Supporting information

Microdroplet chemical reactor prototype based on multiplexed electrospray

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Experimental procedures and reaction conditions

Chemicals and Reagents

The chemicals were bought commercially and were used with no additional processing or purification. Phenylhydrazine (97%) and cyclohexanone (99.8%) were purchased from Thermo Scientific Chemicals (Haverhill, MA, USA). HPLC-gradient grade methanol was purchased from Chimmed (Russia), acetonitrile grade "0" was purchased from Cryochrom (Russia), formic acid (98 %) was purchased from Sigma-Aldrich (Switzerland), 18.2 MΩ·cm water was from a Milli-Q water purification system.

Analysis of reaction mixtures

Analyses were performed by means of high performance liquid chromatography – high resolution mass spectrometry with electrospray ionization (HPLC-ESI-HRMS). Instrument setup for all the analyses consisted of a liquid chromatograph LC-30 Nexera (Shimadzu, Kyoto, Japan) including DGU-A5 vacuum degasser unit, two LC-30AD pumps, CTO-20A column oven, SIL-30AC autosampler, and CBM-20A system controller, coupled with a Q Exactive Plus hybrid mass spectrometer (Thermo Scientific, Waltham, USA) with an Orbitrap mass analyzer.

Chromatograms were recorded using Thermo Xcalibur 3.1 software. A Nucleodur PFP column from Macherey-Nagel (2·150 mm, 1.8 µm, Macherey-Nagel, German) was used to obtain chromatographic separation of the samples. Mobile phase was formic acid (0.1% v/v) in ultrapure water (B) and acetonitrile (A). Gradient elution was set as follows: 20% A from 0 to 1 min, then linear gradient from 20 % to 100 % A in 16 min, then 100 % A held for 1 min, in 1 min the percentage of acetonitrile returns to initial conditions and the 20 % A is held for 5 min (from 19 to 24 min) to ensure the correct equilibration of the column at the initial conditions. The flow rate was set at 450 µL/min. Global run time was 24 min. The column temperature was set at 40° C. Sample injection volume was 5 µL.

The HRMS system operated in the positive ionization mode. The optimal parameters of the ion source were used to ensure: spray voltage 3.5 kV; sheath, auxiliary and sweep gas (N_2) flow rates 30, 8 and 3 arb. units, respectively; desolvation capillary temperature 320 ºC; auxiliary gas heater temperature 120 ºC; S-lens RF level 55 arb. units. Mass spectrometer operated in full-scan mode in the range of 80-330 m/z (ESI+), with a resolution of 70000. An automatic gain control mode (AGC) with the target quadrupole ion trap (C-Trap) filling value of 10^6 was used. Tandem mass spectrometry (MS/MS) experiments were automatically performed in the range of *m/z* 50-300 (ESI+), using the automatic dependent scan function. Collision energy was set at 30 (arbitrary units) for all the MS acquisitions. Xcalibur 3.1 software was used both for acquisition and for elaboration and calculation.

Mass scale calibration was performed using the Calmix calibration mixture (Thermo Scientific, Waltham, USA). N,N-Dimethylformamide and acetone were used as additional standards to increase the accuracy of *m/z* measurements in the low mass region (*m/z* 50-100).

Reaction conditions

The reaction mixture initial composition was consistent across all experiments: a methanol solution containing an equal amount of phenylhydrazine (**1**) and cyclohexanone (**2**) (C=0.0002 mol/L). Further, the conditions varied depending on the type of experiment. The following reaction conditions were used:

a) Study of the reaction of phenylhydrazine with cyclohexanone in methanol under conditions of contact of the reaction mixture with air.

The reaction mixture (60 mL) was kept at room temperature in a sealed flask on a magnetic stirrer for 480 minutes. Immediately after preparation of the reaction mixture and thereafter every 30 minutes a 1 mL aliquot of the reaction mixture was taken and subjected to HPLC-ESI-HRMS analysis according to the above procedure. These measurements were repeated three times. The results are

presented in “Results and Discussion” in the related subsection.

b) Nitrogen purging in the reaction of phenylhydrazine and cyclohexanone.

The reaction mixture (60 mL) was prepared and incubated at room temperature in a dark glass flask under constant nitrogen flow through the reaction mixture for 1380 minutes. Immediately after preparation of the reaction mixture and at regular intervals, a 1 mL aliquot of the reaction mixture was taken and analyzed by HPLC-ESI-HRMS according to the above procedure. These measurements were repeated three times. The results are presented in “Results and Discussion” in the related subsection.

c) Reaction of phenylhydrazine and cyclohexanone in the MCR prototype.

The reaction mixture (80 ml) was prepared and sprayed at room temperature in the MCR prototype (spraying started 4 minutes after preparation of the reaction mixture; the duration of spraying is listed in Table 2). For the series of experiments no. 1 and no. 3 (Table 2), the sample collector was washed several times with the accumulated liquid. An aliquot was taken and further diluted 10 times with methanol. Then, 1 ml of the resulting solution was taken and subjected to HPLC-ESI-HRMS analysis. This procedure was repeated three times for experiments series no. 1 and one times for experiments series no. 3. For the experiments series no. 2, an aliquot of the accumulated liquid was not taken because of its small volume. To extract the accumulated substances, the sample collector was washed twice with methanol, resulting in a total volume of 9 ml. The washing solution was then brought to a volume of 10 ml with methanol, after which 1 ml of the obtained solution was analyzed by HPLC-ESI-HRMS. This procedure was repeated three times. The results are presented in “Results and Discussion” in the related subsection.

Figures and Tables

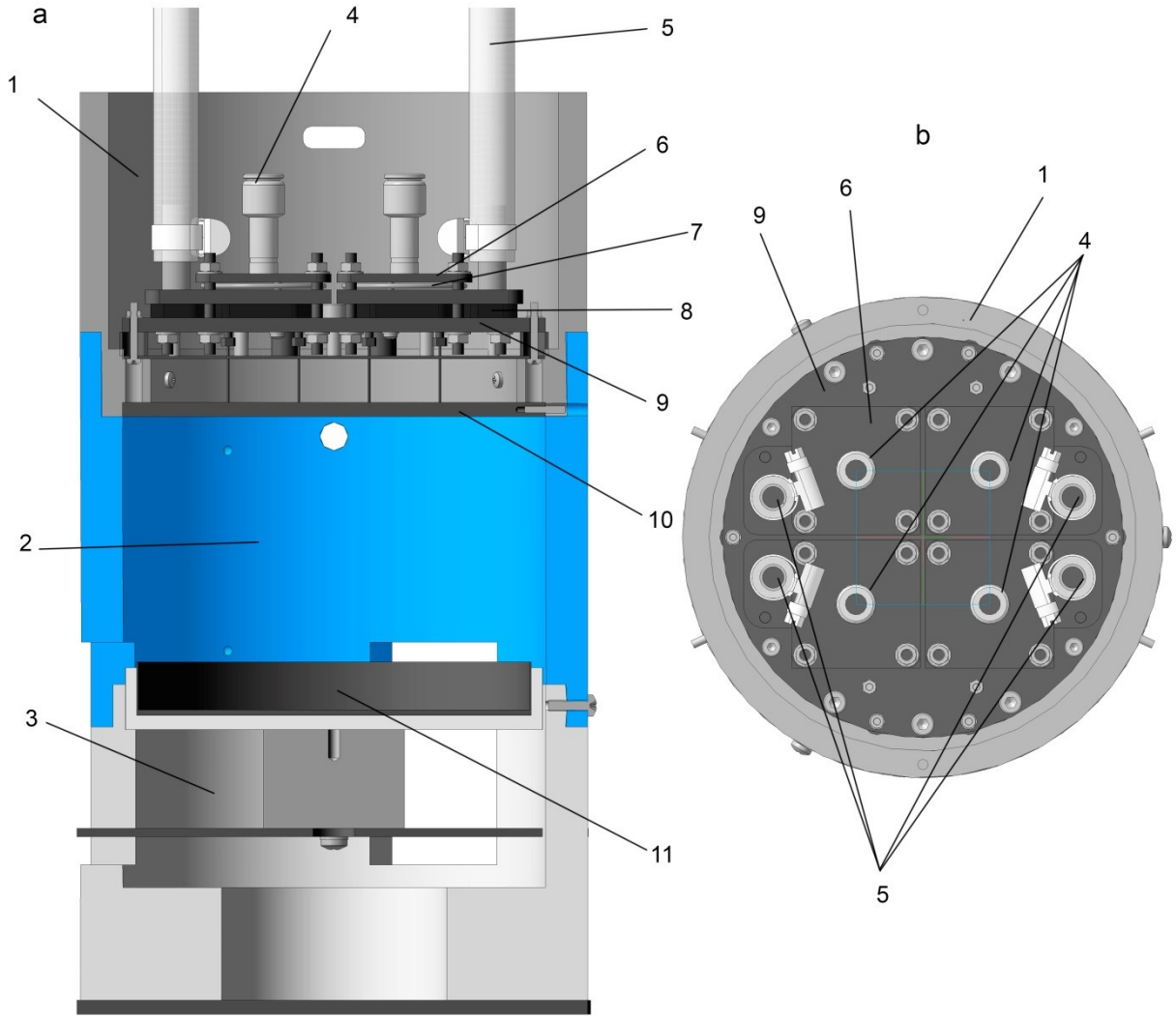

Fig. S1. Schematic illustration of the reactor device. Cross section of the reactor device (a) and top view (b): 1 - the multicapillary pneumoelectrospray system, 2 – the reaction chamber, 3 – the sample collection system, 4 – the liquid inlet fitting, 5 – the gas inlet, 6 - the sprayed liquid input module, 7 – the liquid seal, 8 – the gas seal, 9 - the nebulizing gas inlet module,10 – the transport counter electrode, 11 - the collector of the sample collection system

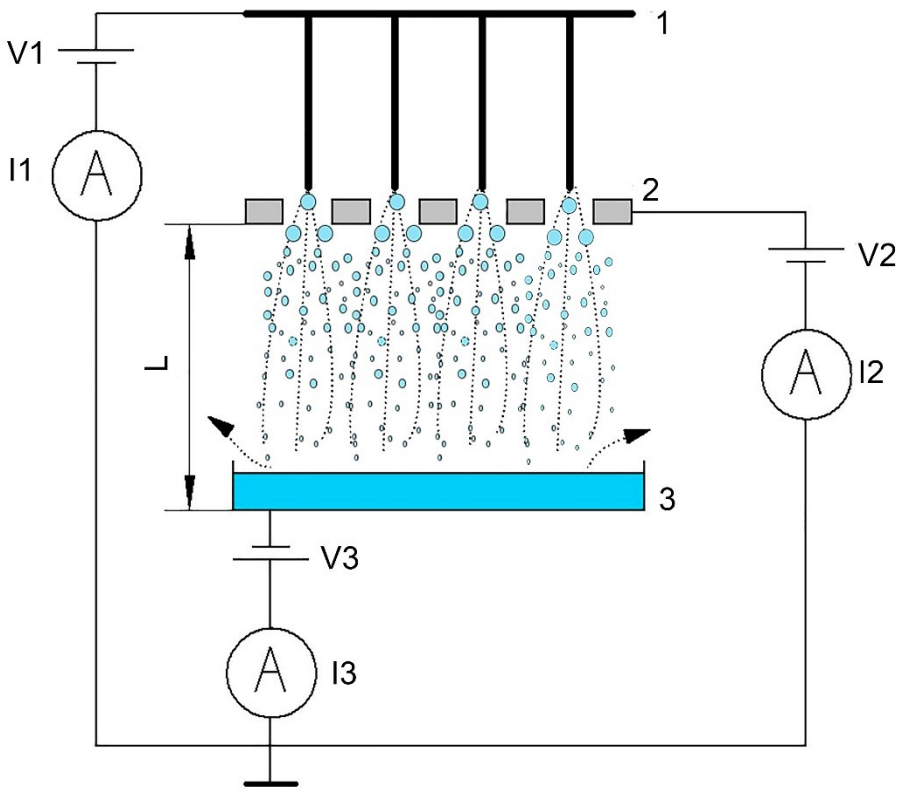

Fig. S2. Electrical scheme of power supply of the reactor device included in the MCR prototype. 1- capillaries of the multicapillary pneumoelectrospray system, 2 – the transport counter electrode, 3 – the collector of the sample collection system. V1 - unit of electric potential supply on the multicapillary pneumoelectrospray system (supply electric potential U1), V2 - unit of electric potential supply on the counter electrode (supply electric potential U2), V3 - unit of electric potential supply on the collector of the sample collection system (supply electric potential U3). I1 – the total current; I2 – the current flowing through the transport counter electrode; I3 - the current flowing through the collector of the sample collection system. The dotted line shows the nebulizing gas flow.

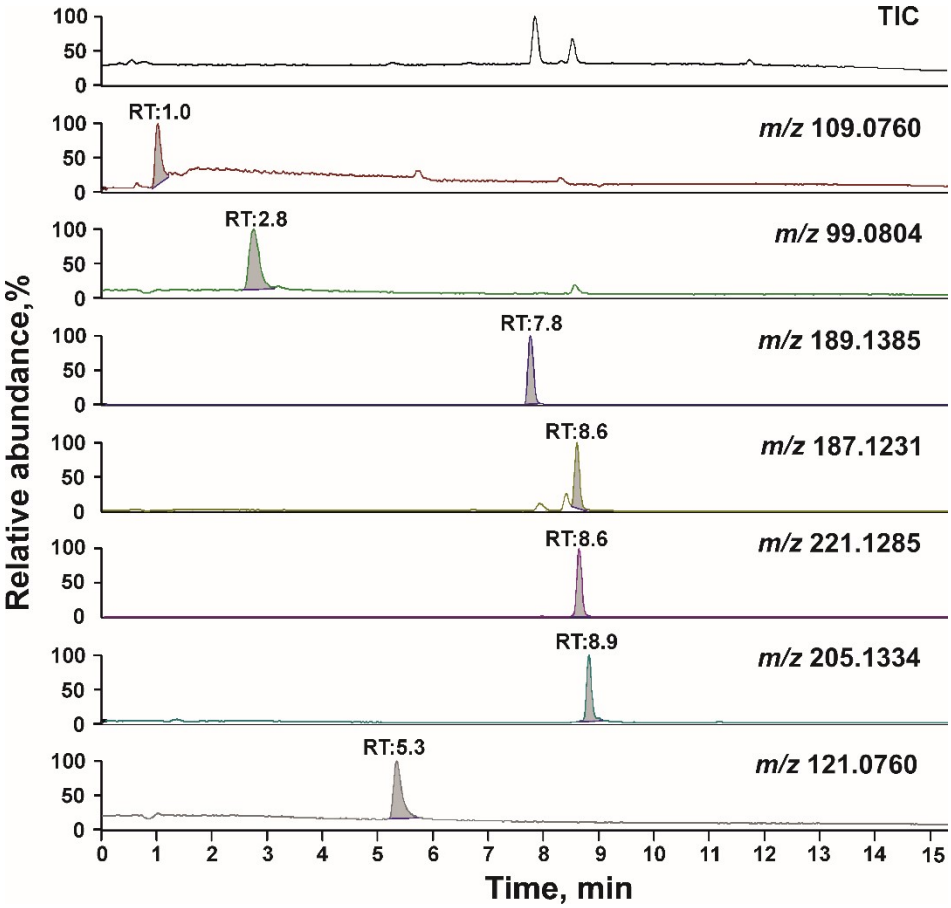

Fig. S3. Total ion chromatogram (TIC) and selected ion mass chromatograms of reaction products of phenylhydrazine with cyclohexanone. Reaction time – 120 min.

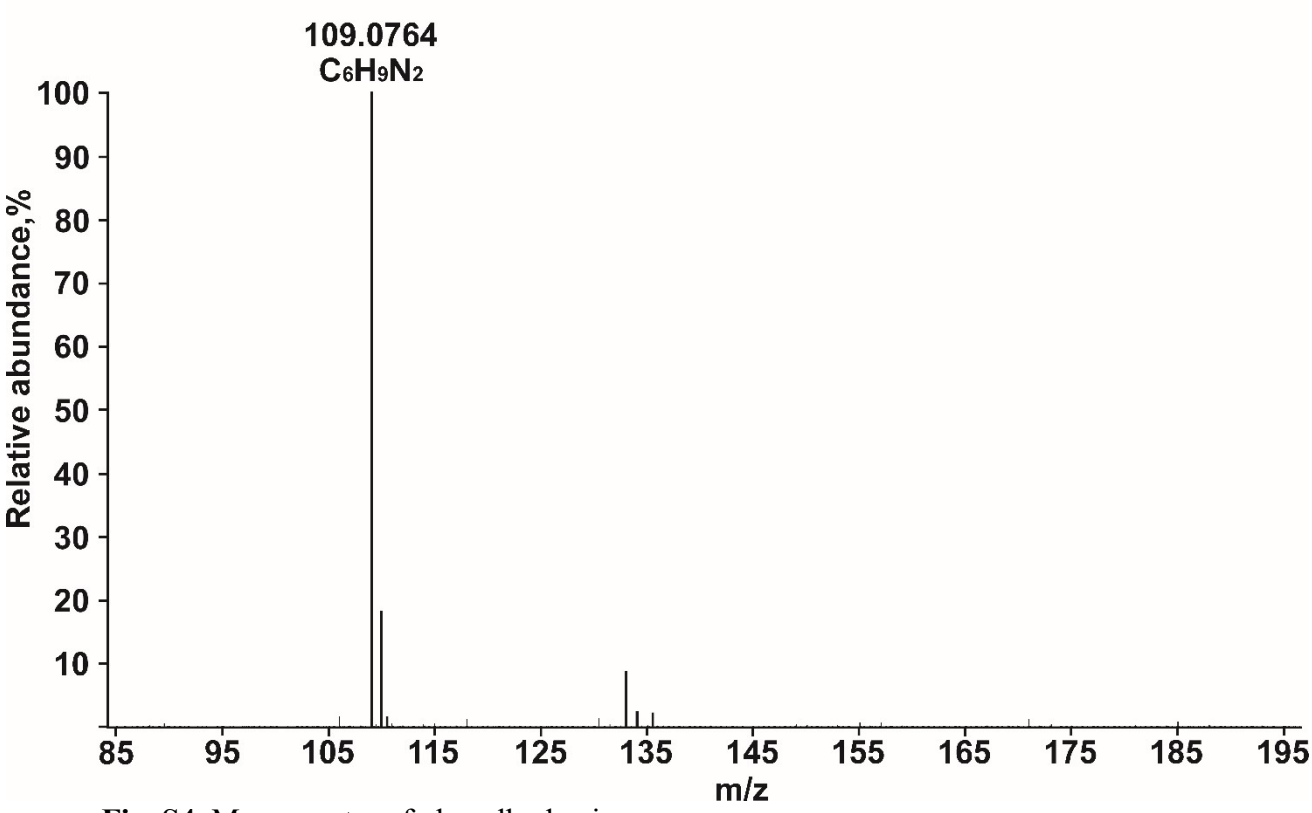

Fig. S4. Mass spectra of phenylhydrazine.

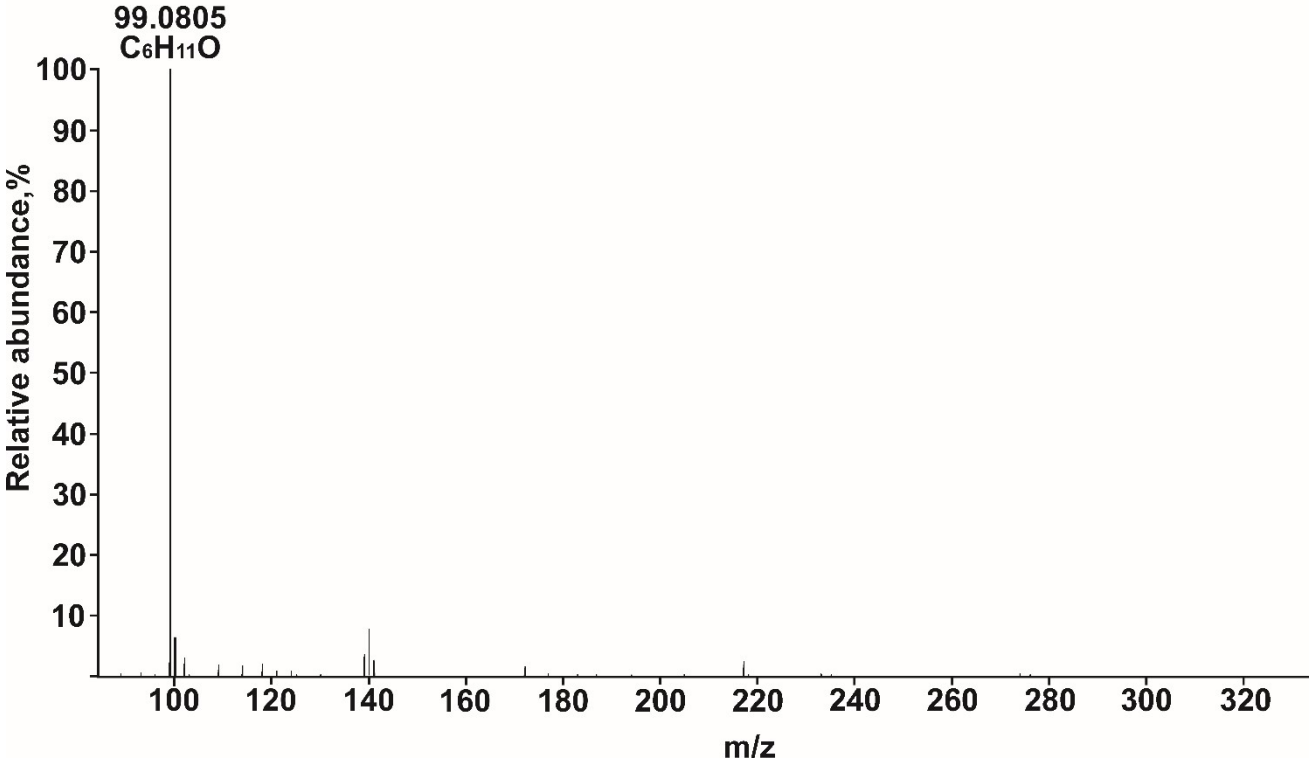

Fig. S5. Mass spectra of cyclohexanone.

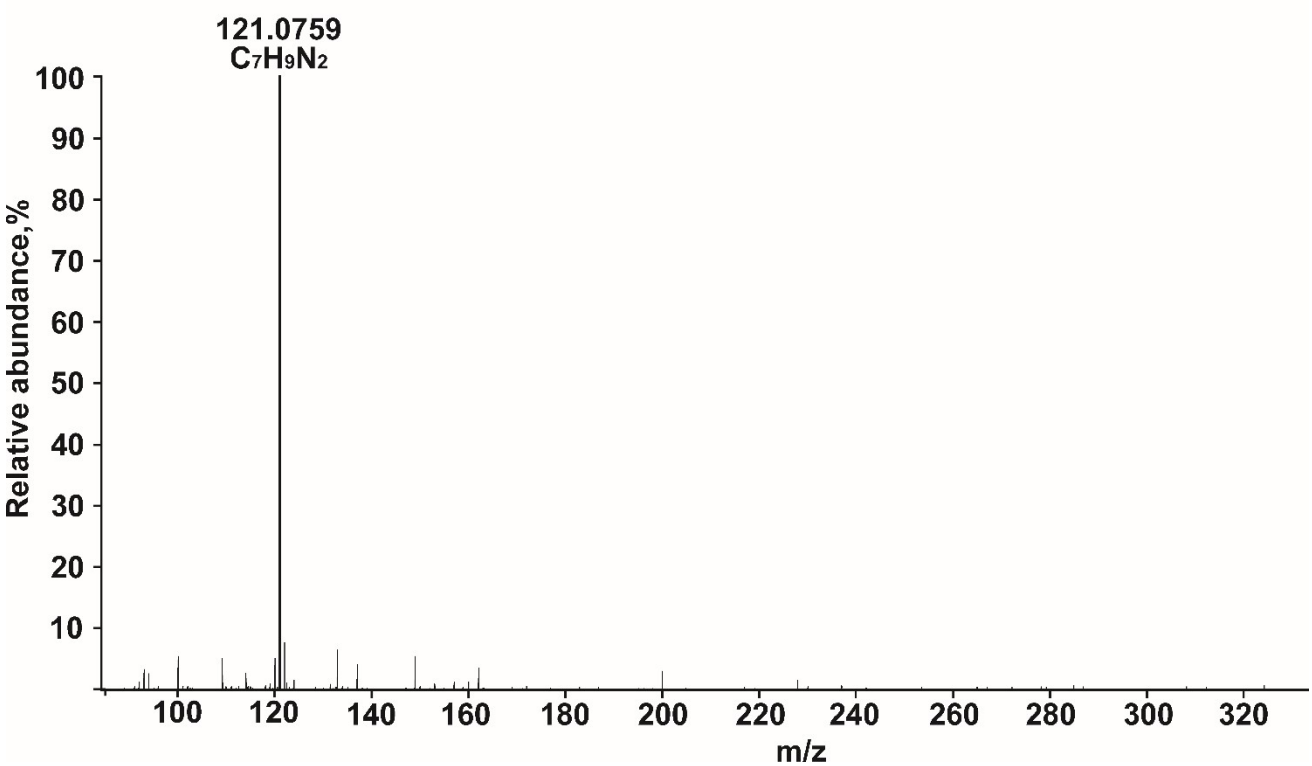

Fig. S6. Mass spectra of formaldehyde phenylhydrazone.

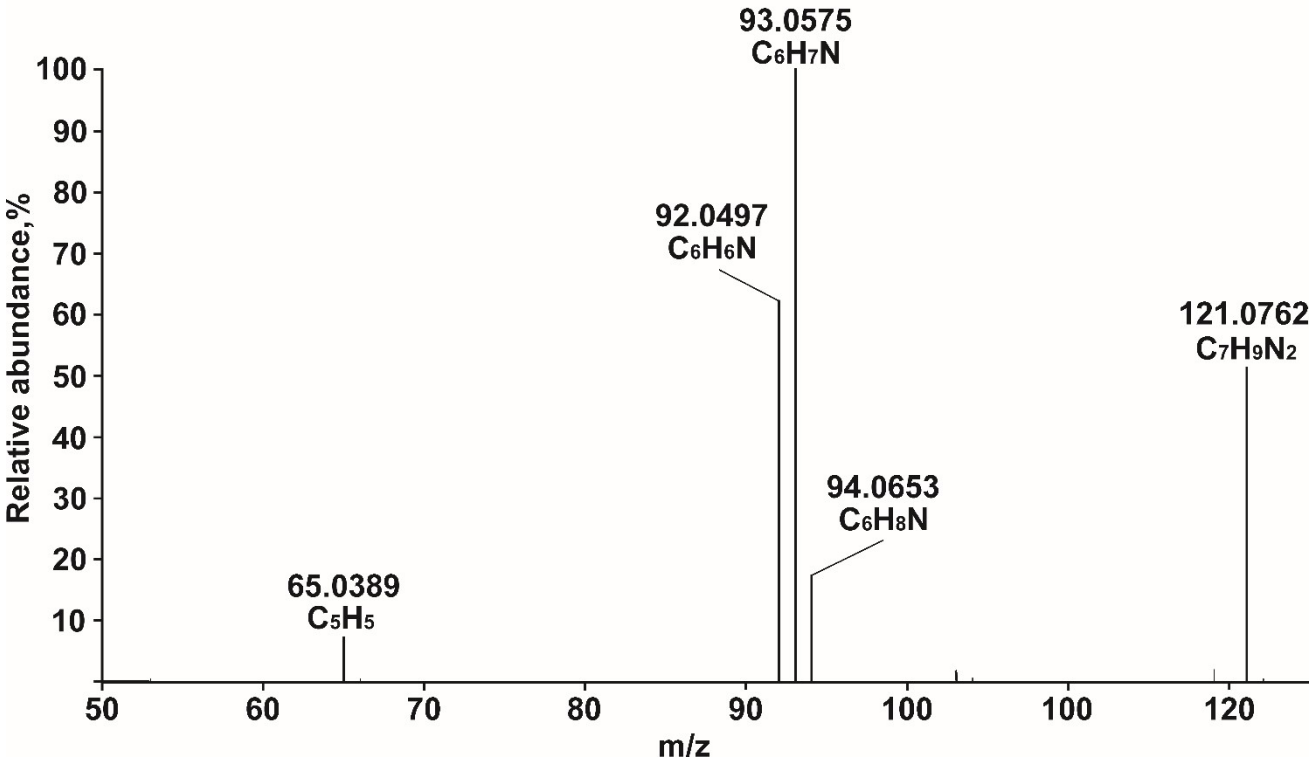

Fig. S7. MS/MS spectra obtained from the $[M+H]^+$ ion fragmentation of formaldehyde phenylhydrazone.

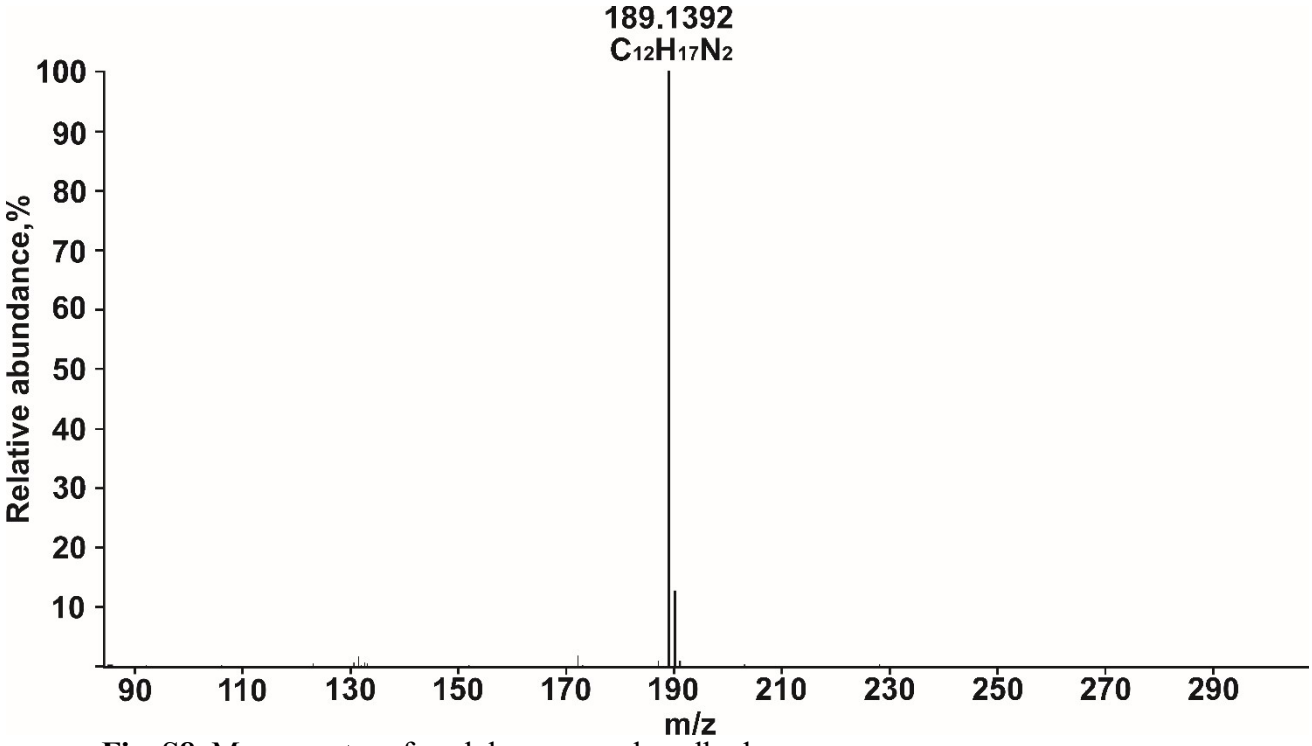

Fig. S8. Mass spectra of cyclohexanone phenylhydrazone.

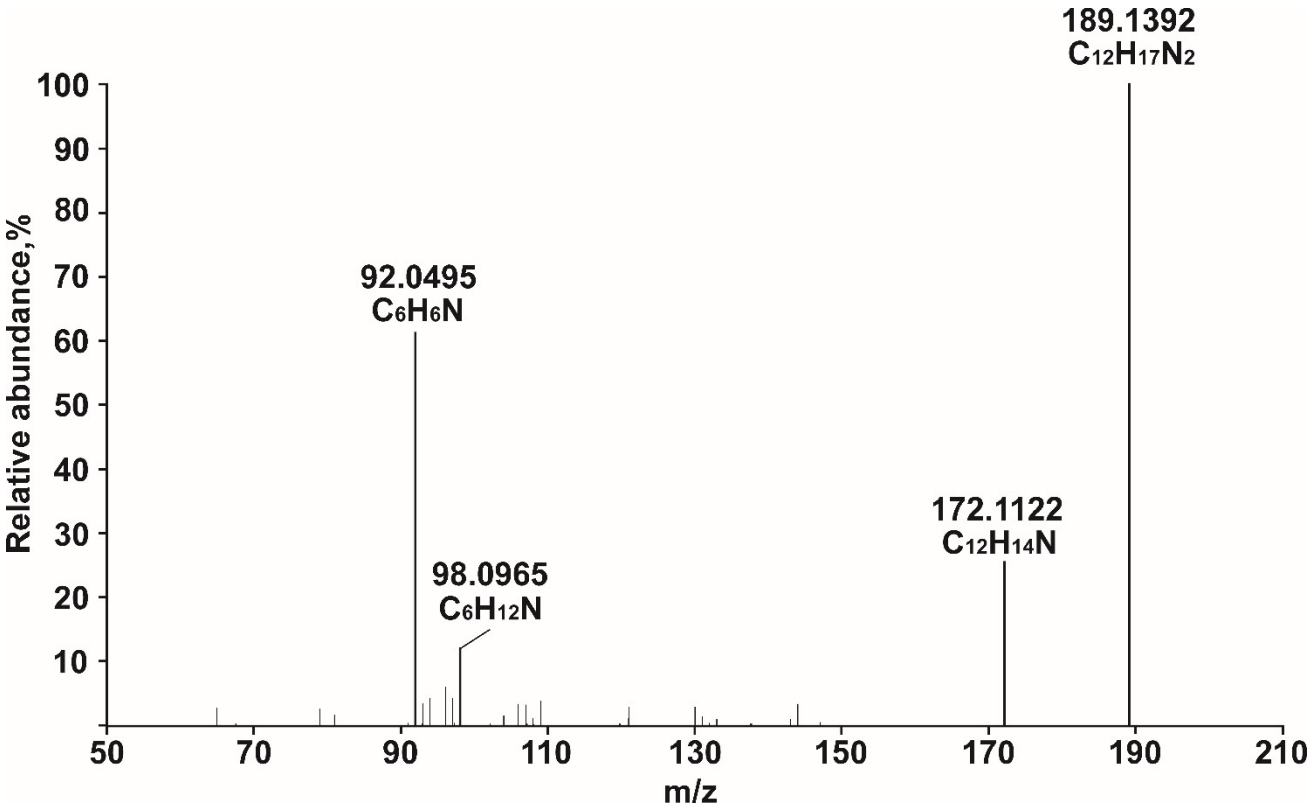

Fig. S9. MS/MS spectra obtained from the $[M+H]^+$ ion fragmentation of cyclohexanone phenylhydrazone.

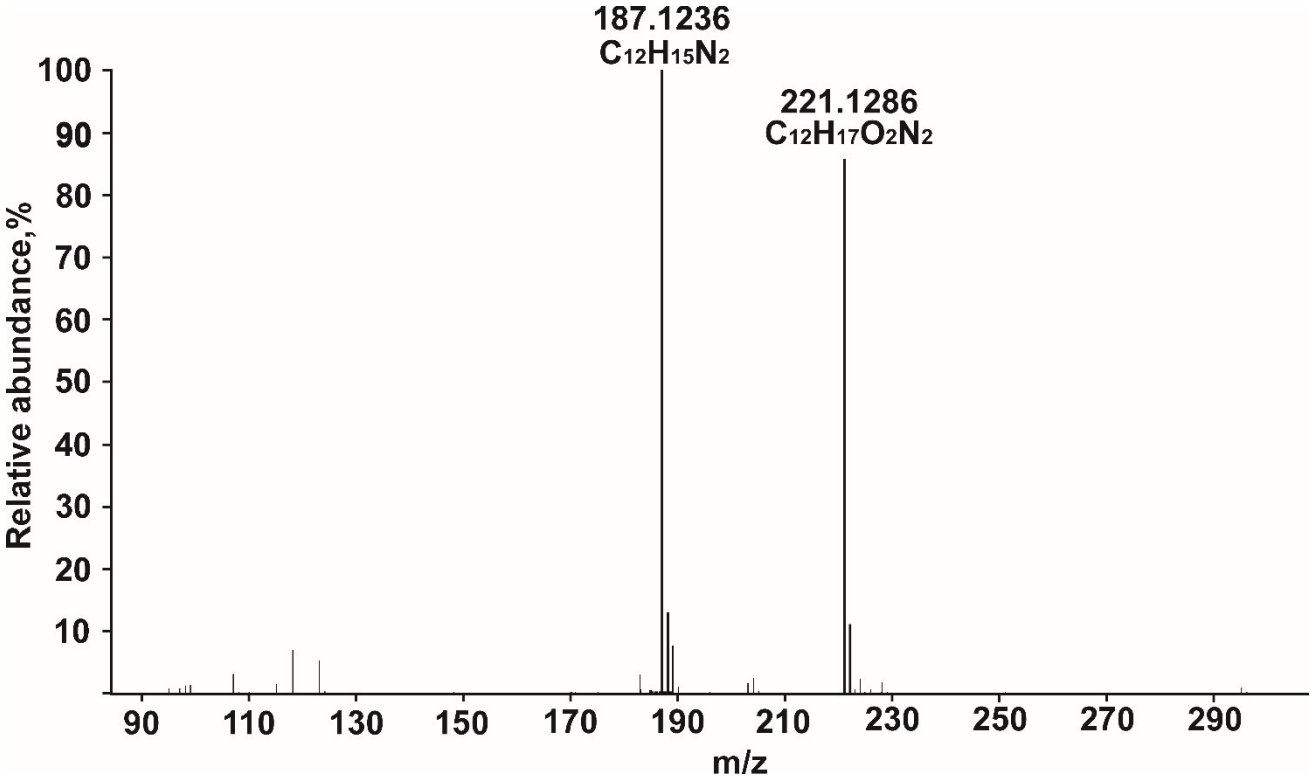

Fig. S10. Mass spectra of 1-(1-hydroperoxycyclohexyl)-2- phenyldiazene.

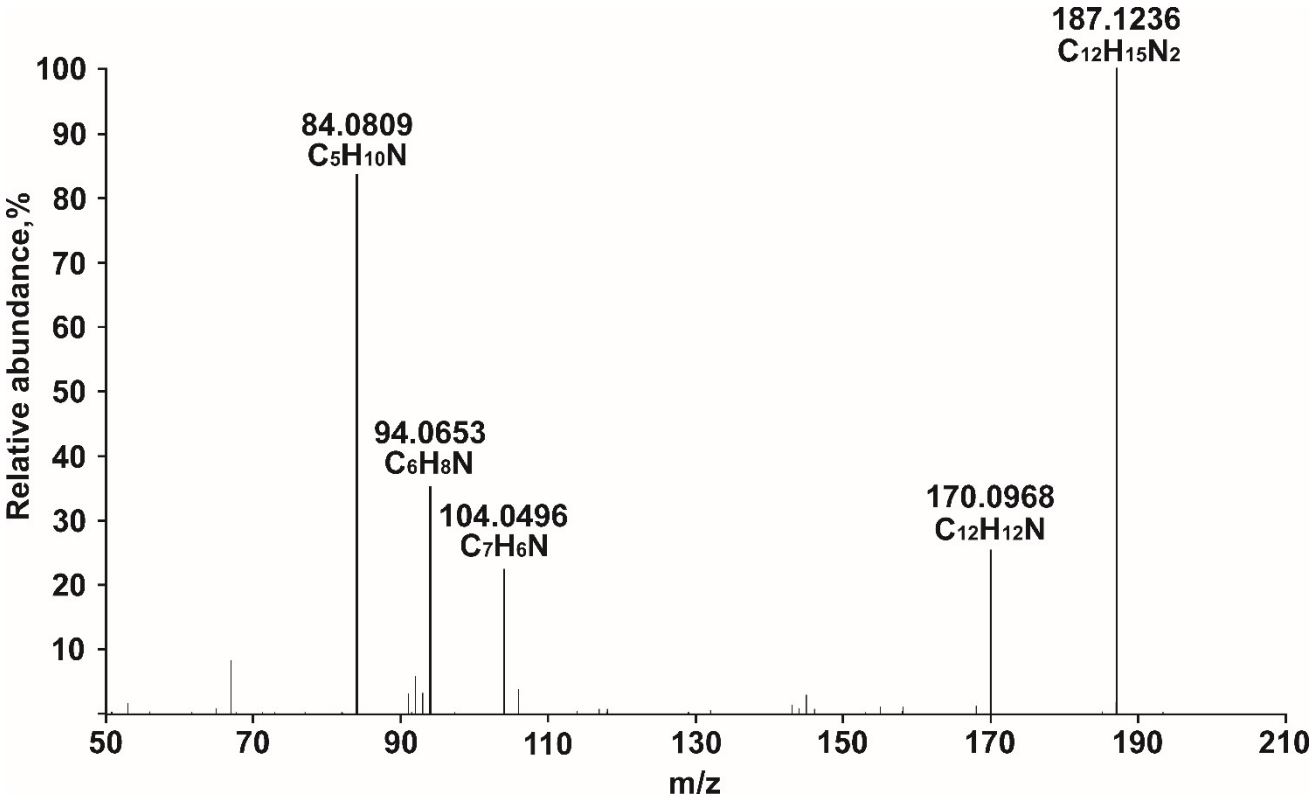

Fig. S11. MS/MS spectra obtained from the ion of m/z 187.1236 fragmentation.

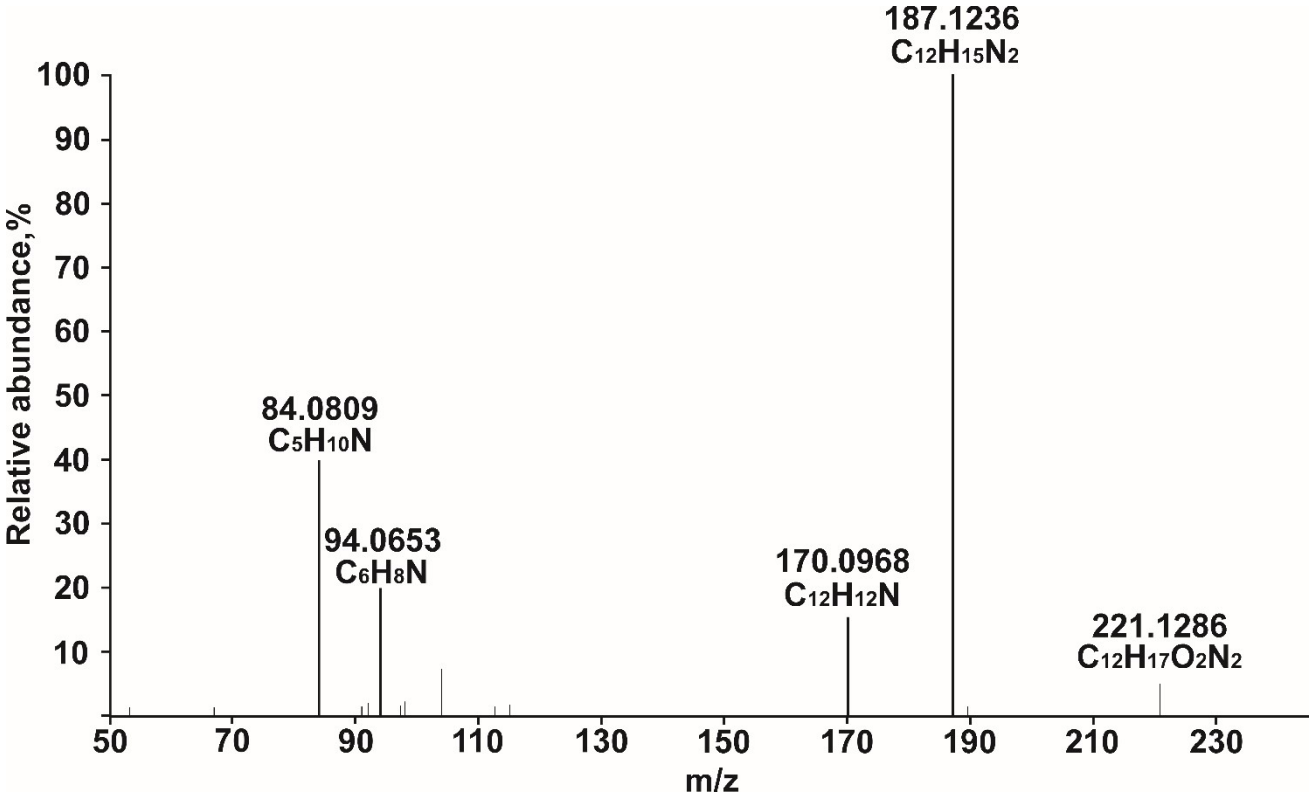

Fig. S12. MS/MS spectra obtained from the $[M+H]^+$ ion fragmentation of 1-(1-hydroperoxycyclohexyl)-2- phenyldiazene.

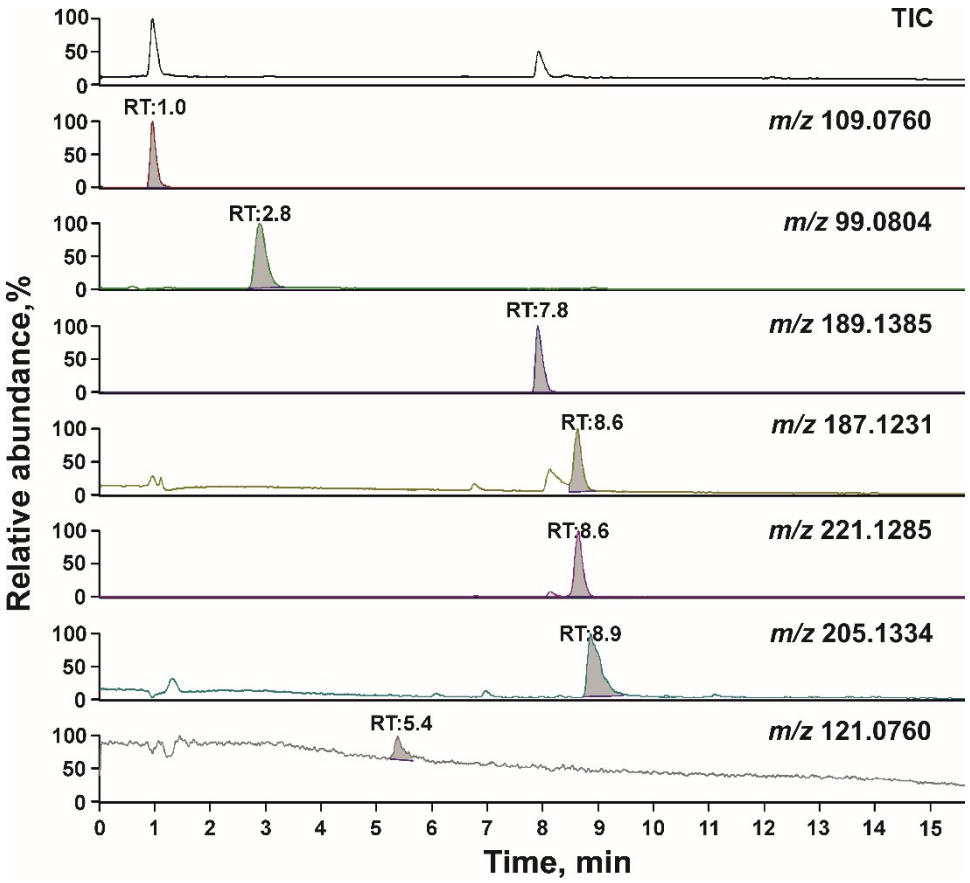

Fig. S13. Total ion chromatogram (TIC) and selected ion mass chromatograms of reaction products of phenylhydrazine with cyclohexanone. Reaction time – 560 min.

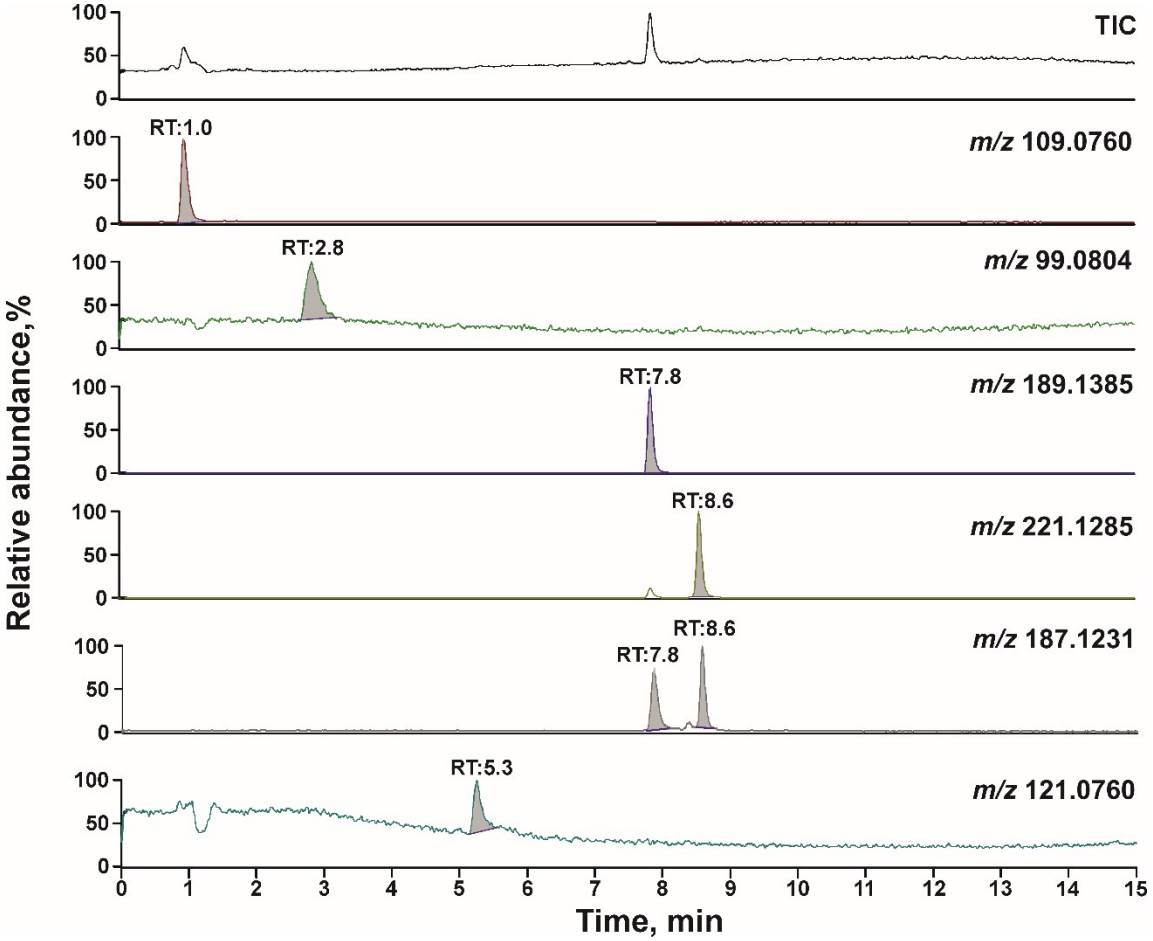

Fig. S14. Total ion chromatogram (TIC) and selected ion mass chromatograms of reaction products of phenylhydrazine with cyclohexanone. Reaction in the MCR prototype, one of the measurements for the experiment series No. 1.

Table S1. The general equations of approximating curves, values of equations coefficients and adjusted R^2 values

No. of curve	General equation	y_0	b	a	adjusted R^2
1	$y=y_0+bx+ax^2$	0.4486± 0.46022	0.10956± 0.03408	$-3.2806\times10^{-5}\pm 2.95333\times10^{-5}$	0.99985
2		0.01289± 0.01125	0.00736± 0.00587	$1.58804\times10^{-5}\pm 6.27884\times10^{-6}$	0.97861
3	$y=y_0+bx$	47.13064± 1.46648	-0.0029± 0.01191	-	-0.18064