1 μM 1-butyl acetate, 36.7 μM D-limonene (in acetonitrile).

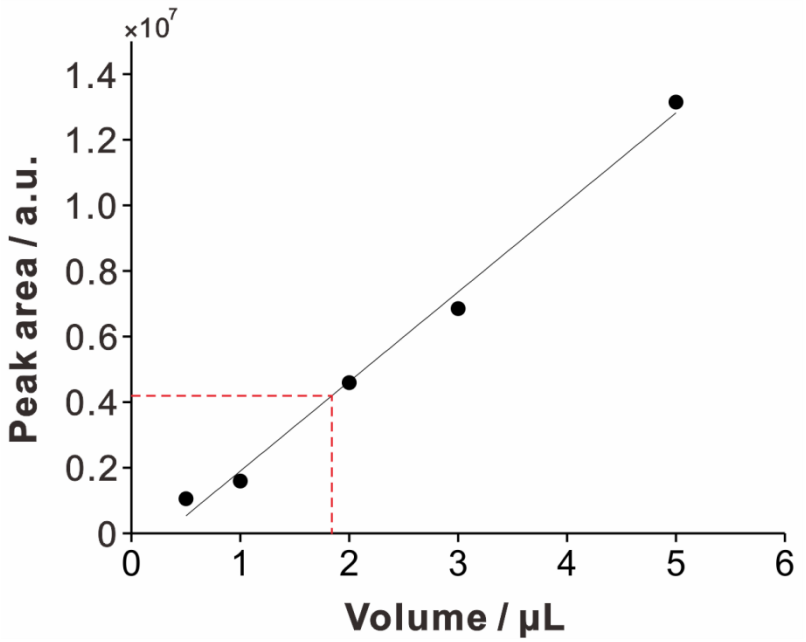

Fig. S2 Determination of the injection volume using the automated sampling/injection system. The black solid markers correspond to the manual injections (without the automated system). The black solid line is a linear function fitted to those data points. The red line extrapolates the peak area (EIC analyte *m/z* 43 ± 0.5 u e^{-1}, internal standard *m/z*, 93 ± 0.5 u e^{-1}) obtained with the automated injection system (n = 3) to the volume axis. The concentration of the 1-butyl acetate standard was 0.5 µM in both cases. Each data point in this graph corresponds to arithmetic average of two replicate results.

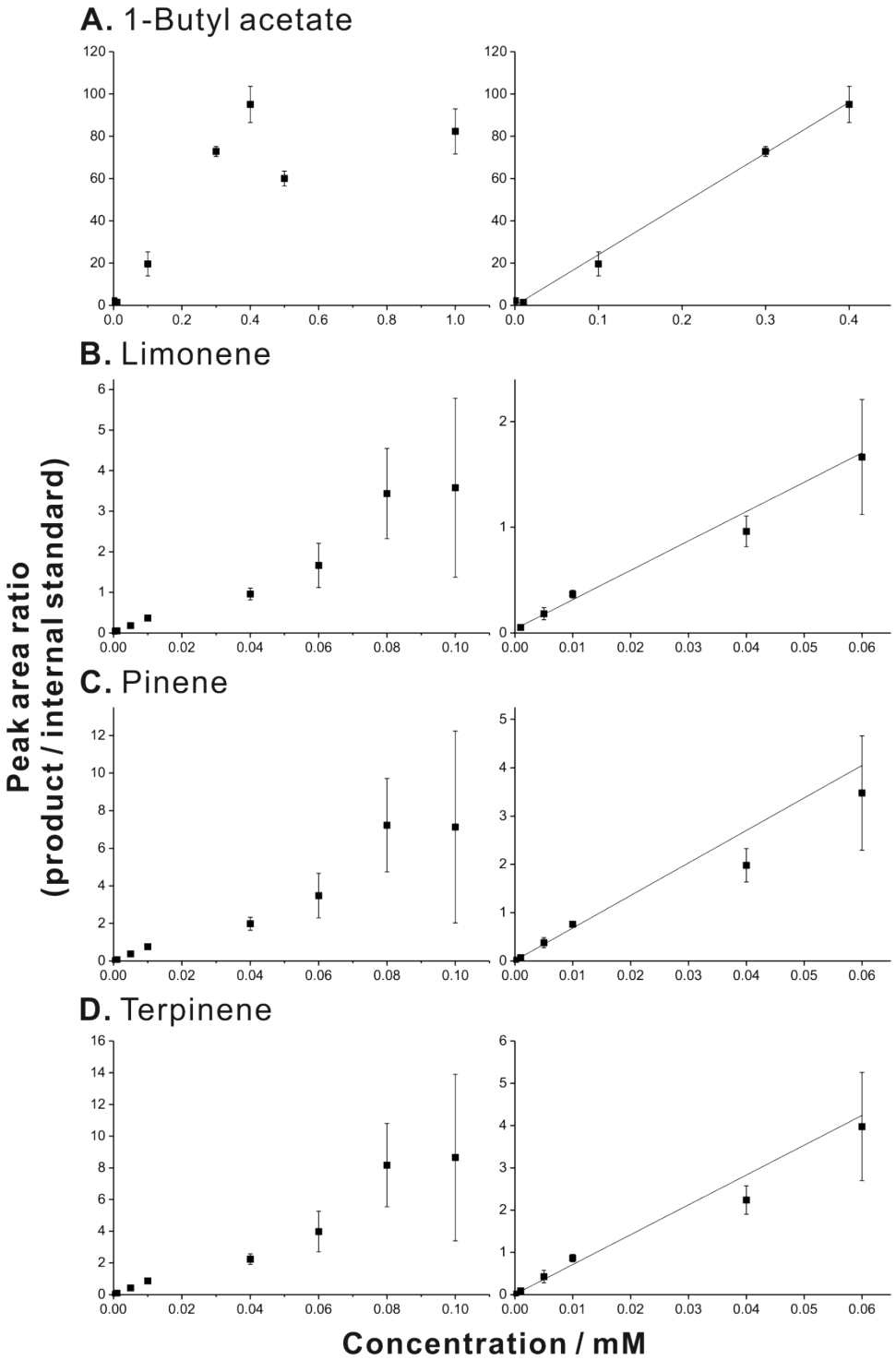

Fig. S3 Quantitative capabilities of the proposed automated sampling system (**Figs. 1** and **2**) coupled with GC-MS. (A) Calibration plot for 1-butyl acetate (D-limonene as internal standard). (B) Calibration plot for D-limonene, (C) β-pinene and (D) γ-terpinene (thymol as internal standard). Error bars correspond to standard deviations (n = 3). For calibration equations, see **Tab. S1**.

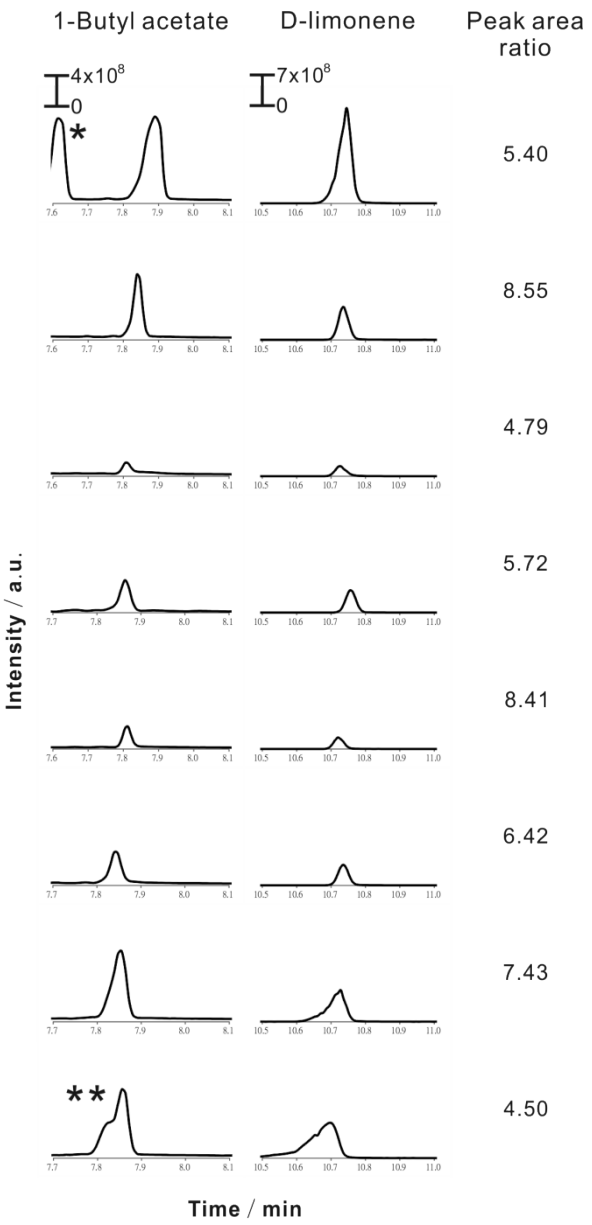

Fig. S4 Raw EICs (analyte *m/z* 43 ± 0.5 u e^{-1}, internal standard *m/z*, 93 ± 0.5 u e^{-1}) for the data in **Fig. 6** (analyses at 126 min). Conditions are the same as in **Fig. 4**. Asterisk (*) indicates a contaminant peak which has a longer retention time when the amount of the reaction product is higher than usual. Two asterisks (**) indicate a fronting feature which is most probably related to an injection artefact.

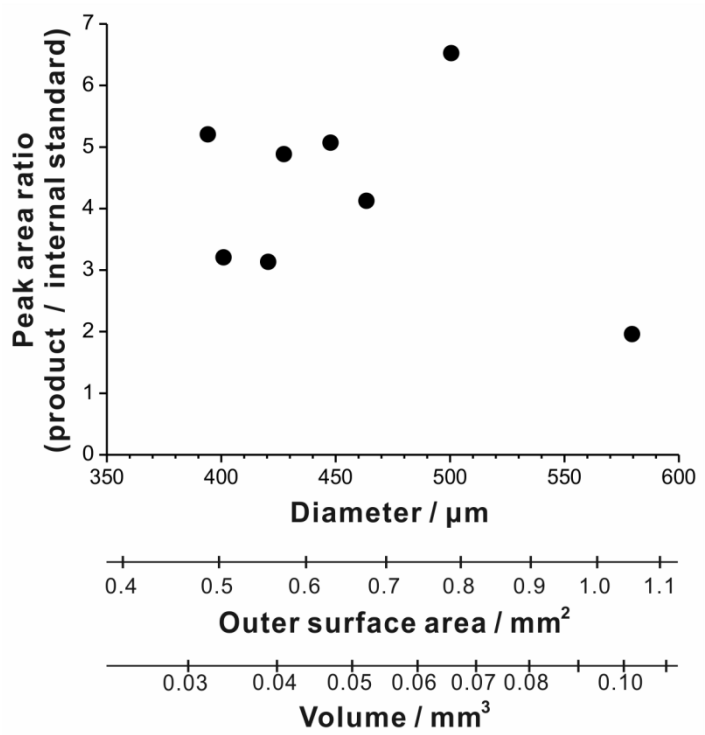

Fig. S5 Dependence of the relative yield of the enzymatic reaction (after 126 min) on physical dimensions (diameter, outer surface area, volume) of the analysed lipase microbeads according to the results of single-microbead assay (**Figs. 6** and **S4**). Peak areas were measured based on the EICs (analyte *m/z* 43 ± 0.5 u e^{-1}, internal standard *m/z*, 93 ± 0.5 u e^{-1}).

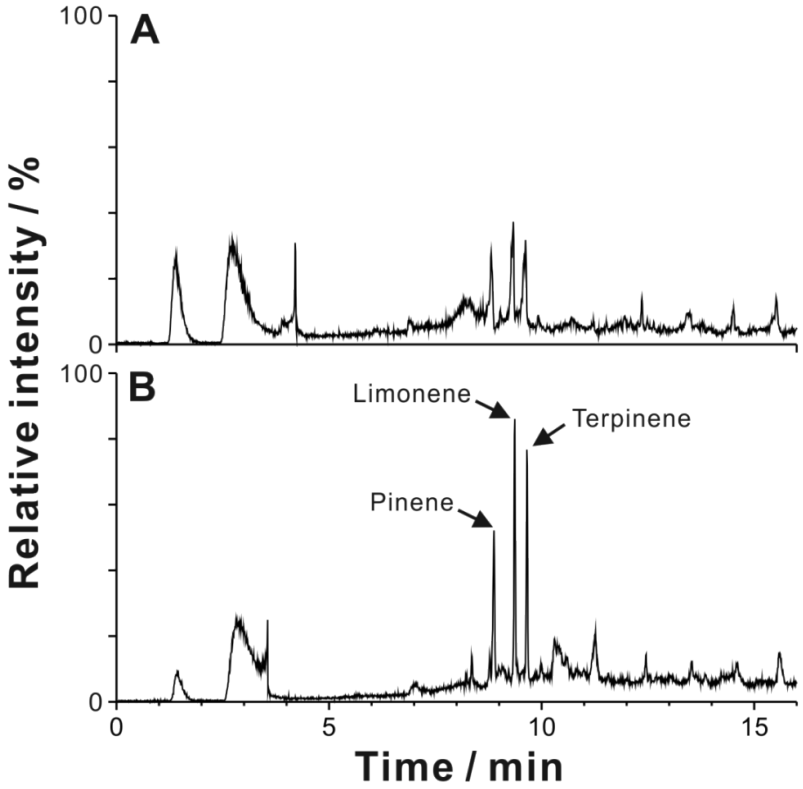

Fig. S6 Monitoring extraction of the lemon peel sample in real time. Extraction solvent: 5 mL acetonitrile. Temperature: 298 K. Shaking speed: 20 rpm. Internal standard: 10^{-5} M thymol. (A) EIC at *m/z* 93 ± 0.5 u e^{-1} obtained at time "zero" (right after inserting the sample to the extraction solvent), and (B) EIC at *m/z* 93 ± 0.5 u e^{-1} obtained at a later stage of extraction (140 min). These mass spectra are from the same experiment as the one illustrated in **Fig. 7A**.