Supporting Information:

1) Monitoring batch reaction:
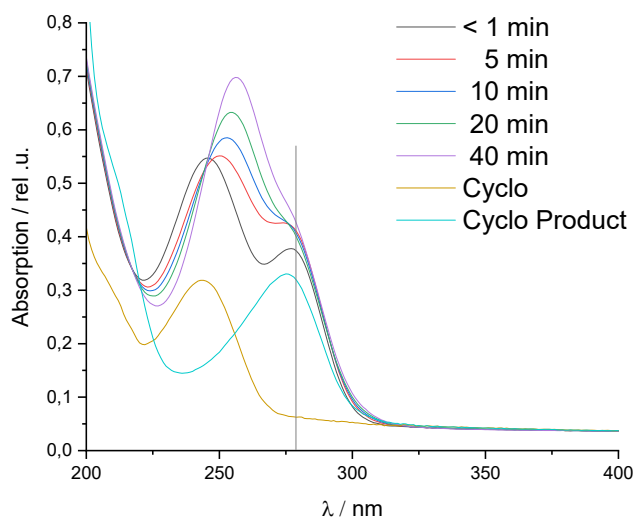
UV-VIS batch reaction

Fig. S.1 Selection of UV-VIS spectra of the time-dependent monitoring of a reaction mixture (100 µM Pip, Fal and Cyclo), reaction time 0 min = adding Pip to Fal and Cyclo mixture + reference spectra of Cyclo and its product **5**-Cylco, grey line at λ = 279 nm, measurement was repeated at only 279 nm with higher measurement frequency of 25 Hz for fig. 3

NMR batch reaction

As acidic conditions are difficult to measure in non-aqueous media, a series of solvents with increasing acid content were investigated. Six seldom have been studied for this purpose for the reaction with 1,3-cyclohexandione, and two for the reaction with Acetylacetone, as described in detail in experimental part section A reaction combination 1) to 8).

The resulting amount of product at sampling time was calculated from the integral of the triplet signal at 2.80 ppm (for 1,3-Cyclohexandione and for Acetylacetone: $N_{product}$ = 2) (NMR-spectra are provided in the supporting info). This signal is calculated in relation to the signal of the hydrogens ($N_{dioxane}$ = 8) of the internal standard, dioxane, at 3.66 ppm by the following equation:

$$c_{Produkt} = \frac{\int I_{produkt}(3.66\ ppm)}{\int I_{dioxane}(2.80\ ppm)} \cdot \frac{N_{dioxane}(H\ singlet, 3.66\ ppm)}{N_{product}(H\ triplet, 2.80\ ppm)} \cdot c_{dioxane}$$
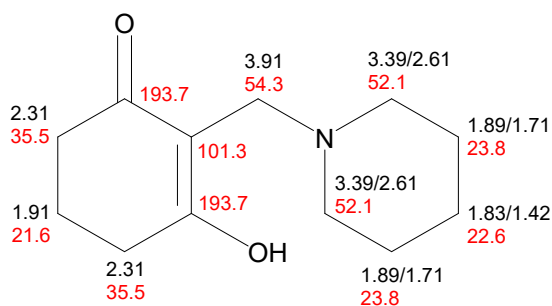
With c_x as the concentration, $\int I_x$ as the integral at the specific hydrogen signals, and N_x as the number of hydrogen atoms causing the signal.

NMR Spectra of 3-hydroxy-2-(piperidin-1-ylmethyl)cyclohex-2-en-1-one

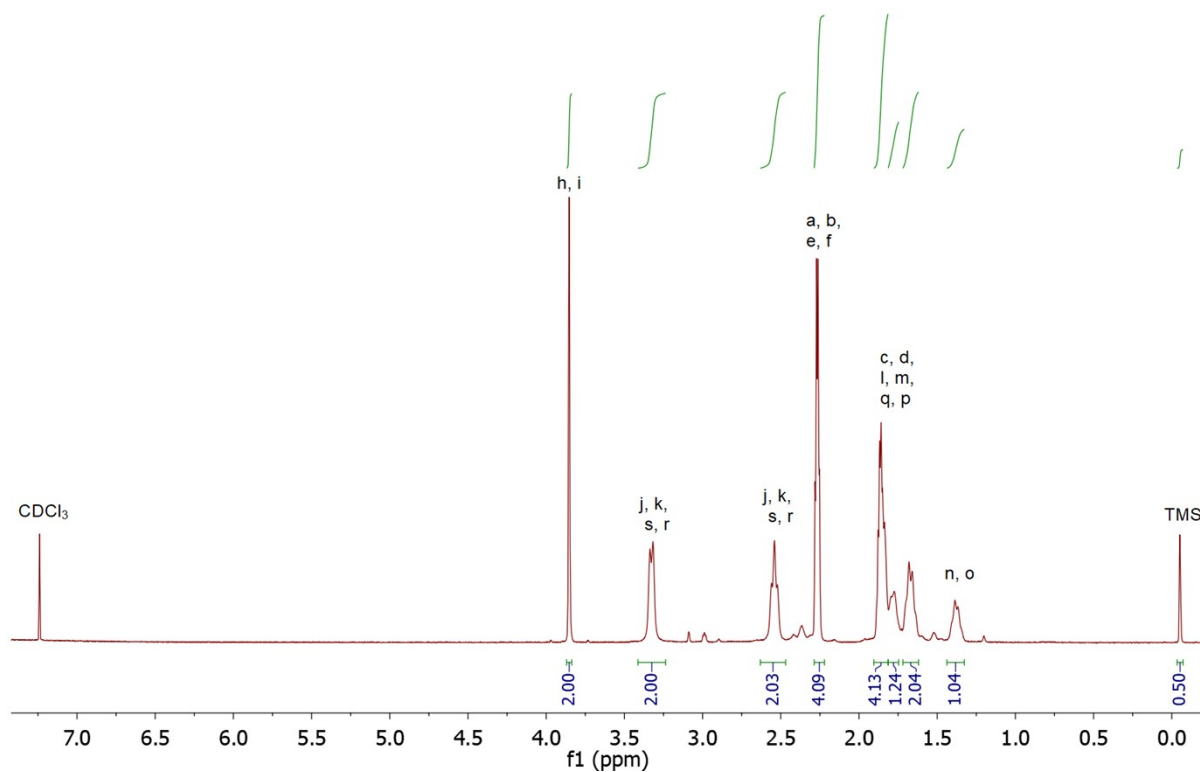Labelling: Hydrogen: black, Carbon: red
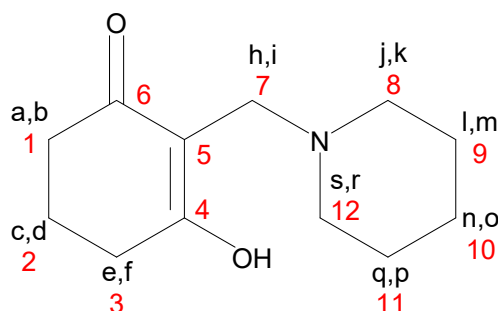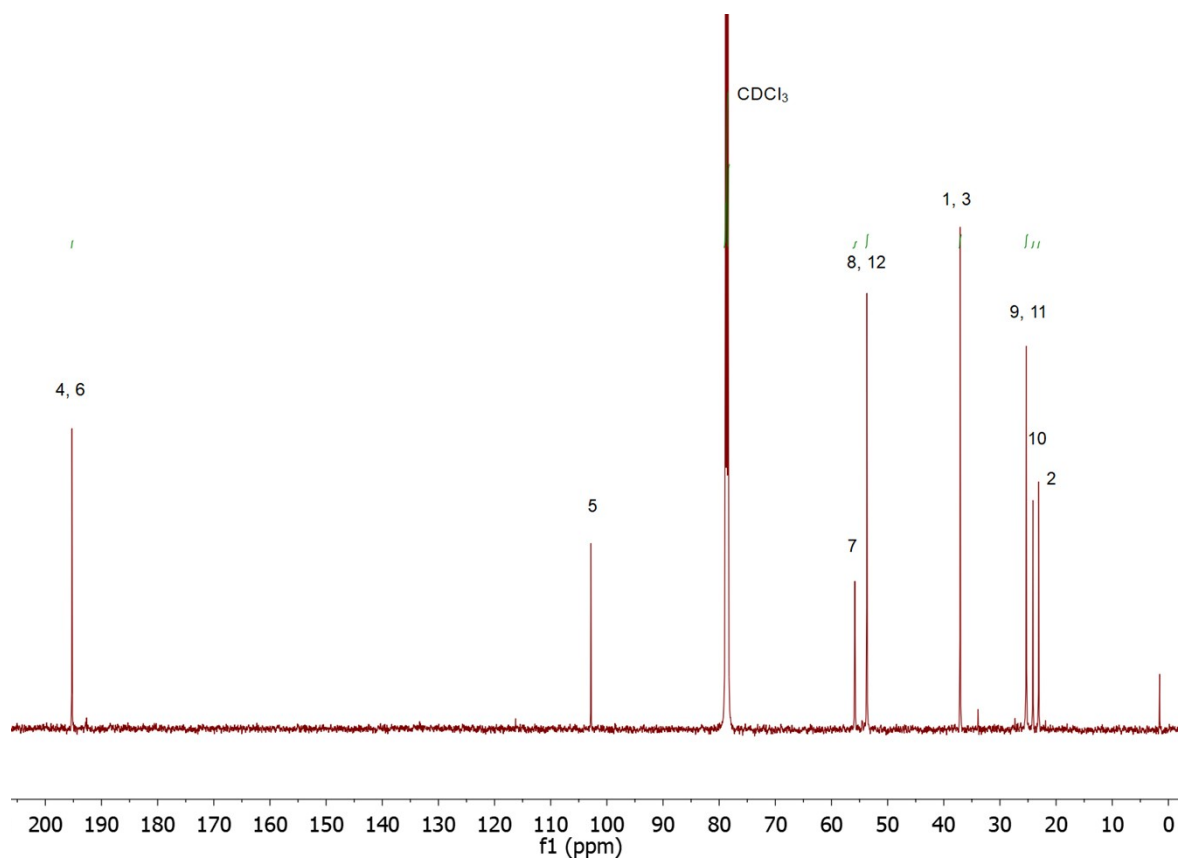

^{1}H, ^{13}C-shifts in CDCl$_3$ + TMS:

^{1}H-spectra in CDCl$_3$ + TMS:

^{1}H NMR (600 MHz, C$_D$Cl3, 25 °C): δ = 3.85 (s, 2H), 3.34 (d, *J* = 12 Hz, 2H), 2.52 (t, *J* = 12 Hz, 2H), 2.26 (t, *J* = 6 Hz, 4H), 1.84-1.87 (m, 4H), 1.85 (m, 1H), 1.64-1.66 (m, 2H), 1.37-1.41 (m, 1H)).
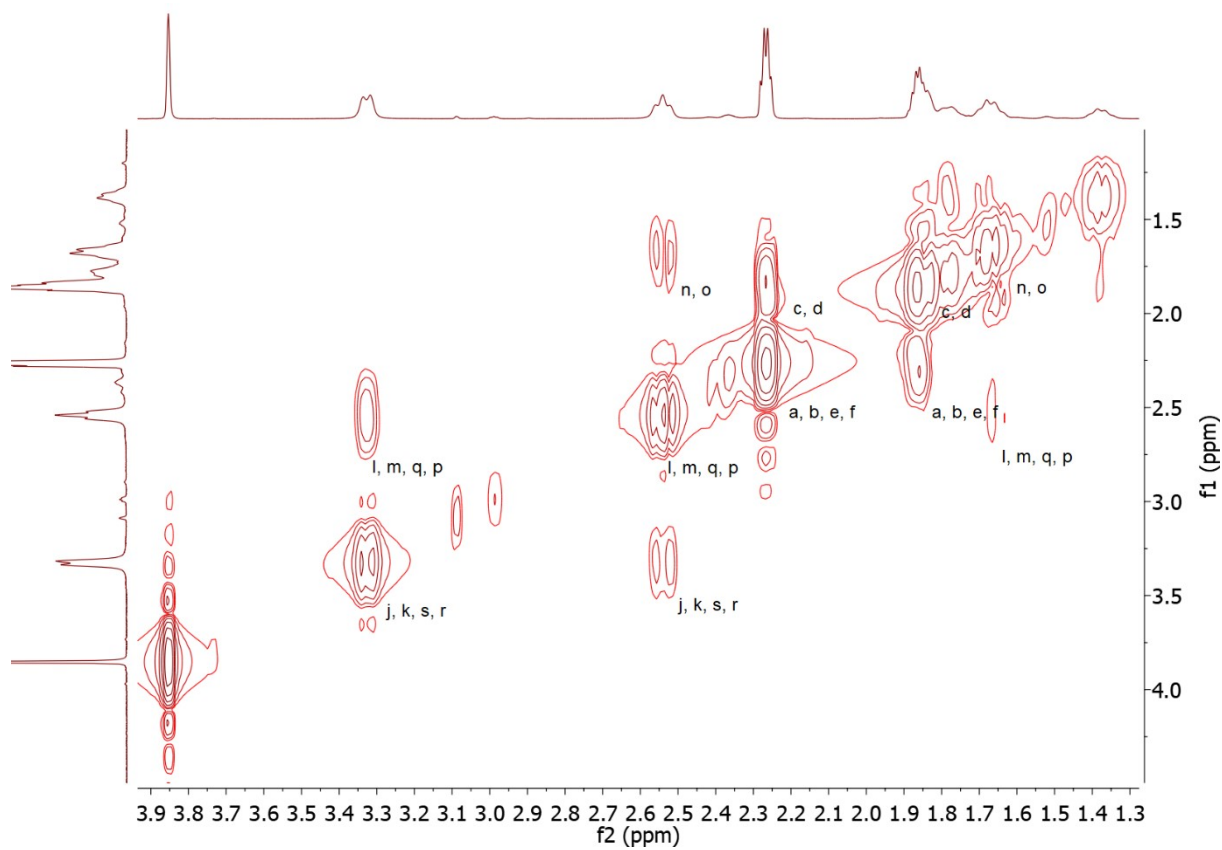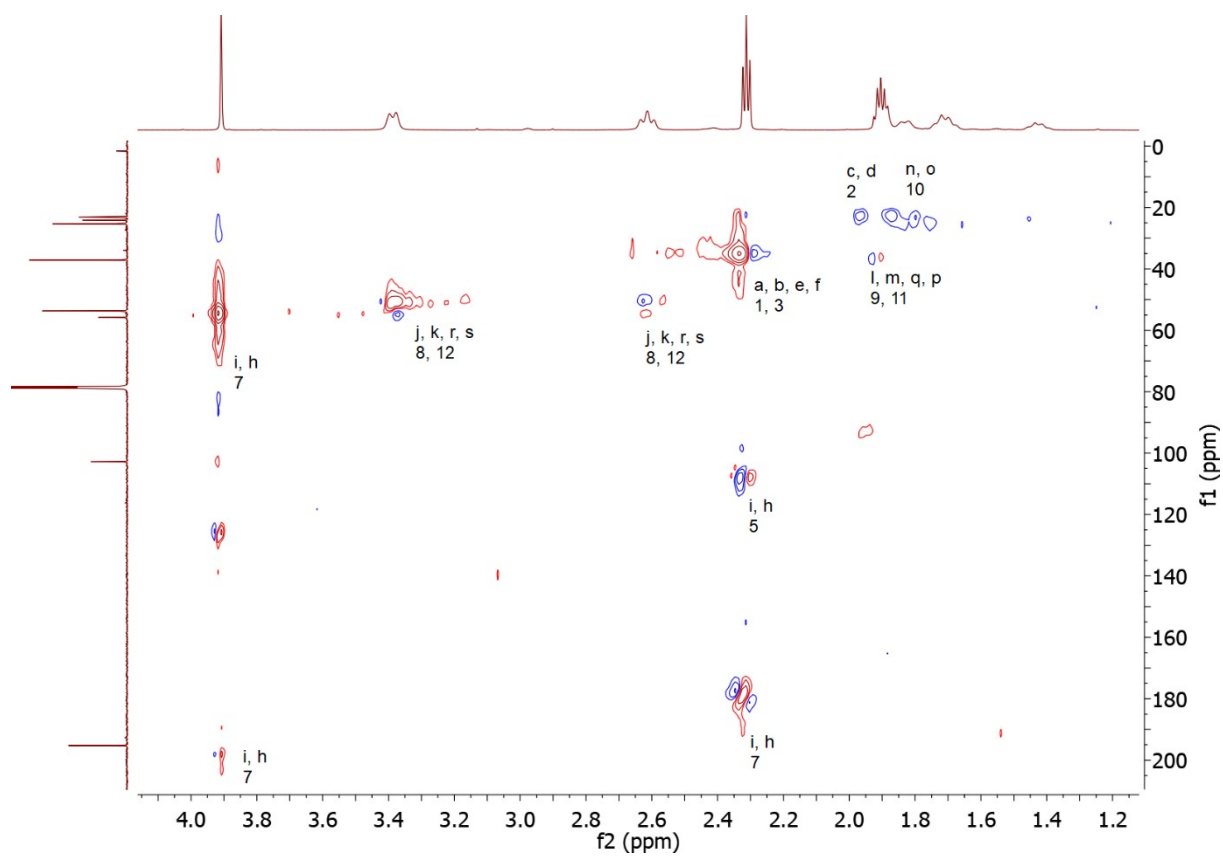
^{13}C-spectrum in CDCl$_3$ + TMS:

^{13}C NMR (100 Mhz, CDCl$_3$, 25 °C): δ = 195.6, 102.5, 57.0, 55.0, 37.5, 26.0, 25.0, 24.0
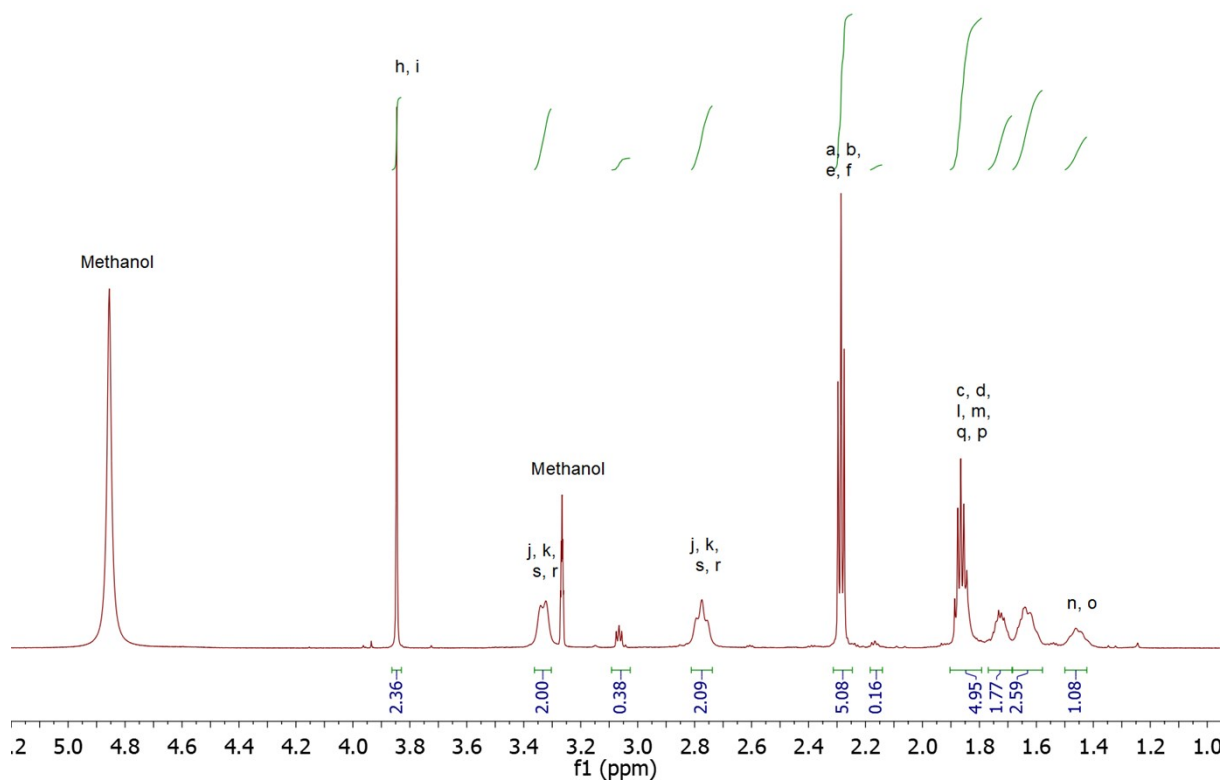

COSY-spectrum in CDCl$_3$ + TMS:
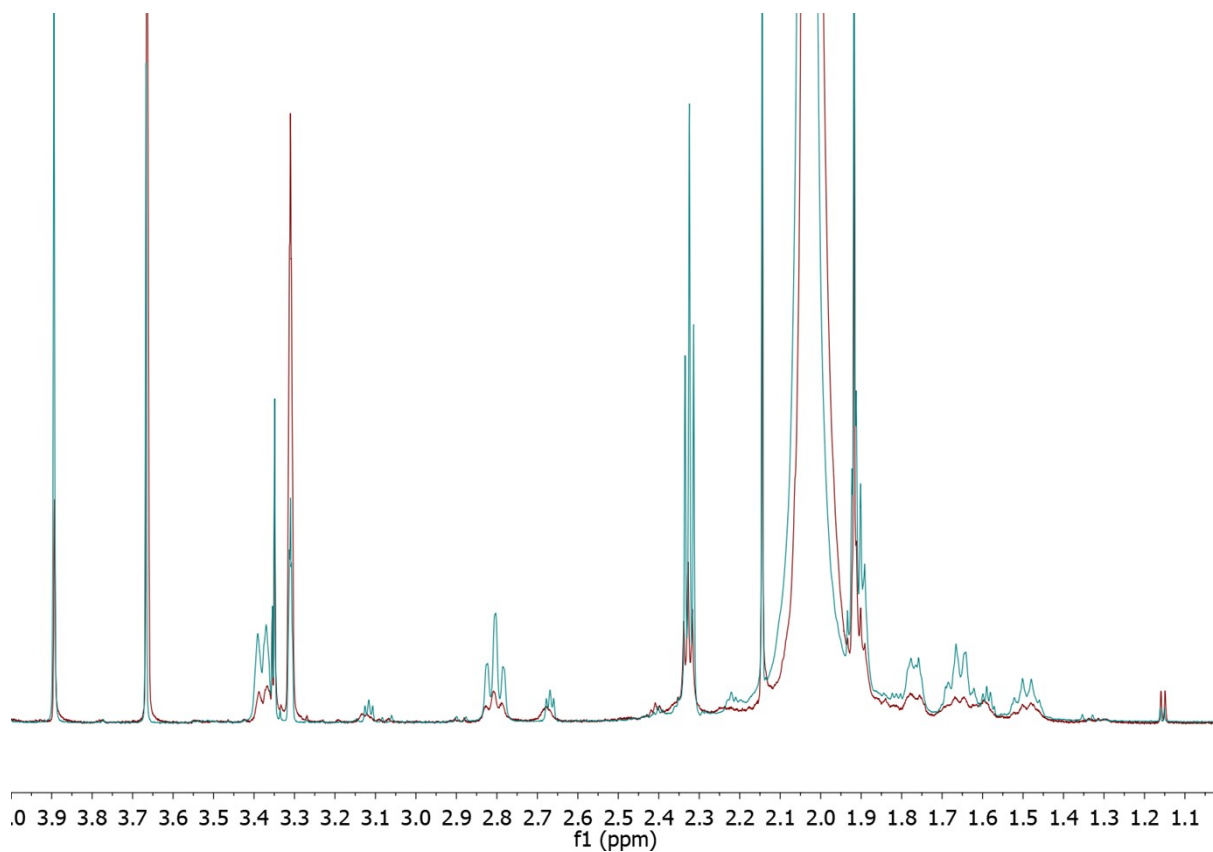
HSQC in CDCl₃ + TMS:

HMBC in CDCl₃ + TMS:

4
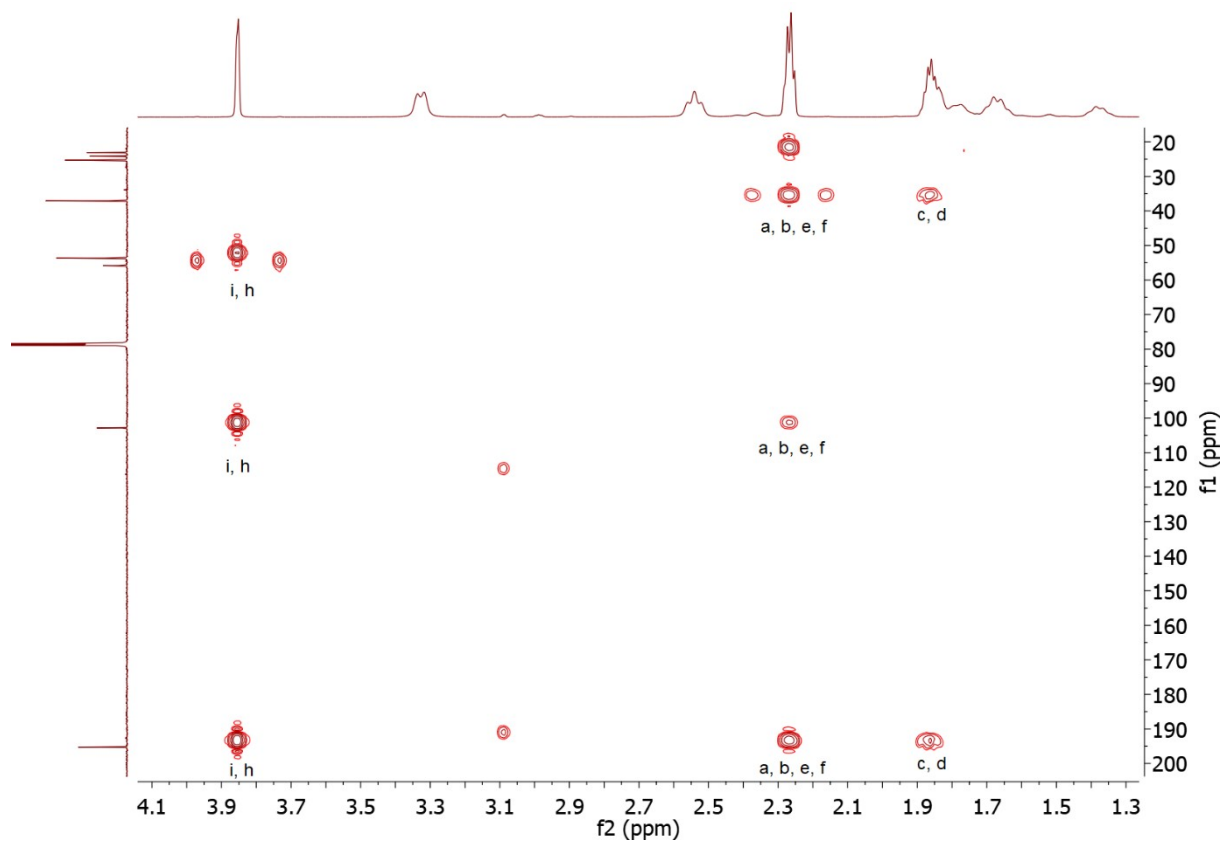
¹H-spectra in CD₃OD:

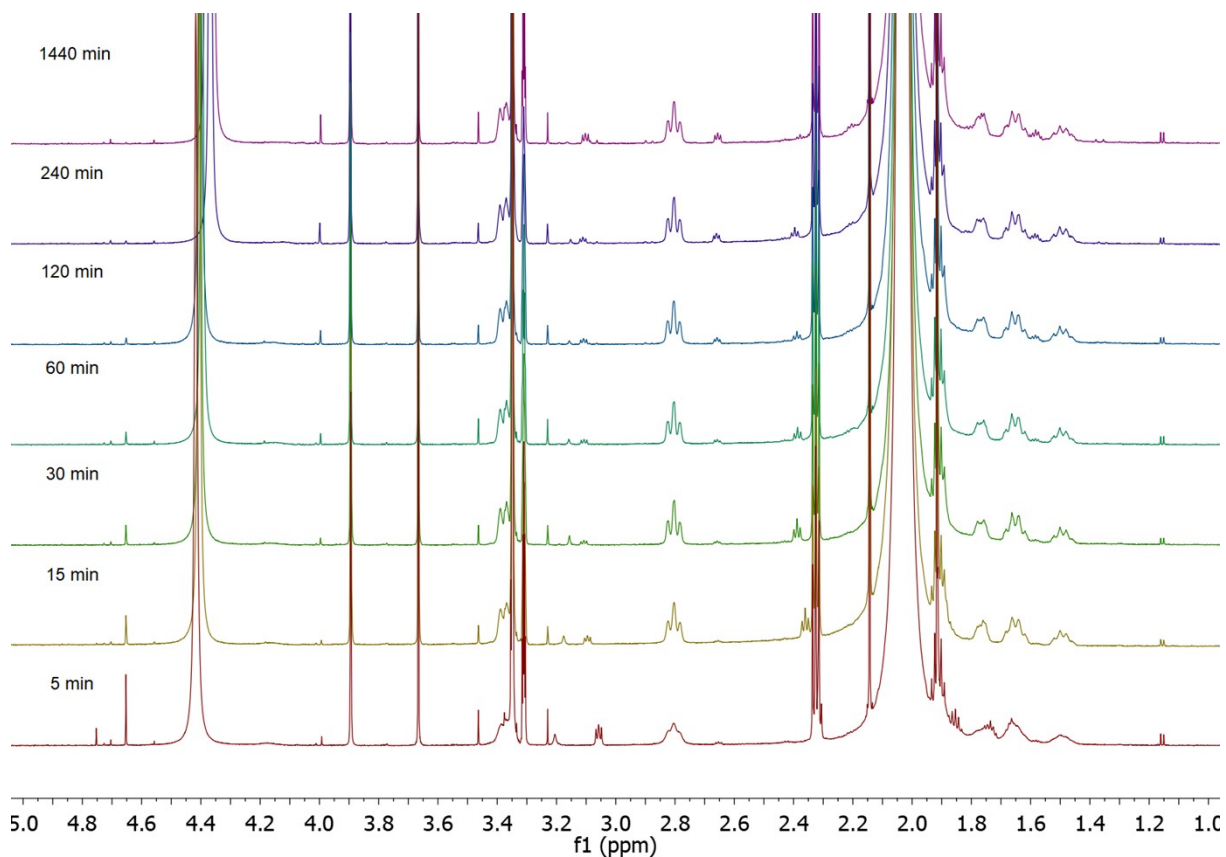**Below: 1H-NMR** spectra of reaction (Acetonitrile, 100 mM 1,3-cyclohexandione, 100 mM Pip, 120 mM FA) after 120 min reaction time (red) and spiked with 5 mg product (blue).

Kinetic investigation of the reaction:
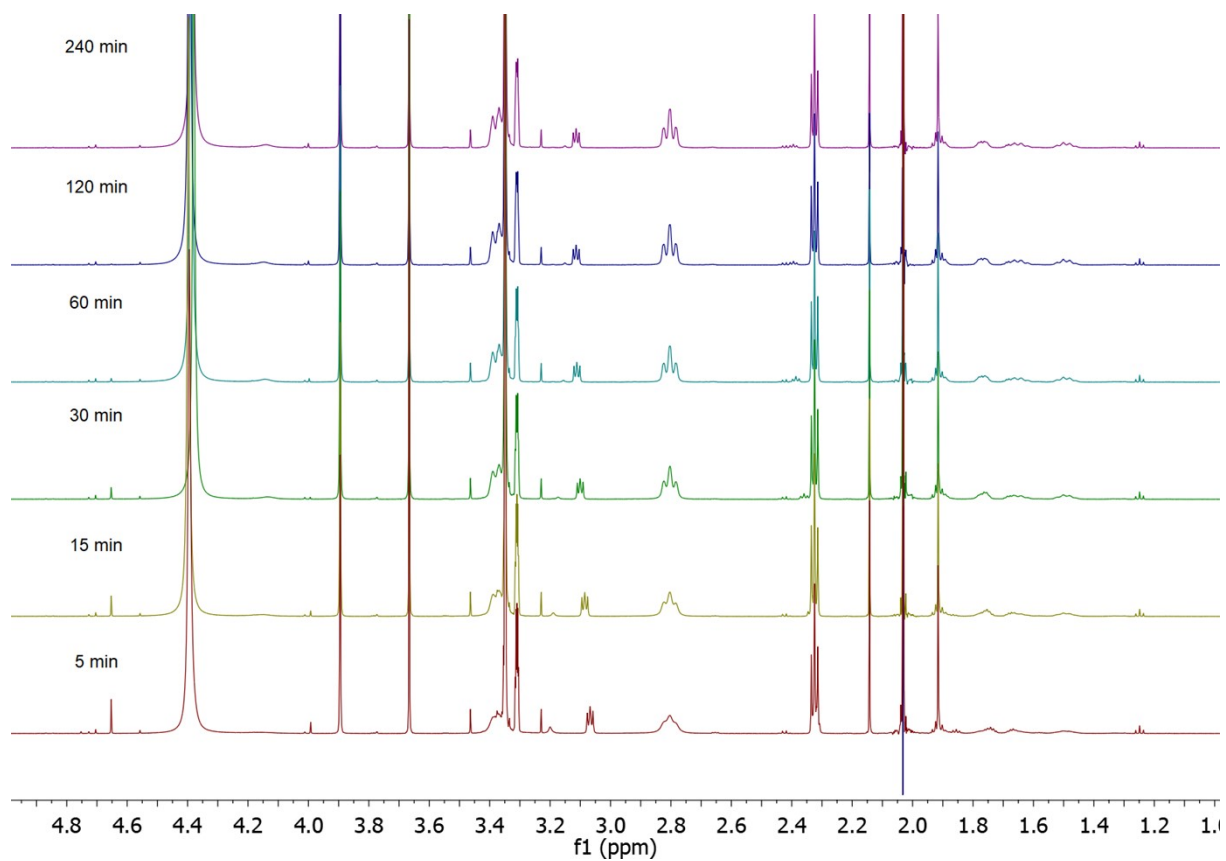
1) Cyclo without TFA: 100 mM Pip, 100 mM 1,3-cyclohexanedione, 120 mM Fal, no TFA, in CAN

2) Cyclo with TFA: 100 mM Pip, 100 mM 1,3-cyclohexanedione, 120 mM Fal, 0.1 % (v/v) TFA, in ACN

3) AcAc without TFA: 100 mM Pip, 100 mM Acetylacetone, 120 mM Fal, no TFA, in ACN
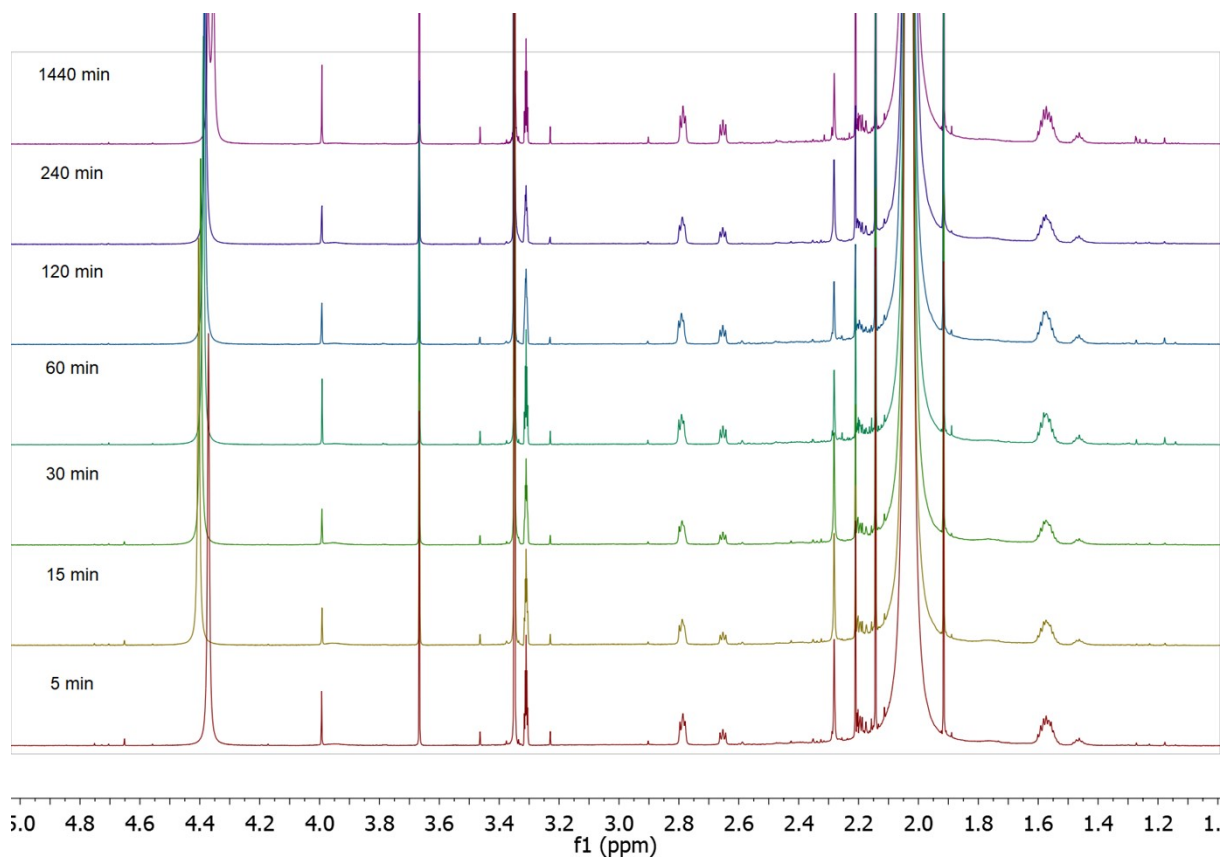
4) AcAc without TFA: 100 mM Pip, 100 mM Acetylacetone, 120 mM Fal, 0.1 % (v/v) TFA in ACN
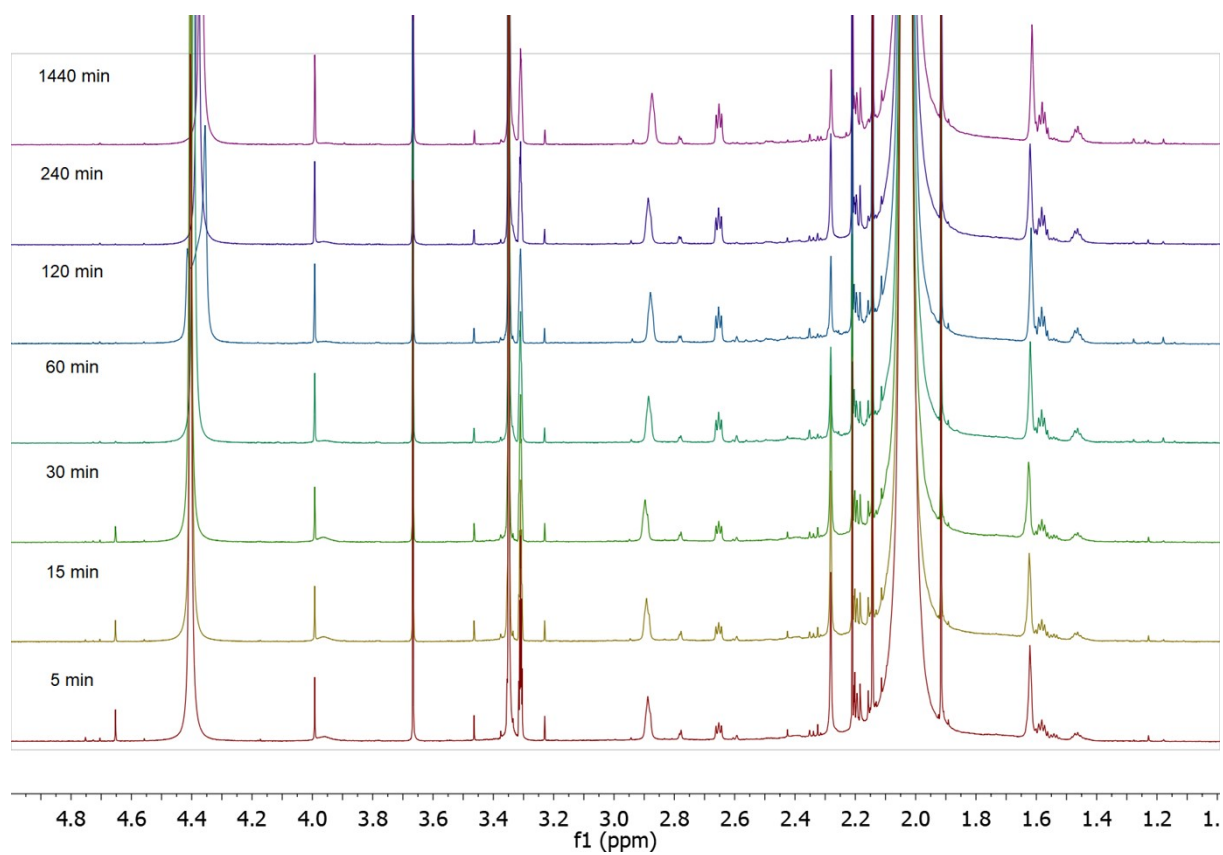
NMR Spectra of the crude reaction mixture of (E)-4-hydroxy-3-(piperidin-1-ylmethyl)pent-3-en-2-one after 24h
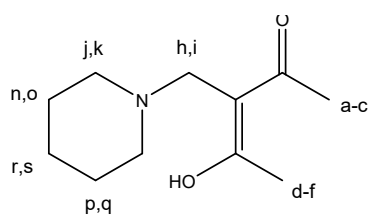
¹H NMR (600 Mhz, CD$_3$OD/ACN=2/1, rxn. mix, 25 °C): δ = 3.99 (s, 2H), 2.62-2.64 (t, *J* = 6 Hz, 4H), 2.2 (s, 6H), 1.52-1.61 (m, 4H), 1.42-1.49 (m, 2H)).

2) Detection efficiencies of reactant, intermediate and product species

The conversion ratios in this paper were calculated from the intensities of each component that were used for or generated during the Mannich reaction. The formation of each ion that is detected b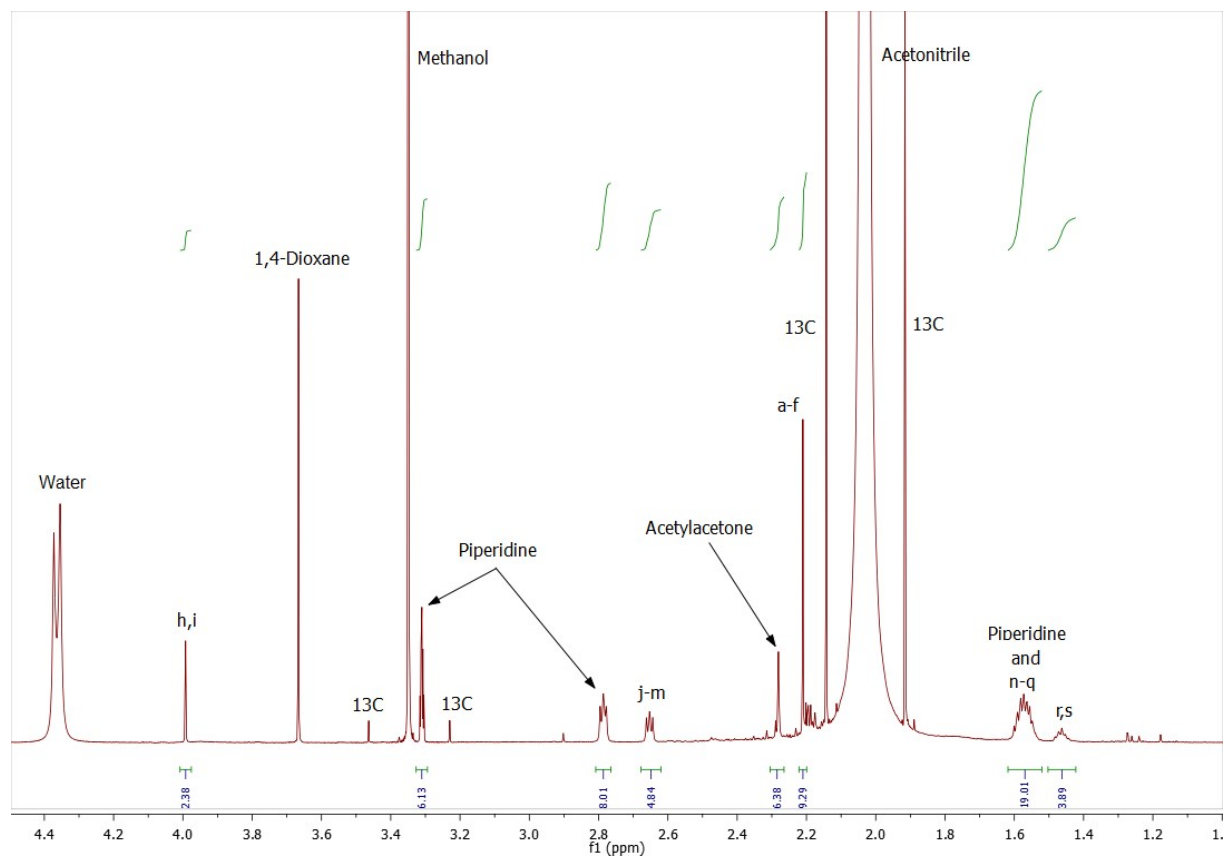y the MS can be influenced by another component due to charge competition or ion suppression during the ES process.[29] Therefore, the product of the Mannich reaction with Cyclo was synthesized in batch, isolated and cleaned up (see experimental part B). A calibration curve of the product and the ketone starting material were done at the lowest and highest tube length (rectangles, Fig. 4). Furthermore, mixtures of both components were measured to compare the intensities of each component in mixture (circles, Fig. 4) against their calibration curve intensity at same concentration. The intensity of both components is lower in the mixture than in their neat component solution. Due to higher ionization efficiency the intensity decrease in mixture is bigger for the ketone than for the product. At 2 cm tube length the intensities in the mixture reach 92-99% for the product and 83-90% for the ketone and at 60 cm 77-92% for the product and 56-74% for the ketone of the calibration curve intensity. Using that mixture intensity factors to correct the intensity for the calculation of the reaction conversion ratio at these tube lengths show a low impact. The impact was tested for all measurements following in the next sections and the deviation reaction conversion ratio was at maximum < 5% and the average was around 2% deviation.
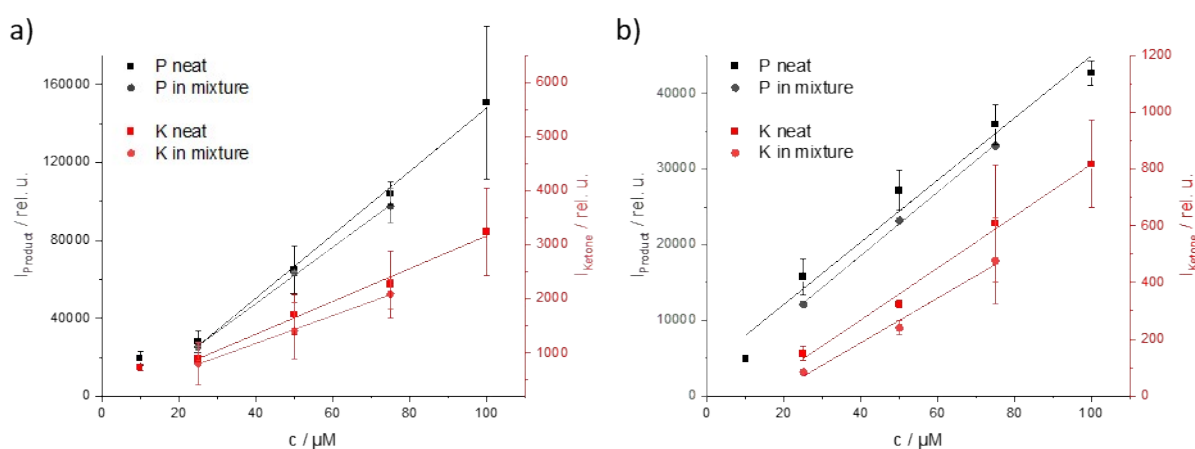

Fig. S.2 Charge competition measurements: Calibration curve of product (P) and ketone (K) (both rectangles) and their mixtures (25:75, 50:50, 75:25 [all µM in ACN], circles) - a) @2cm and b) @60cm tube length - both at 4 kV, 350°C MS inlet, 10µl/min, 2 and 1 L/min sheath gas and auxiliary gas
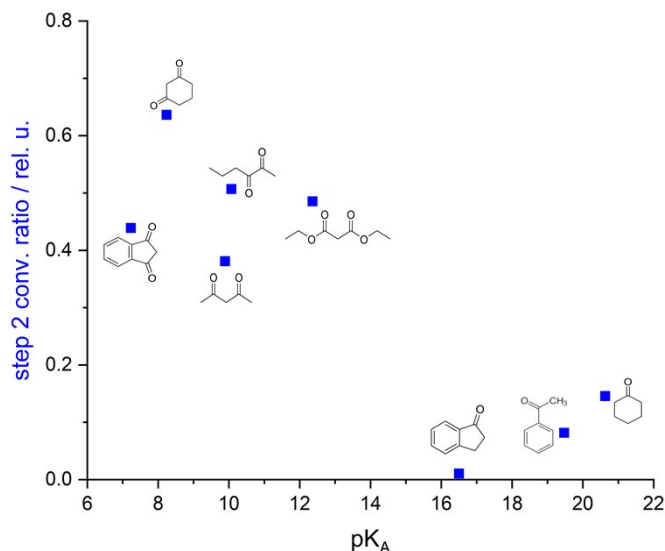
3) pK$_A$ values

Fig. S.3 Intermediate to product (step II) conversion ratios of different enolizable ketones over their keto pK$_A$ value the concentration ratio of each ketone to Fal and Pip in Fig. 5 was 1:1:1; Instrument settings: l_{guide} = 40 cm, 4 kV, 350 °C MS inlet, 10 μl/min, 2 and 1 L/min sheath and auxiliary gas

4) Calculation of droplet lifetimes over temperature

For Fig. 8f the equation for the radius of a droplet R(T, t$_0$) from Kebarle eq. 9a (Anal. Chem. Vol 65, 22, 1993, 977 A) is given as:

$$R = R_0 - \frac{\alpha M \tilde{v}\,(T) p°(T)}{4R\;\rho(T)\quad T}\,t$$

R_0 - radius of the primary droplet
α - condensation coefficient
M - molar mass of the solvent molecule
$\tilde{v}(T)$ - average molecular velocity of the solvent gas
$p°(T)$ - bulk vapor pressure of the solvent
$\rho(T)$ - density of the solvent
R_g - univesal gas constant
T - temperature of the droplet
t - time

All T-dependent terms were expressed individually to calculate the three parameters for the solvent acetonitrile:Definition of $\tilde{v}\,(T)$, the average molecular velocity:

$$\tilde{v}\,(T) = \sqrt{\frac{3RT}{M}} = \sqrt{\frac{3R}{M}}\sqrt{T}$$

The bulk density $\rho(T)$ [http://www.ddbst.com/en/EED/PCP/DEN_C3.php] and bulk vapour pressor $p°(T)$ [http://www.ddbst.com/en/EED/PCP/VAP_C3.php] are tabulated values for discrete values of T. To calculate the values used in the setupthe data with the respective functions were numerically fitted with dimensionless temperature values T/K.

$$p°(T/K) = a_p T^{b_p}$$

$$\rho(T/K) = a_\rho + b_\rho T$$

$$a_p = 2.51319 \times 10^{-19} \, kPa$$

$$a_\rho = 1101.096 \, kg/m^3$$

$$b_p = 8.13979$$

$$b_\rho = -1.087 \, kg/m^3$$

*Please note that the vapor pressure as used here is only a function of temperature $p(T/K)$ and not droplet diameter, therefore surface curvature effects on the vapor pressure are not accounted for.

Substituting these numerical fit functions of the temperature-dependent vapor pressure and bulk density into the original equation allowed for the calculation of the droplet evolution over time for a given temperature value T in Kelvin. The equation below followed, which was used for the calculation of the data points of figure 8f.

$$R = R_0 - \frac{\alpha M}{4R} \sqrt{\frac{3R}{M} \frac{a_p T^{b_p}}{a_\rho + a_\rho T}} \frac{1}{\sqrt{T}} t$$
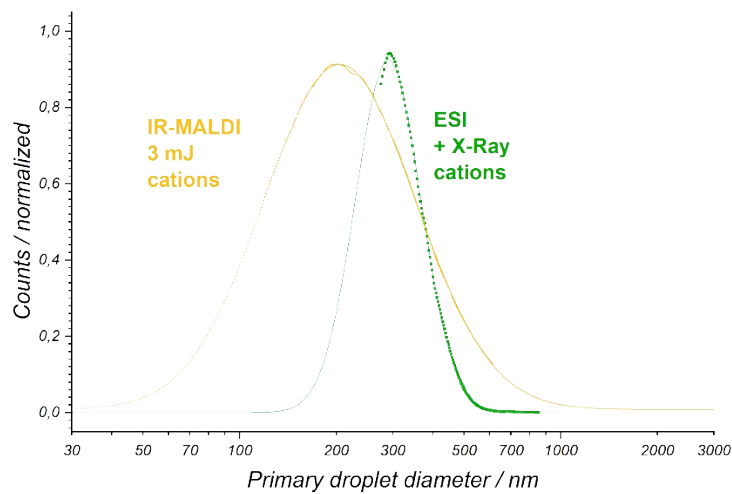
5) Primary droplet size IR-MALDI vs. ESI

Fig. S5 Droplet diameter of the primary produced droplets via ESI (green) and IR-MALDI (yellow) – measured with differential mobility analyzer system (DMA 3081/3085/3085A, Classifier 3082, CPC 3788, AAN 3088) operated with the Aerosol Instrument Manager Software 10.2.0.11, all by TSI GmbH Co & KG, Germany.