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Electronic Supplementary Information

Development of Antimicrobial Ion Jelly Fibers

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General

All reagents and solvents were obtained commercially, unless otherwise noted, and appropriately purified, if necessary. ^{1}H and ^{13}C NMR spectra were recorded at room temperature on a Bruker AVANCE II+ 300 MHz NMR spectrometer, using D_2O as solvent and the residual solvent peak as reference. Water content of all ionic liquids was measured by Karl Fisher titration with a Methrom 831 KF coulometer.

DSC measurements were obtained from service of REQUIMTE-DQ-FCT (Portugal) and carried out with a Setaram DSC 131 calorimeter at a scanning rate of 10 $^{o}C\ min^{-1}$.

Experimental procedures

Synthesis of choline-based ionic liquids. These ionic liquids were synthesized following the common procedure described in the literature by Ohno[1]. Choline chloride (1.047 g, 7.5 mmol), dissolved in 75 mL of methanol, was slowly passed through a column filled with 12.5 g of resin AMBERLITE IRA-400 (OH). The dripping choline hydroxide solution was collect in an Erlenmeyer containing a slight excess of the desired carboxylic acid dissolved in methanol. The resulting solution was passed

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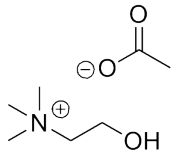
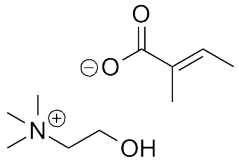
through a small column filled with Celite® 577, activated basic aluminum oxide and silica-gel, for removal of unreacted carboxylic acid and visible particles. After solvent removal by evaporation under reduced pressure, the resulting product was dried *in vacuo* at 50 ºC overnight.

Choline acetate (1)



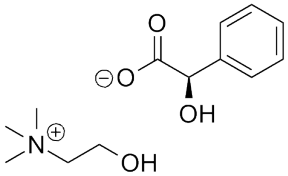
Obtained as a white to pale yellow solid (1.058 g, 86.3 %). ^{1}H RMN δ (ppm): 4.05 (m, 2H), 3.51 (m, 2H), 3.19 (s, 9H), 1.91 (s, 3H). ^{13}C RMN δ (ppm): 181.0, 67.4, 55.6, 53.8, 23.1.

Choline tiglate (2)



Obtained as a yellow viscous liquid (1.397 g, 91.6 %). ^{1}H RMN δ (ppm): 6.43 (m, 1H), 4.03 (m, 2H), 3.48 (m, 2H), 3.17 (s, 9H), 1.71 (m, 6H). ^{13}C RMN δ (ppm): 178.0, 133.4, 132.6, 67.4, 55.5, 53.8, 13.3, 12.5.

Choline (-)-mandelate (3)



Obtained as a pale yellow viscous liquid (1.458 g, 76.2 %). ^{1}H RMN δ (ppm): 7.39 (m, 5H), 4.96 (s, 1H), 4.00 (m, 2H), 3.43 (m, 2H), 3.12 (s, 9H). ^{13}C RMN δ (ppm): 179.3, 140.5, 128.7, 128.1, 127.0, 74.9, 67.3, 55.5, 53.8.

Choline (±)-ibuprofenate (4)

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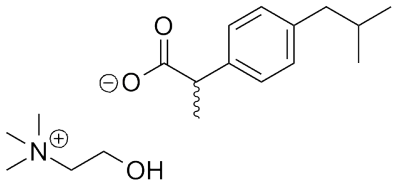

Obtained as a white to pale yellow solid (1.964 g, 84.6 %). ^{1}H RMN δ (ppm): 7.20 (m, 4H), 4.00 (m, 2H), 3.59 (m, 1H), 3.44 (m, 2H), 3.16 (s, 9H), 2.42 (d, 2H), 1.80 (m, 1H), 1.37 (d, 3H), 0.84 (d, 6H). ^{13}C RMN δ (ppm): 183.4, 140.6, 140.1, 129.3, 127.2, 67.3, 55.5, 53.8, 48.1, 44.3, 29.7, 21.7, 18.5.

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NMR Spectra

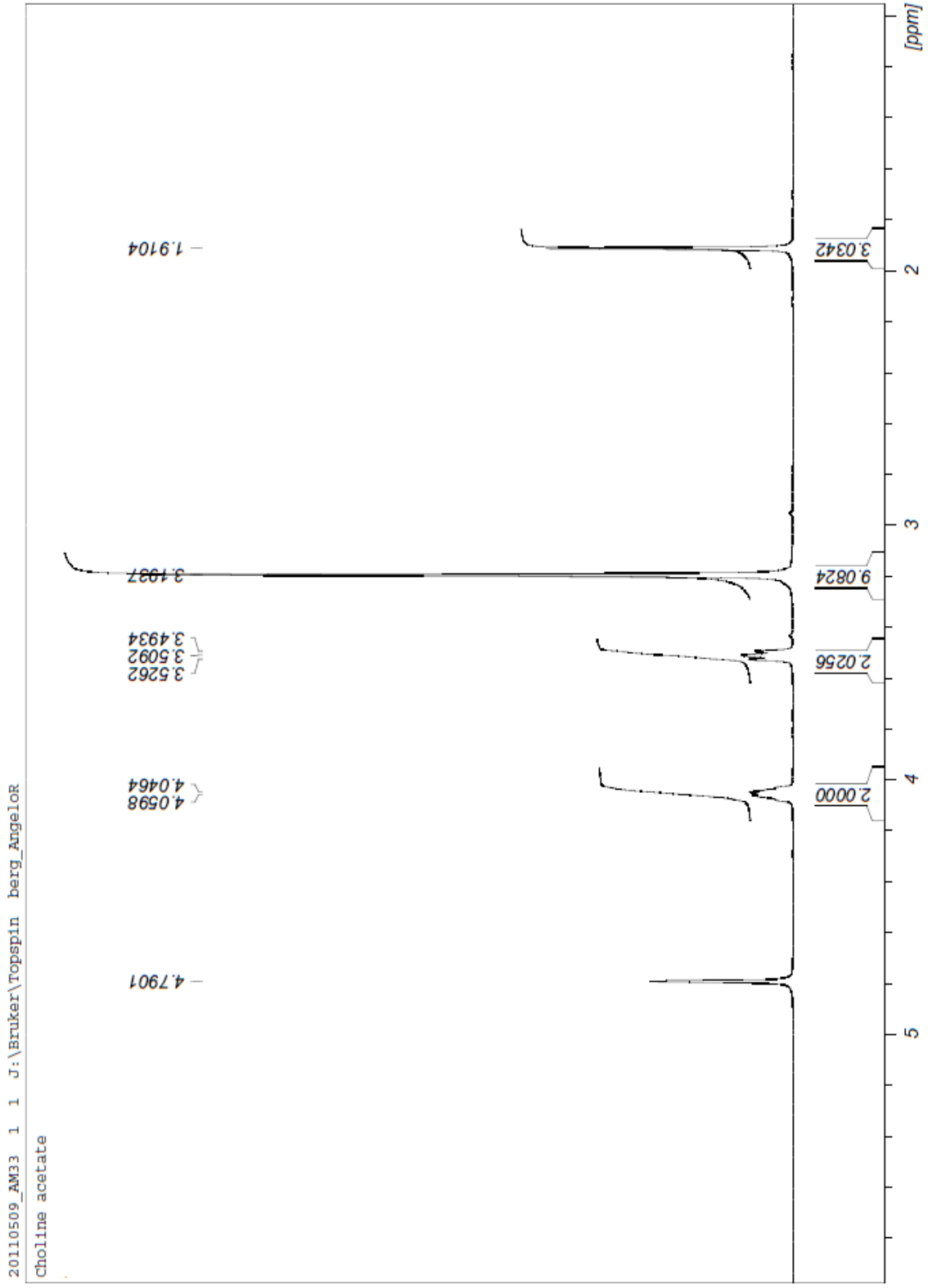

Figure 1 – ^{1}H NMR spectrum of choline acetate (**1**) in D_2O at room temperature.

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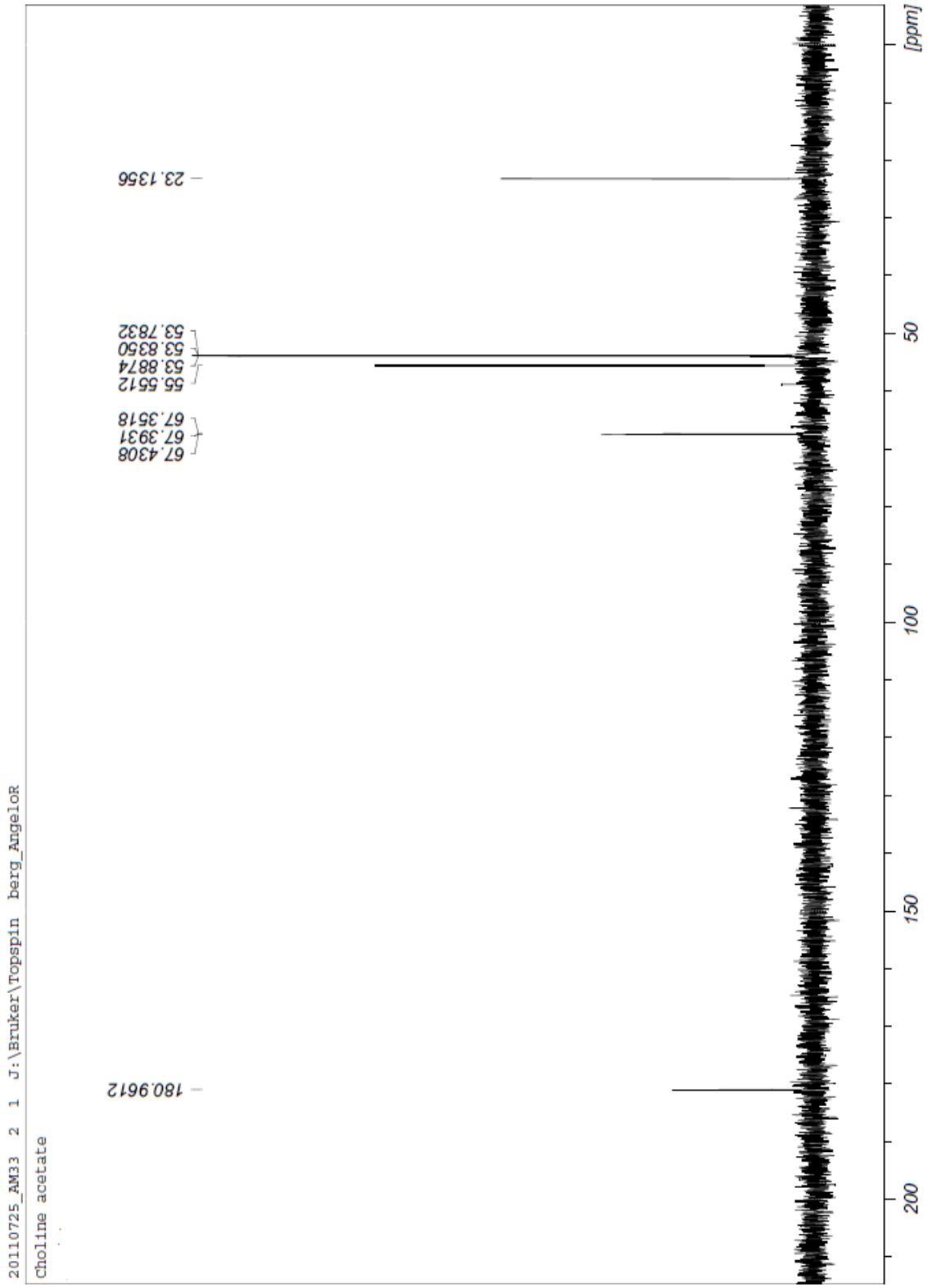

Figure 2 – ^{13}C NMR spectrum of choline acetate (**1**) in D_2O at room temperature.

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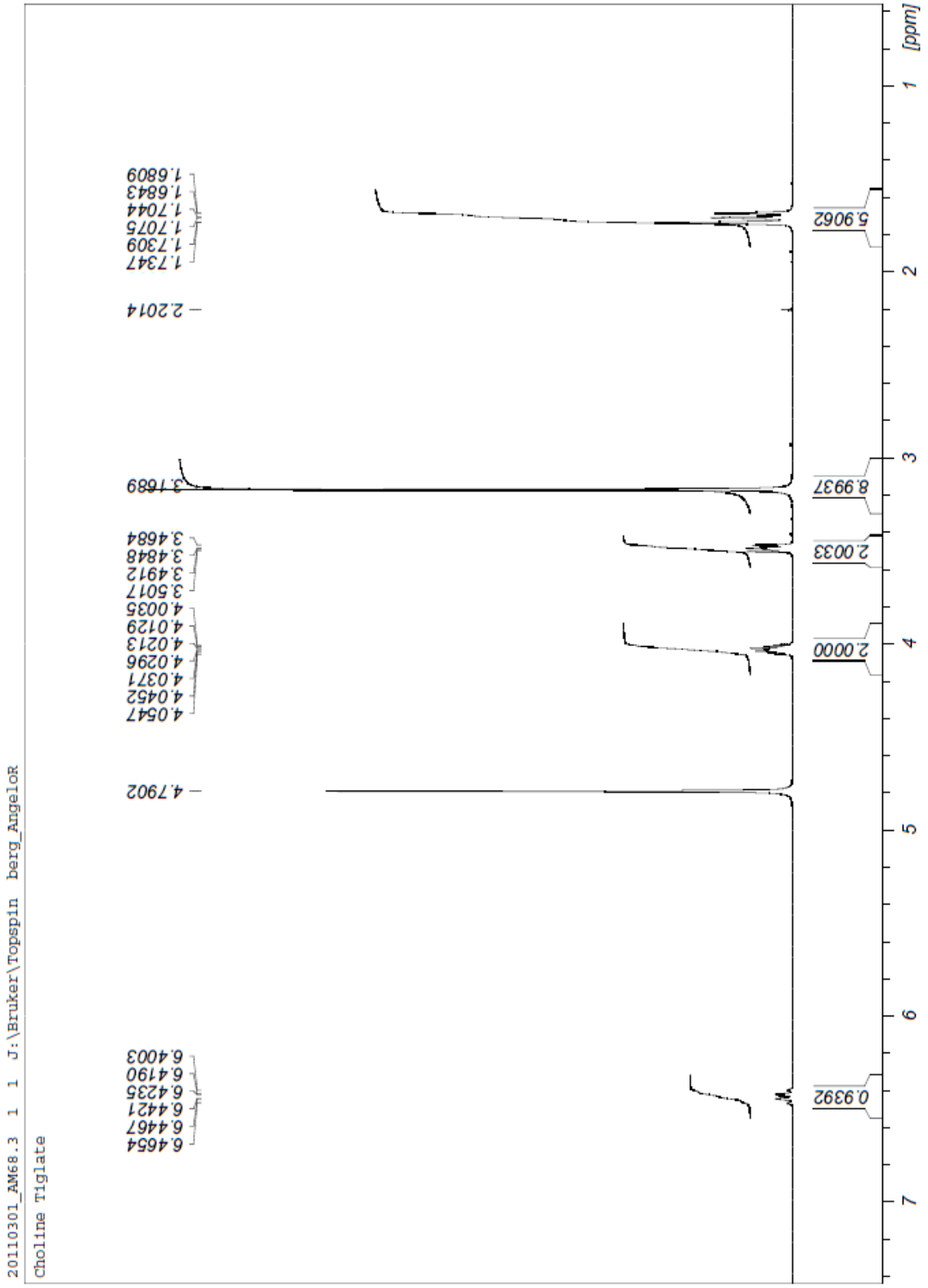

Figure 3 – ^{1}H NMR spectrum of choline tiglate (**2**) in D_2O at room temperature.

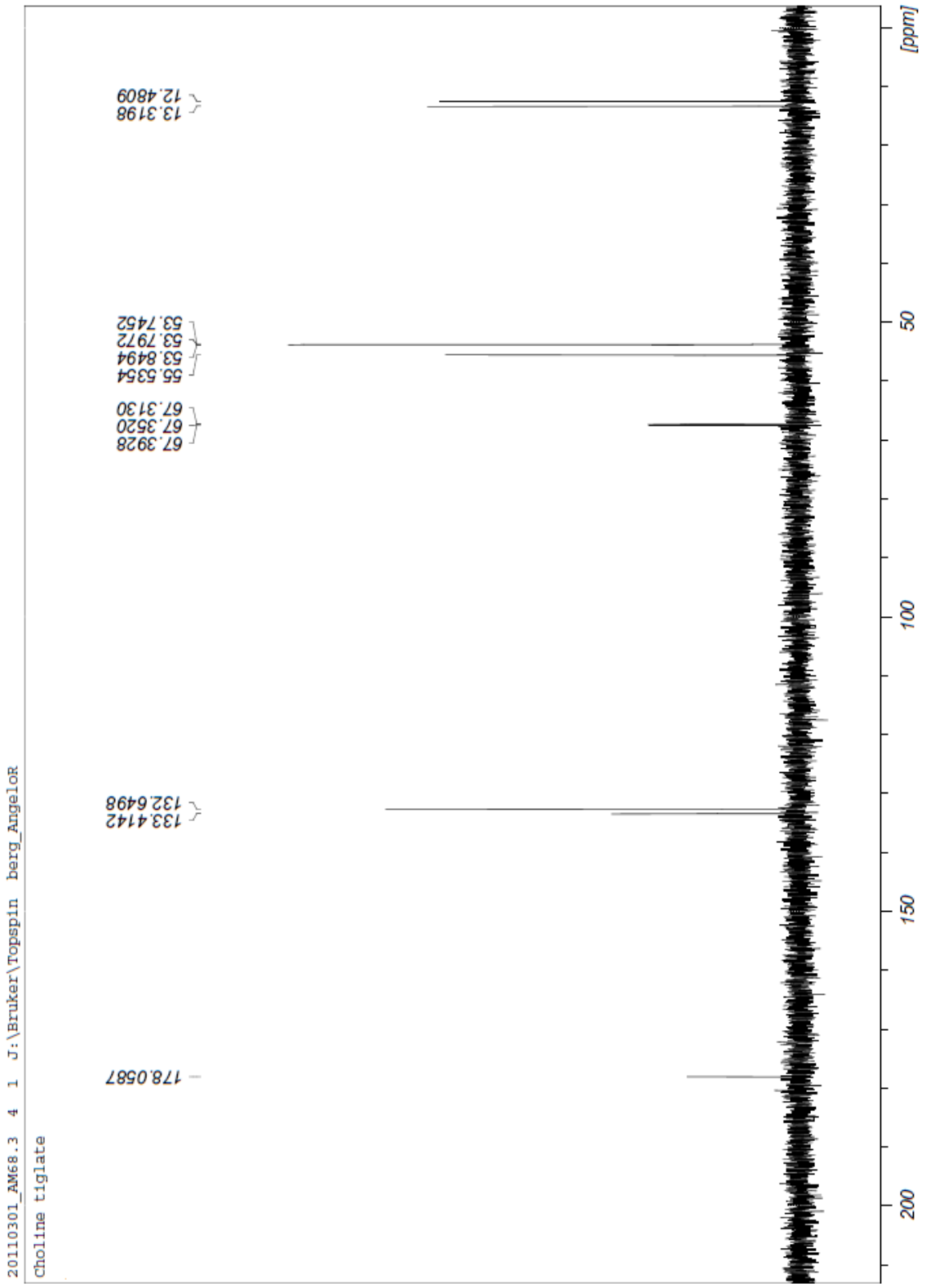

Figure 4 – ^{13}C NMR spectrum of choline tiglate (**2**) in D_2O at room temperature.

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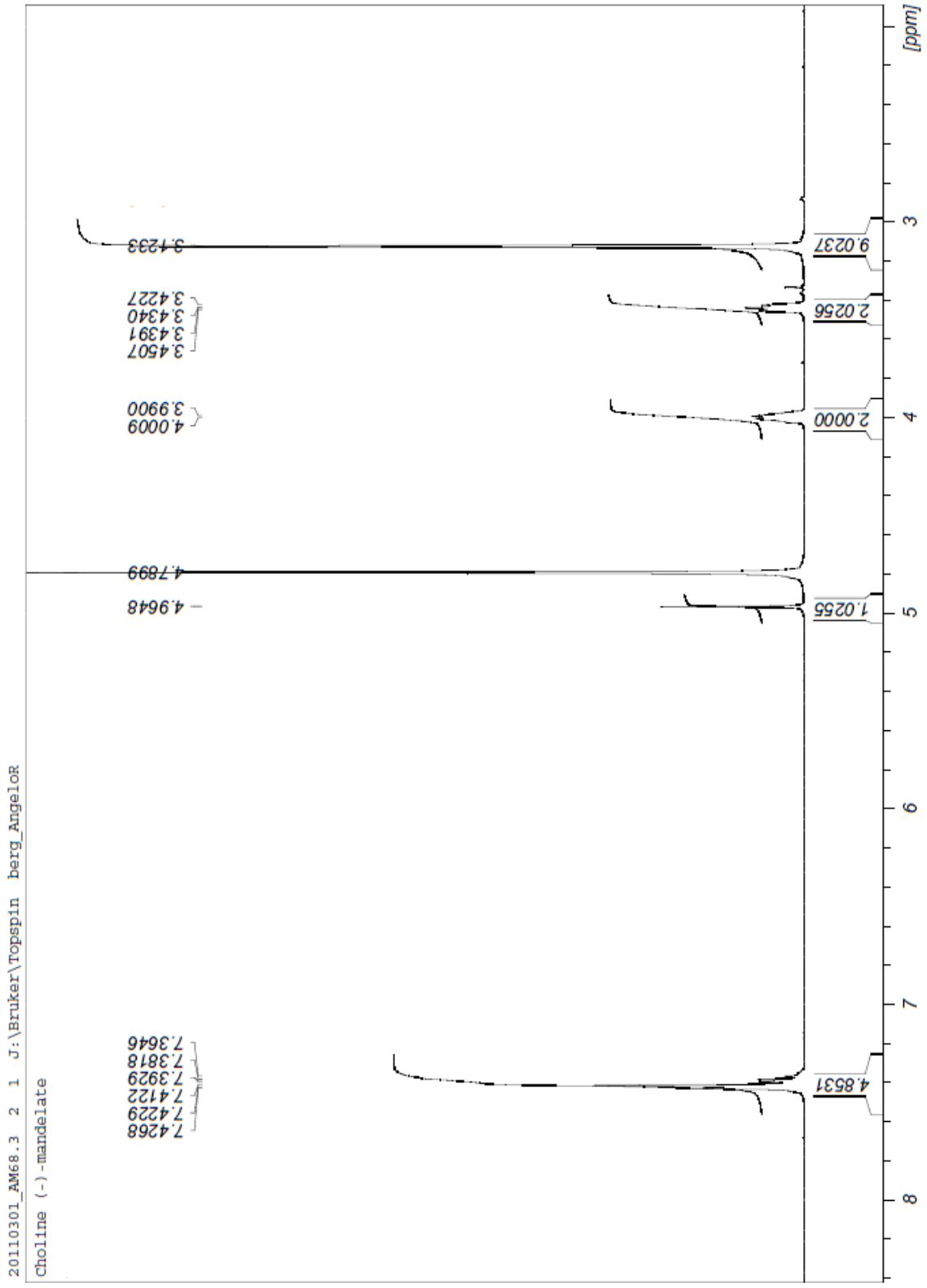

Figure 5 – ^{1}H NMR spectrum of choline (-)-mandelate (**3**) in D_2O at room temperature.

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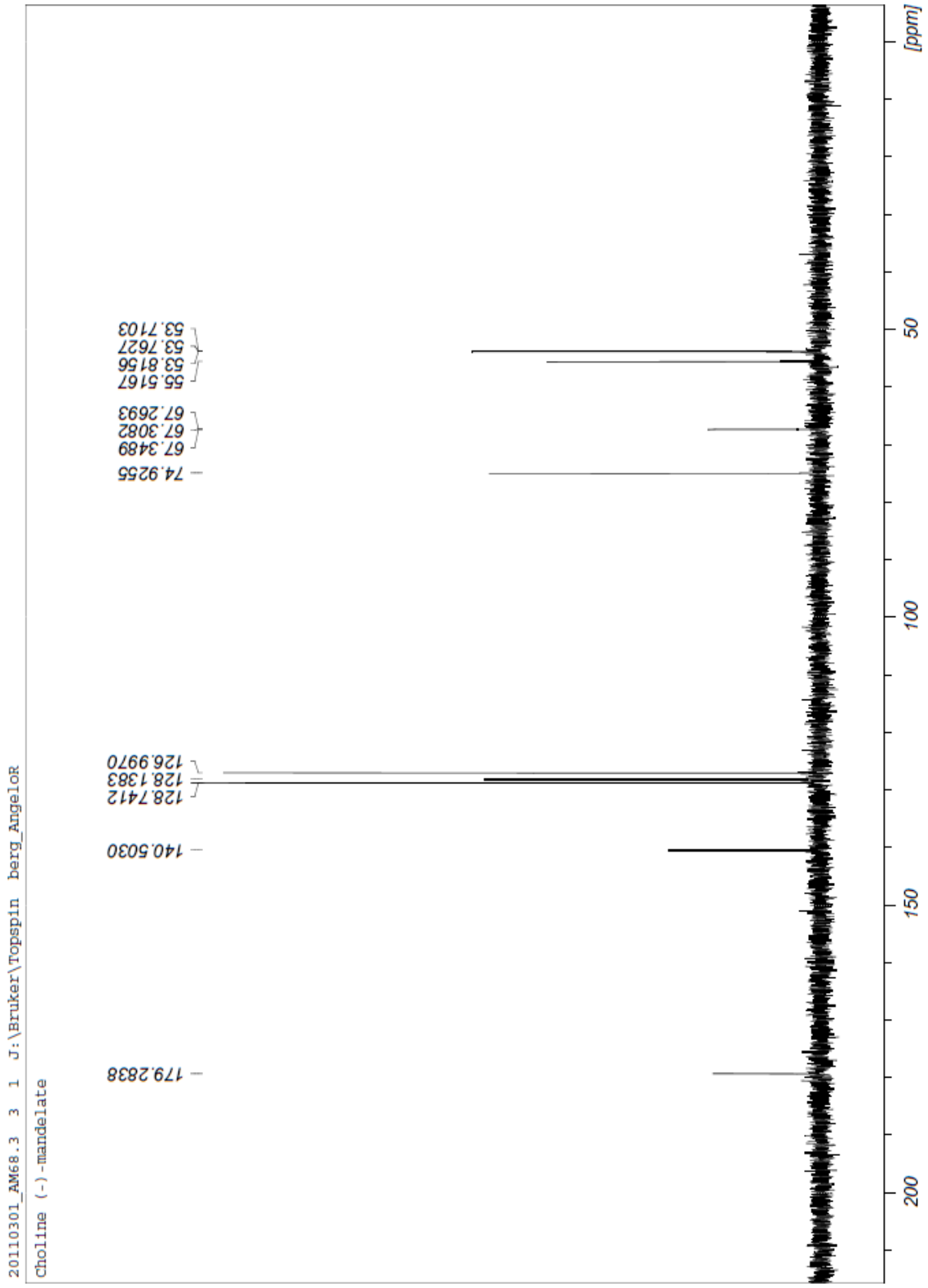

Figure 6 – ^{13}C NMR spectrum of choline (-)-mandelate (**3**) in D_2O at room temperature.

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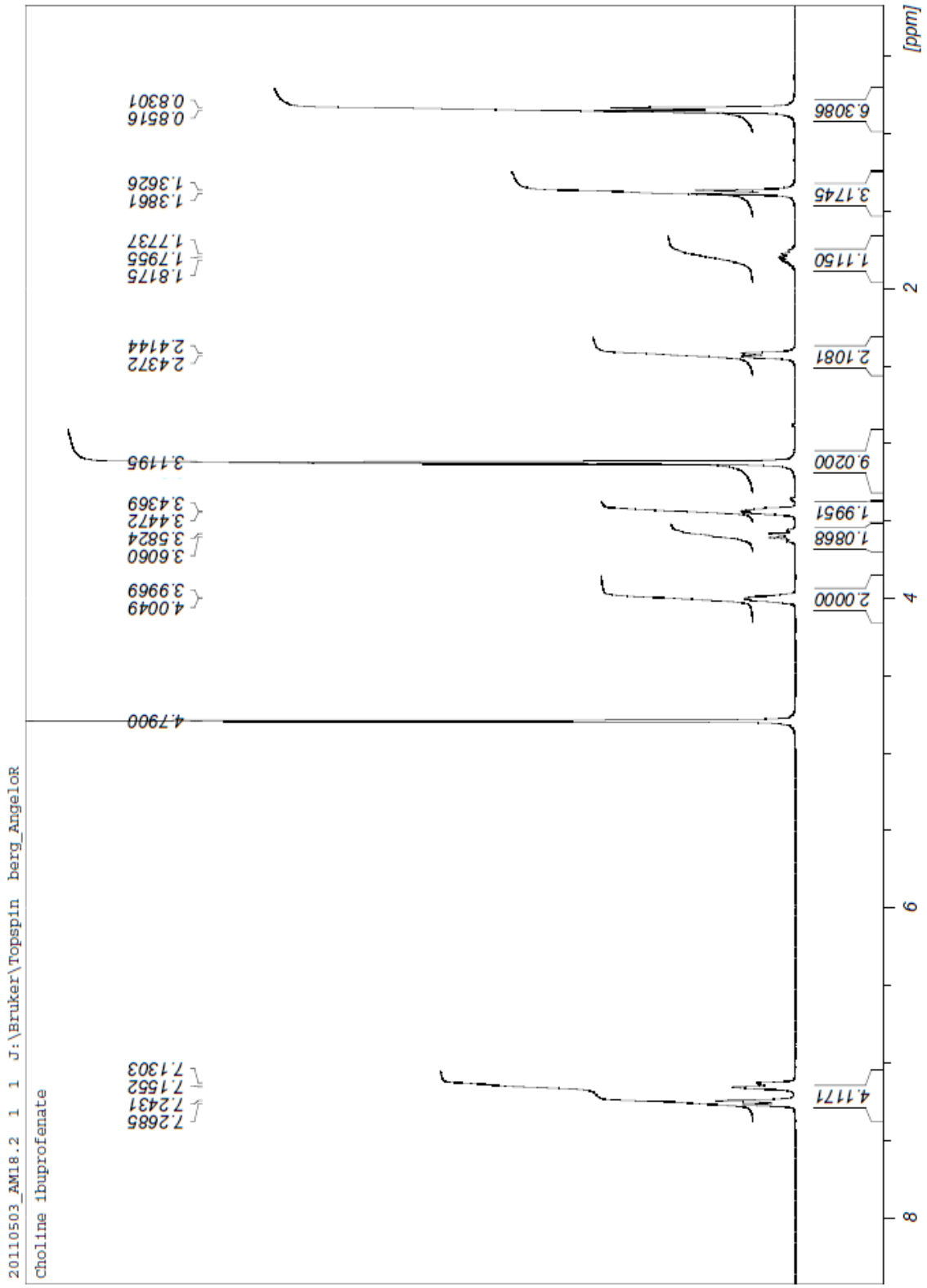

Figure 7 – ^{1}H NMR spectrum of choline (±)-ibuprofenate (**4**) in D_2O at room temperature.

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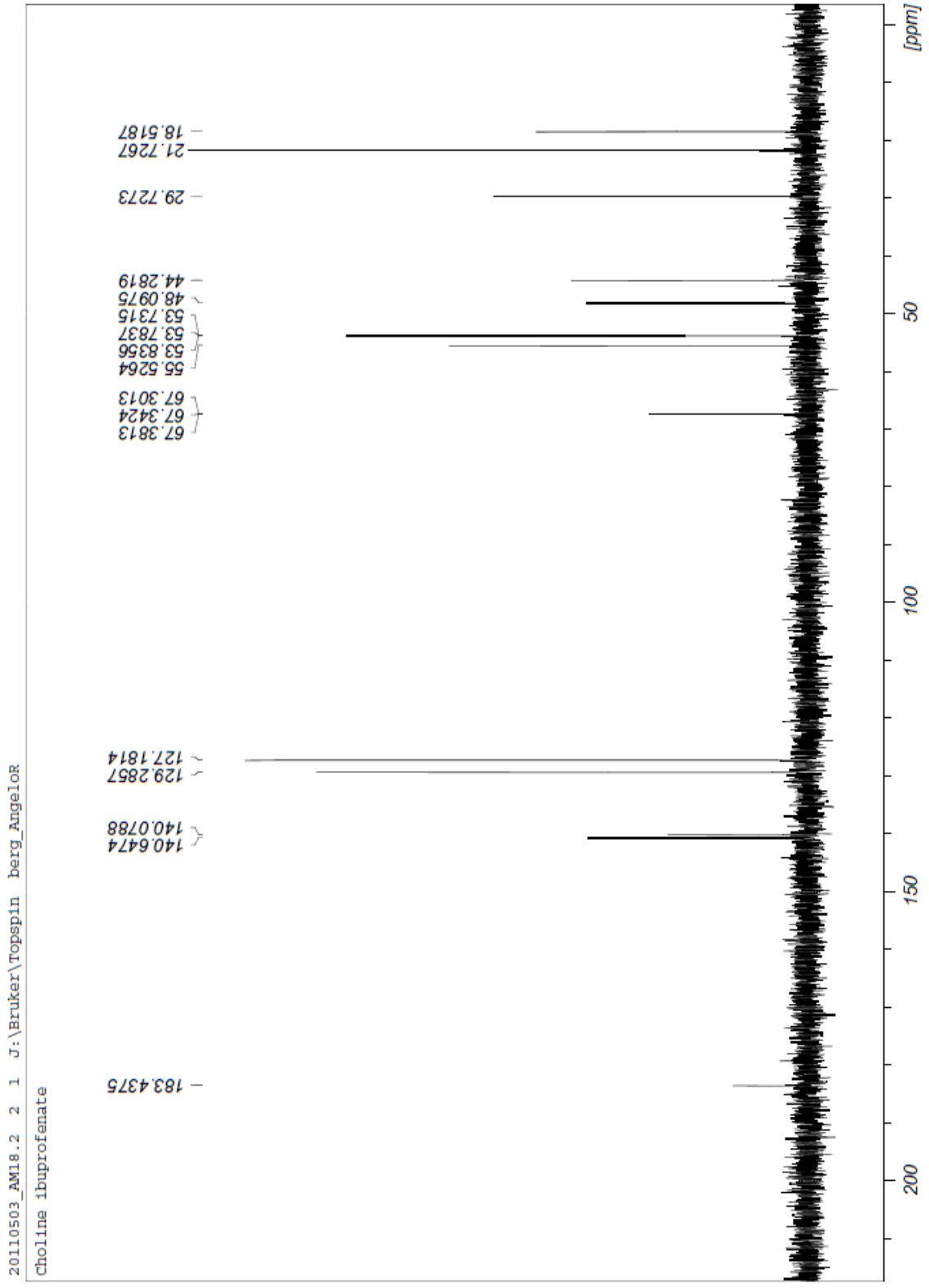

Figure 8 – ^{13}C NMR spectrum of choline (±)-ibuprofenate (**4**) in D_2O at room temperature.

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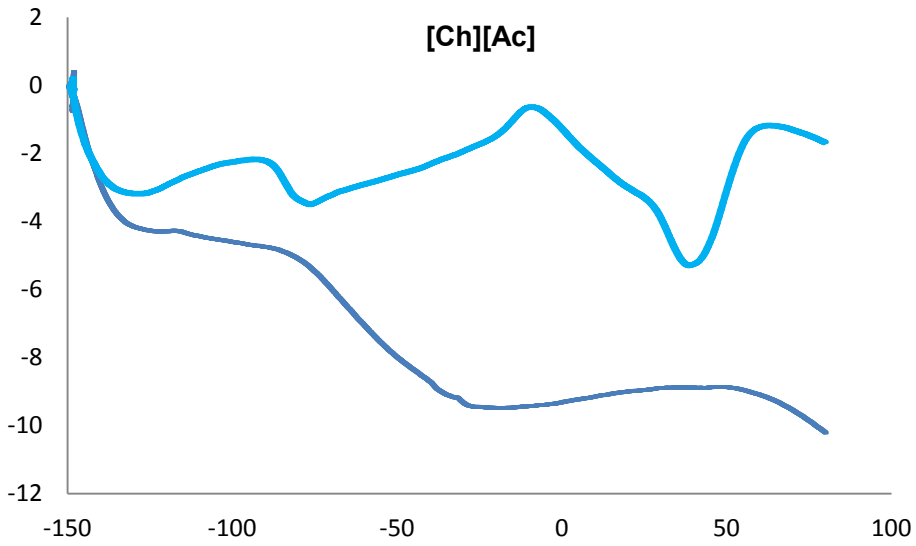
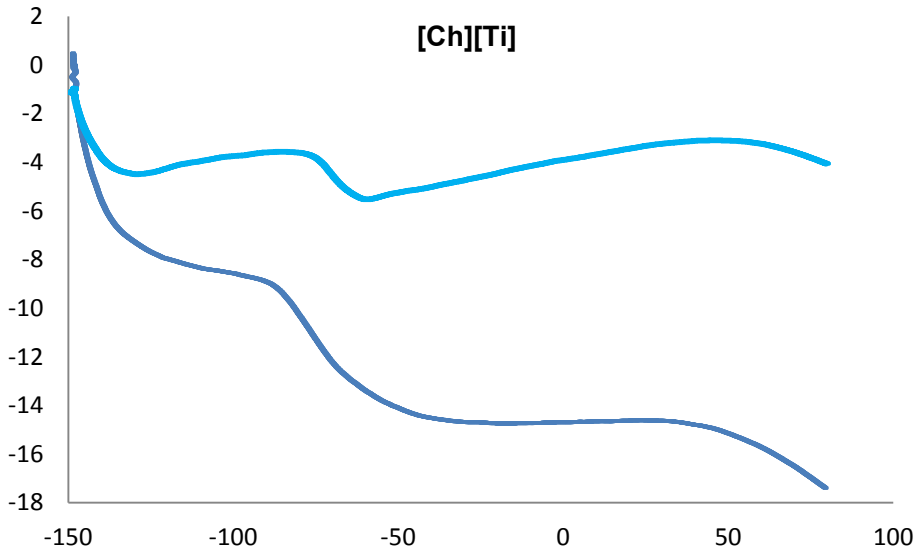
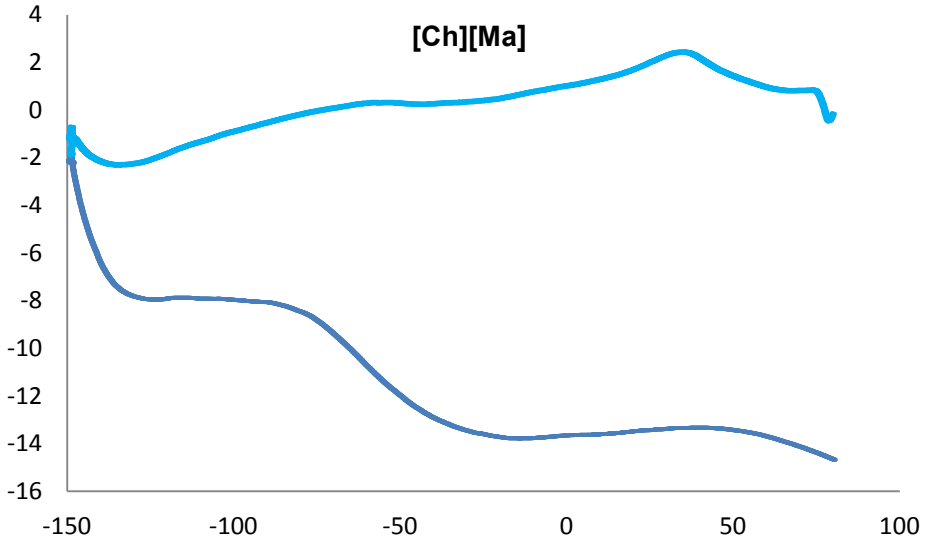
DSC analysis

Both choline-based ILs and the resulting IJs were submitted to DSC analysis to analyze their phase transitions.







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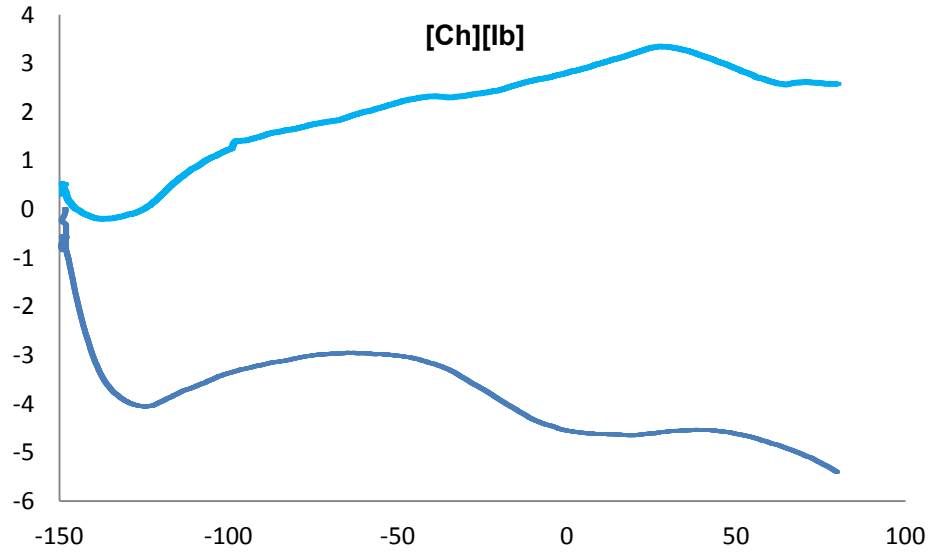

Figure 1 – DSC curves of Ionic Liquid (IL) and Ion Jelly (IJ) (IJ, IL, ↑ exothermic, abscissa units: W/g, ordinate units: ºC). [Ch][Ac] – Choline Acetate; [Ch][Ib] – Choline ibuprofenate; [Ch][Ma] – Choline Mandelate; [Ch][Ti] – Choline Tiglate.

Cytotoxicity evaluation on human Caco-2 cells

Cell toxicity assays were performed using human Caco-2 cells. Briefly, Caco-2 cells were seeded at a density of 2×10^4 cells/well in 96-well plates and their media (RPMI 1640 supplemented with 10% of FBS) was replaced every 48 hours. The experiments were performed using cells after reaching confluence - 72 hours after seeding. After that, medium was removed and cells were washed twice with PBS and treated for 4h with different concentrations of Choline-based ILs in culture medium supplemented with 0,5% FBS [15000-500 µM]. Control cells contained culture medium alone. After 4 hours of incubation, the medium was removed and 100 µL of a CellTiter 96® AQueous One Solution Cell Proliferation Assay reagent (containing MTS and PES) was added to each well and left to react for 4 hours. This solution reagent contains a tetrazolium compound (3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium, inner salt; MTS] and an electron coupling reagent (phenazine ethosulfate, PES). PES has an enhanced chemical stability which allows it to be combined with MTS and leading to a stable solution. MTS is bio-reduced by cells into a coloured formazan product that is soluble in tissue culture medium. Formazan was quantified spectrophotometrically at 490 nm in a BioTek FLx800 microplate reader

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(BioTek, USA). Each sample was incubated in six different wells and obtained value was the average of 3 independent assays. Cell viability was determined by the ratio between the measured absorbance.

Ion Jelly SEM

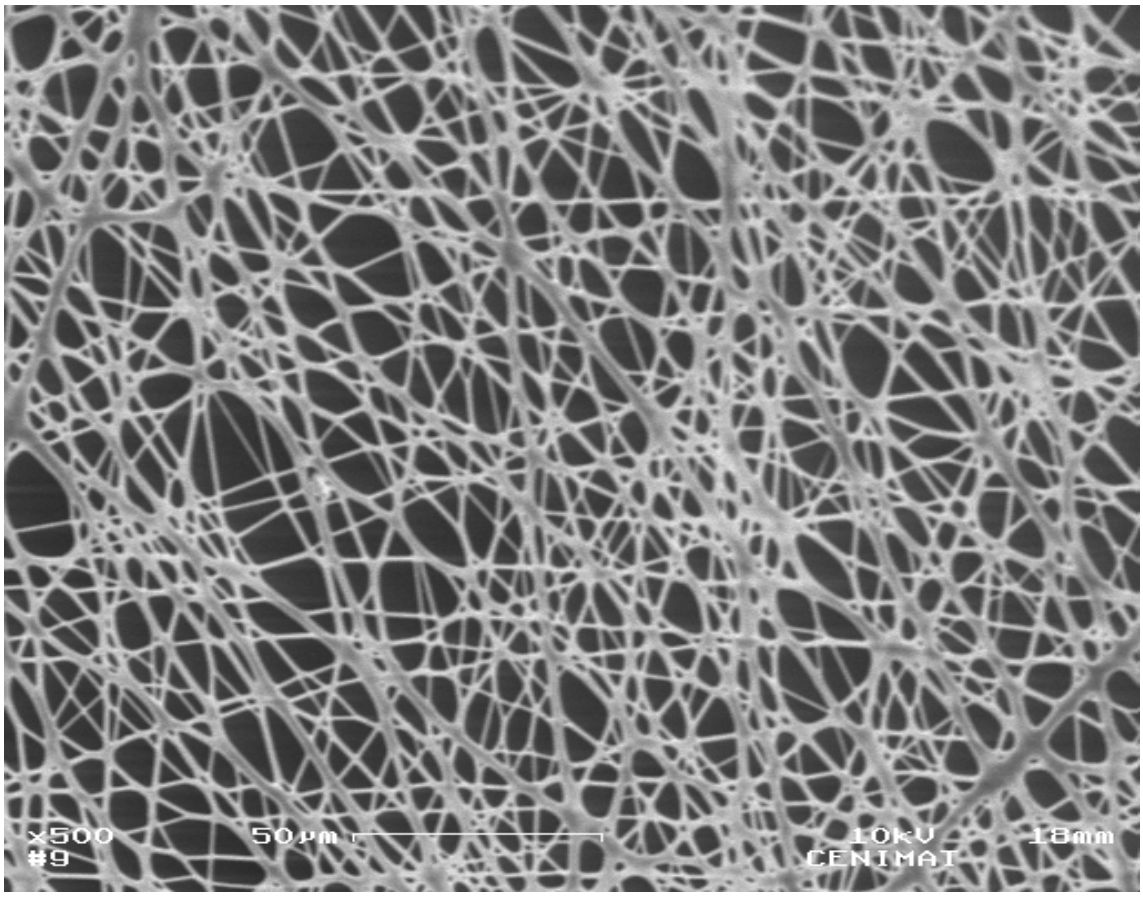

Figure E1- [Ch][ib] fibers.

References

1. Y. Fukaya, Y. Iizuka, K. Sekikawa and H. Ohno, *Green Chem.*, 2007, **9**, 1155.