

# Supporting Information for

## Flow Synthesis of L- $\alpha$ -Glycerylphosphorylcholine: Studies on Synthetic Routes Applicable to a Flow Reactor and Optimization of Reaction Conditions

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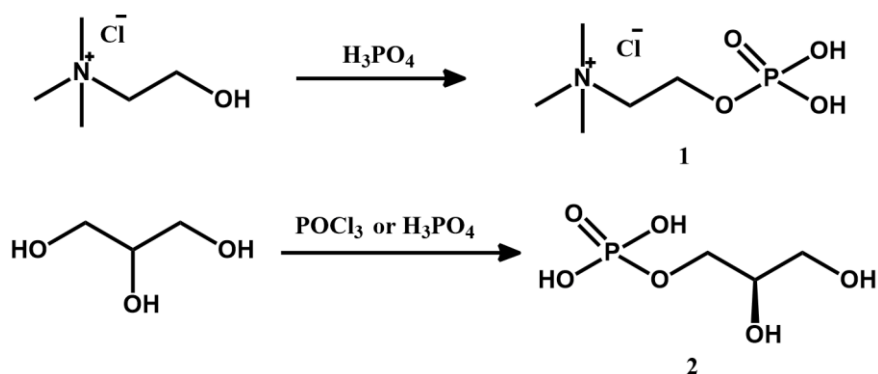
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## Material synthesis



**Scheme S1.** First step reactions to prepare L- $\alpha$ -GPC.

### **N,N,N-trimethyl-2-(phosphonoxy)ethanaminium chloride (compound 1)**

Choline chloride (50 g, 0.358 mol) and phosphoric acid (100 g, 1.02 mol) were mixed in a flask equipped with a Dean-Stark trap. The solution was mixed during warming up to 170 °C for 6 hours. After the reaction was ended, the reaction mixture was dried in vacuo and phosphocholine (0.286 mol, 80%) was obtained as colorless liquid.

### **(R)-2,3-dihydroxypropyl dihydrogen phosphate (compound 2)**

Glycerol (32.8 g, 0.358 mol) and phosphoric acid (100 g, 1.02 mol) were mixed in a flask equipped with a Dean-Stark trap. The solution was mixed during warming up to 170 °C for 6 hours. After the reaction was ended, the reaction mixture was dried in vacuo but (R)-2,3-dihydroxypropyl dihydrogen phosphate was not synthesized.

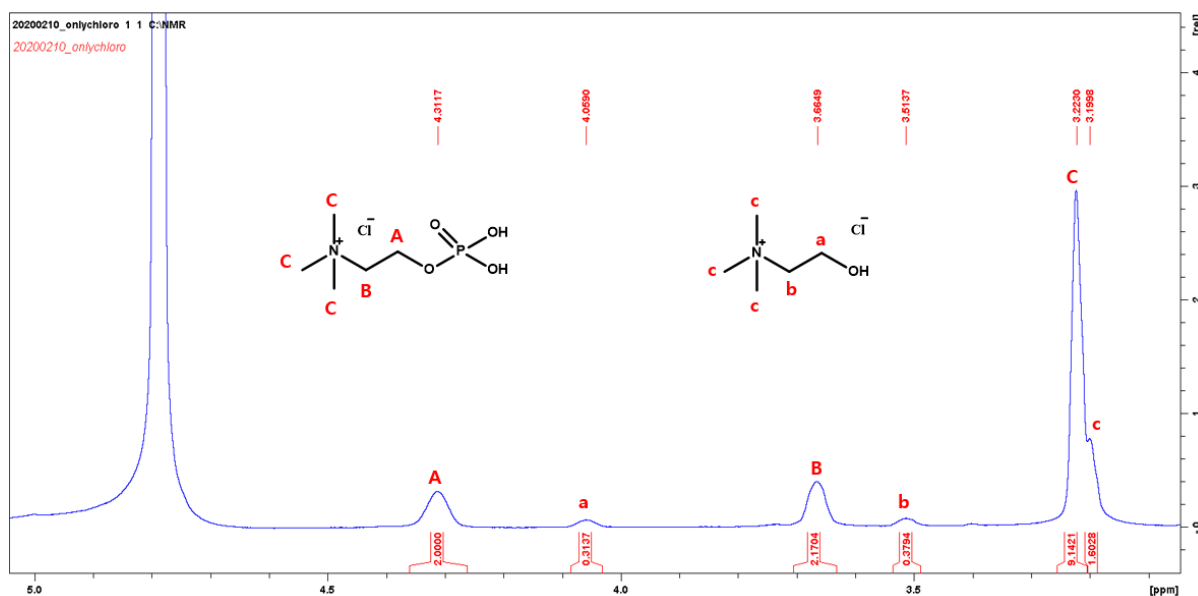
### **(R)-2,3-dihydroxypropyl dihydrogen phosphate (compound 2)**

POCl<sub>3</sub> (1.75 g, 12.5 mmol) and 2 mL of anhydrous chloroform were mixed in a flask under nitrogen atmosphere and stirred for 10 min at room temperature. Glycerol (0.132 mg, 1.44 mmol) was slowly added to the flask and the reaction was carried out at room temperature for 15 hours. After the reaction was ended, the reaction mixture was quenched by adding deionized water to remove unreacted POCl<sub>3</sub>. The solution was dried in vacuo at 60 °C, but (R)-2,3-dihydroxypropyl dihydrogen phosphate was not synthesized.

## NMR data

**Table S1.** Optimized reaction conditions and conversions of first step reactions in a batch reactor.

Entry	reactant	phosphorylation reagent	solvent	temperature (°C)	reaction time (hr)	conversion <sup>a</sup> (%)
1-1	choline chloride	POCl <sub>3</sub>	H <sub>2</sub> O/CHCl <sub>3</sub>	25	4	86
1-2	Choline chloride	H <sub>3</sub> PO <sub>4</sub>	-	170	6	< 20
1-3	choline chloride	H <sub>3</sub> PO <sub>4</sub>	-	170	6	80
1-4	glycerol	POCl <sub>3</sub>	CHCl <sub>3</sub>	25	15	-
1-5	glycerol	H <sub>3</sub> PO <sub>4</sub>	-	170	6	-



**Figure S1.** <sup>1</sup>H NMR spectrum of entry 1-1.

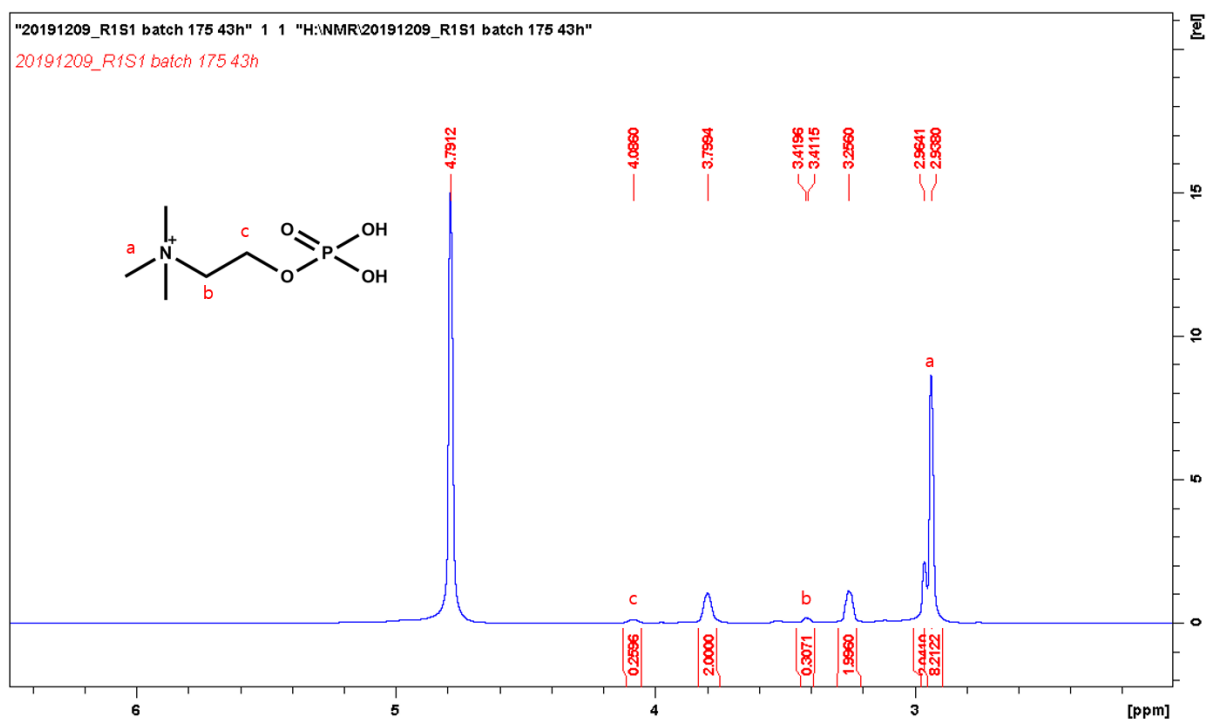


Figure S2.  $^1\text{H}$  NMR spectrum of entry 1-2.

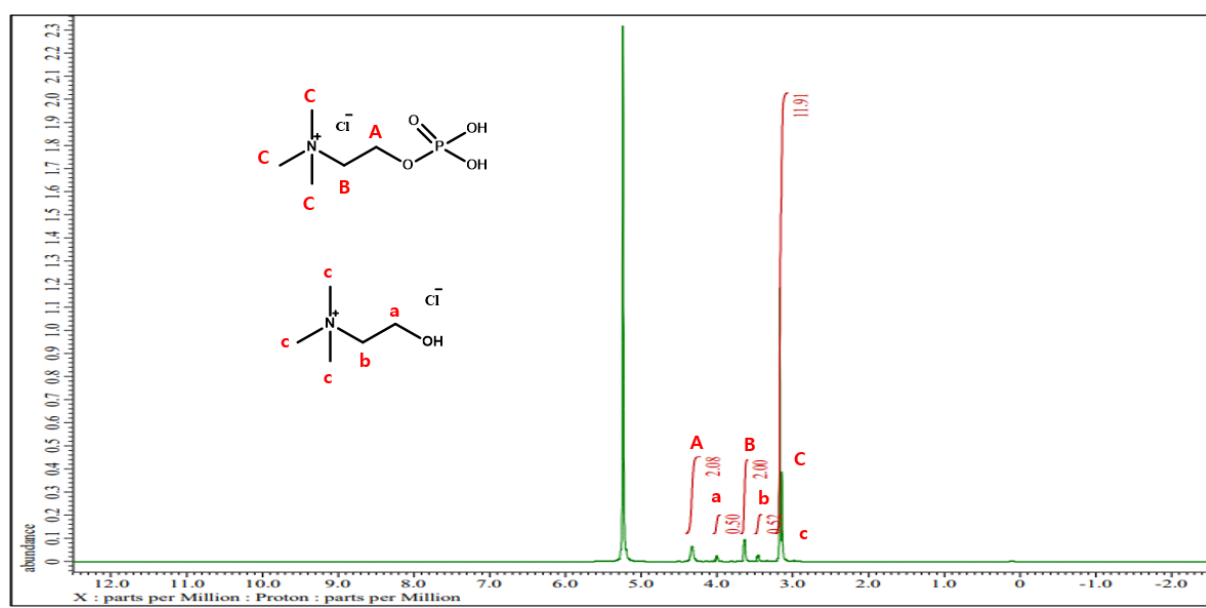
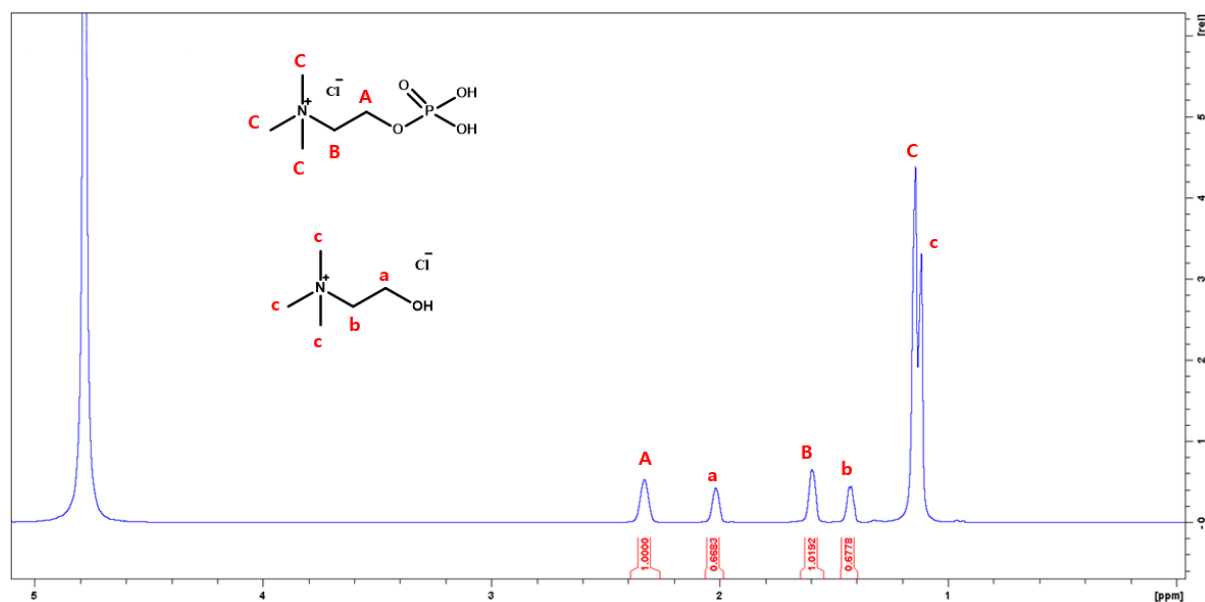


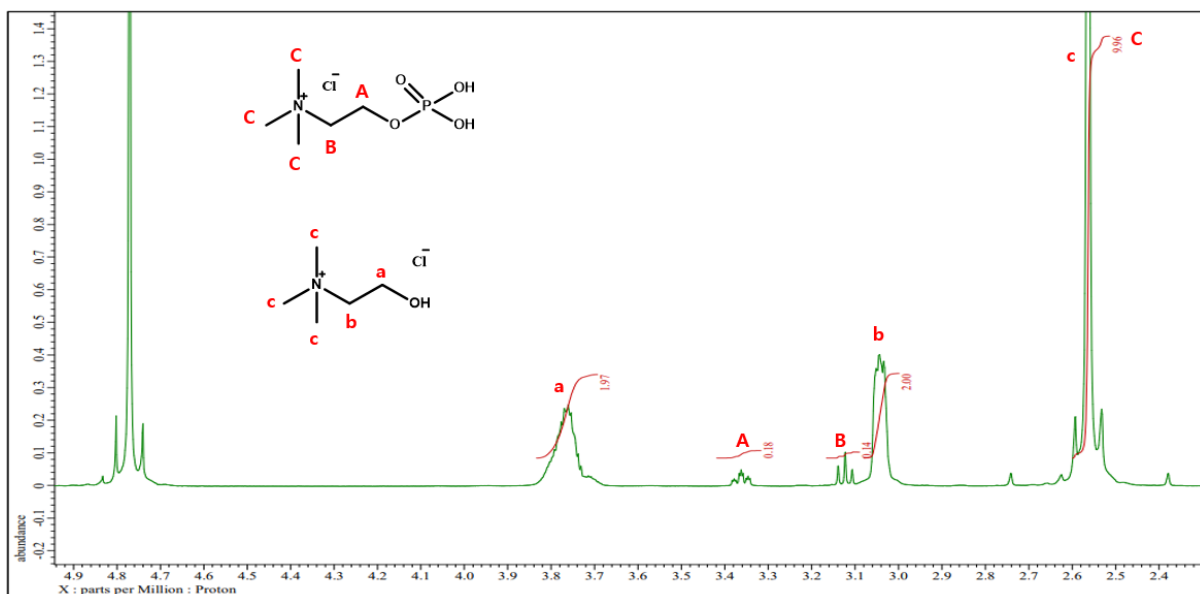
Figure S3.  $^1\text{H}$  NMR spectrum of entry 1-3.

**Table S2.** Reaction conditions and conversions of first step reactions in a flow reactor.

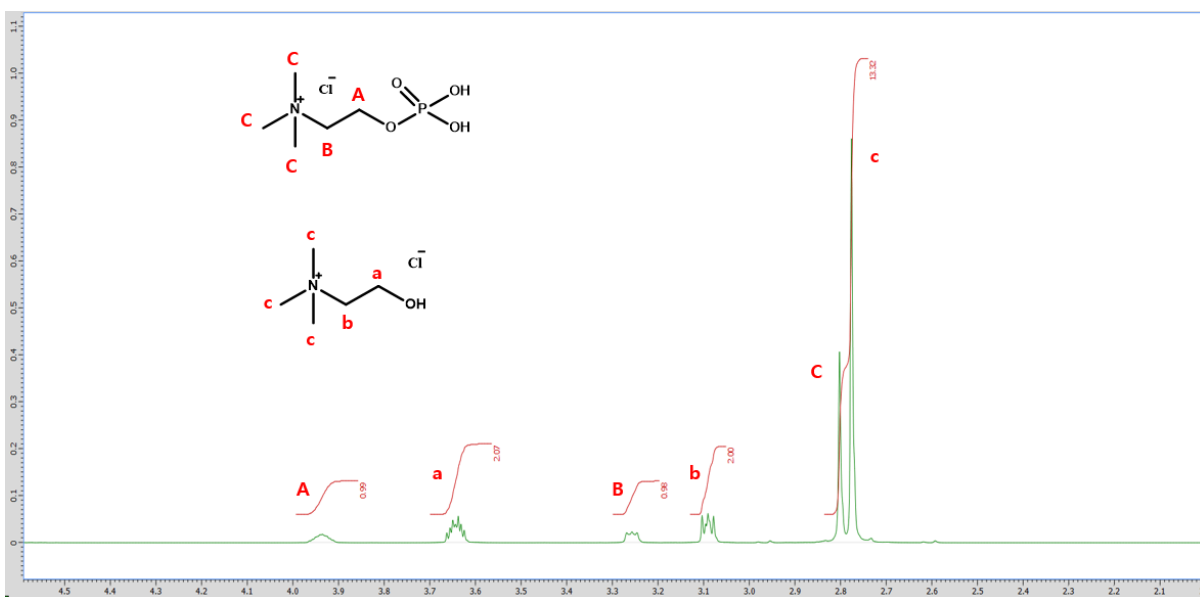
entry	H <sub>2</sub> O <sup>a</sup> (mL)	POCl <sub>3</sub> / choline chloride <sup>b</sup>	reactor volume (mL)	temperature (°C)	flow rate (μL/min)	reaction time	Conversion <sup>c</sup> (%)
2-1	0.3	8	22.5	25	31.25	1hr 38min	60
2-2	0.3	8	22.5	0	31.25	4hr	8
2-3	0.3	8	22.5	30	31.25	35min	32
2-4	0.3	8	22.5	35	31.25	30min	38
2-5	0.3	6	22.5	25	31.25	1hr 56min	88
2-6	0.3	4	22.5	25	31.25	2hr 24min	36
2-7	0.15	6	30	25	31.25	3hr 25min	97



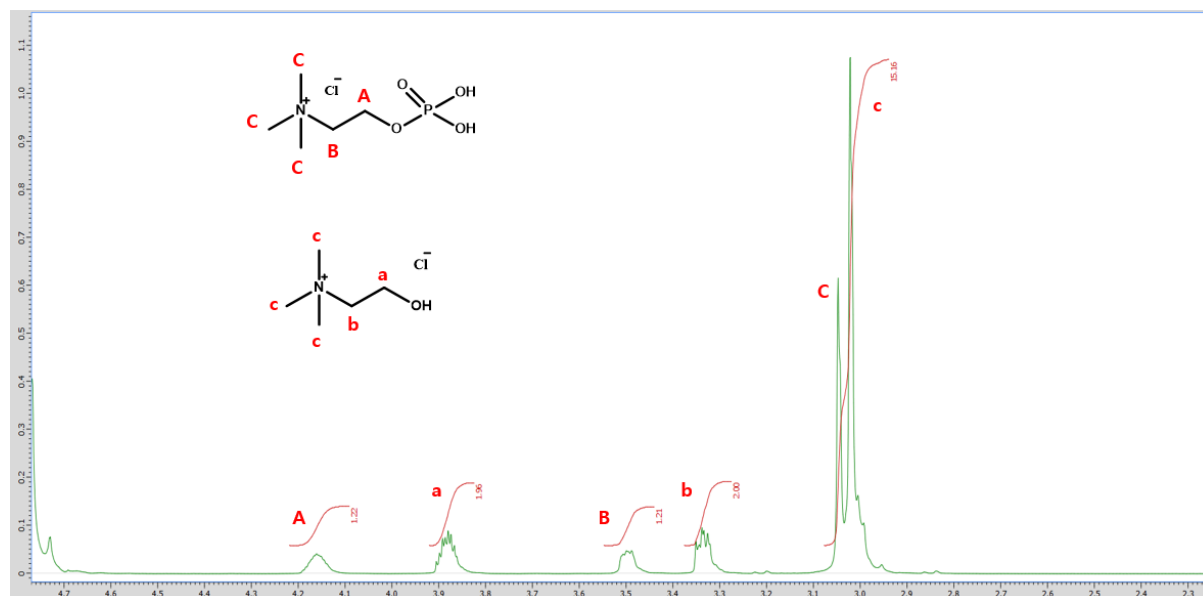
**Figure S4.** <sup>1</sup>H NMR spectrum of entry 2-1.



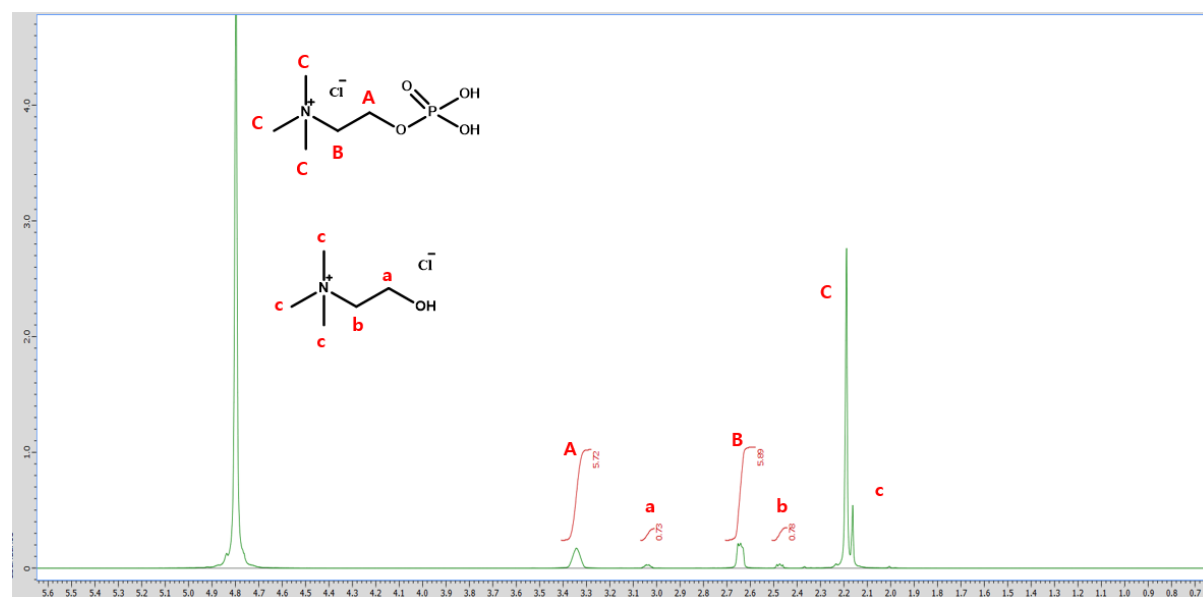
**Figure S5.** <sup>1</sup>H NMR spectrum of entry 2-2.



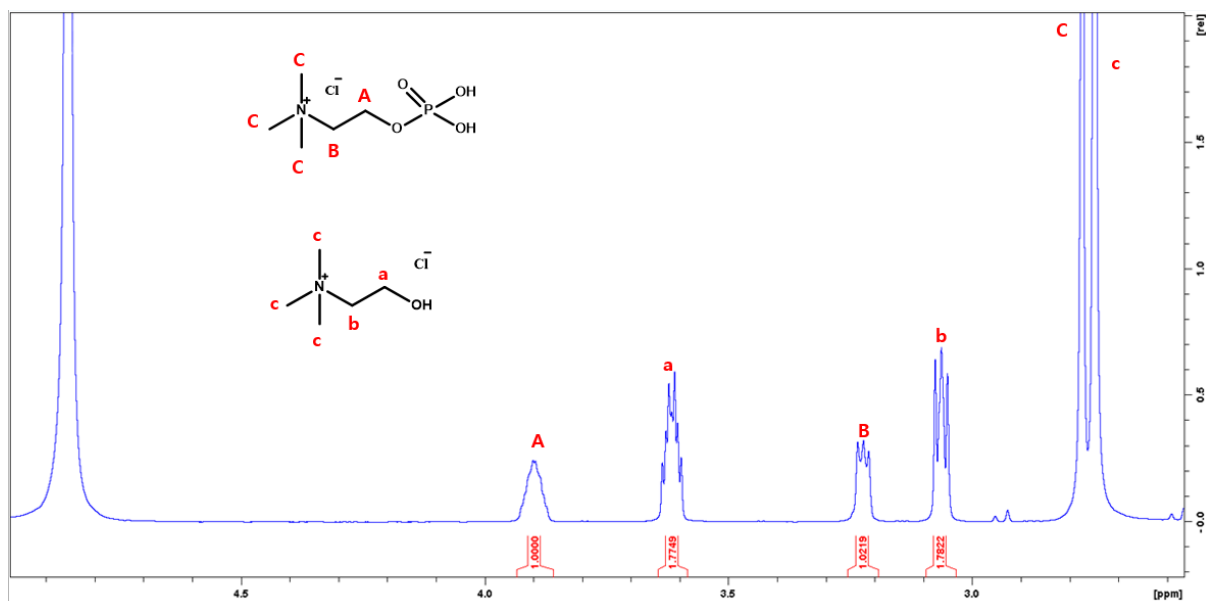
**Figure S6.** <sup>1</sup>H NMR spectrum of entry 2-3.



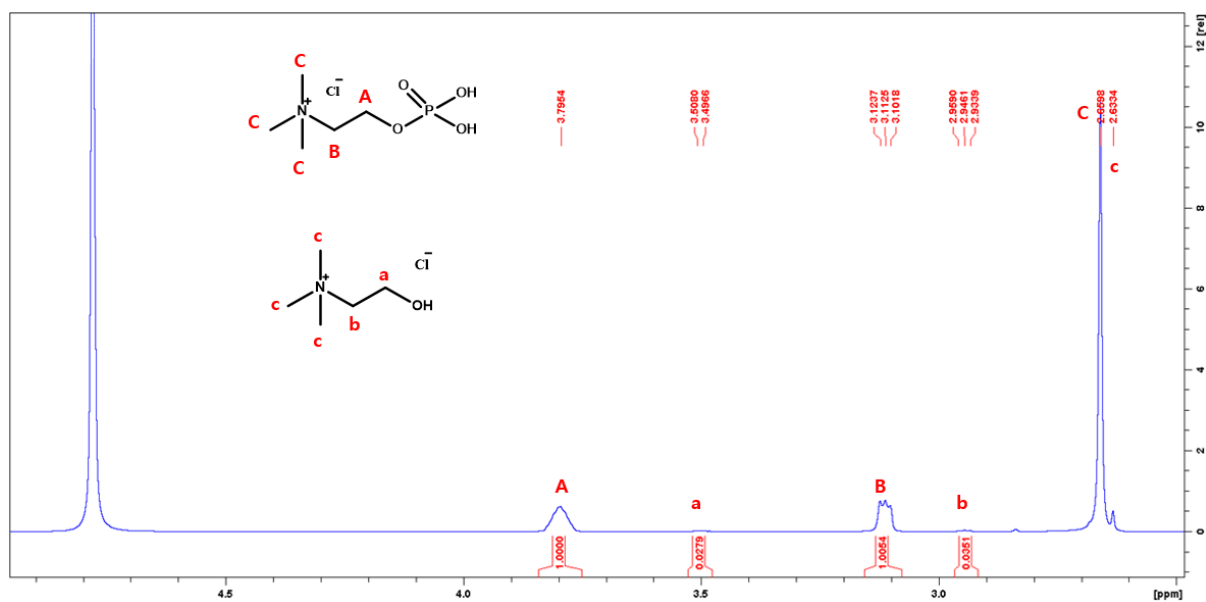
**Figure S7.**  $^1\text{H}$  NMR spectrum of entry 2-4.



**Figure S8.**  $^1\text{H}$  NMR spectrum of entry 2-5.



**Figure S9.**  $^1\text{H}$  NMR spectrum of entry 2-6.

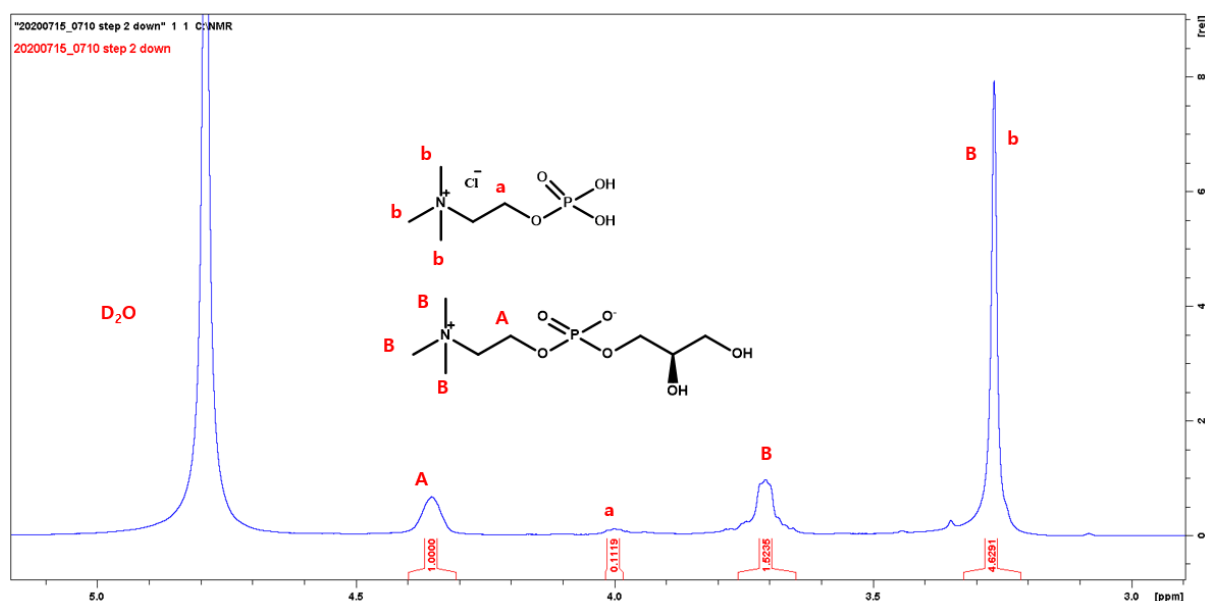


**Figure S10.**  $^1\text{H}$  NMR spectrum of entry 2-7.

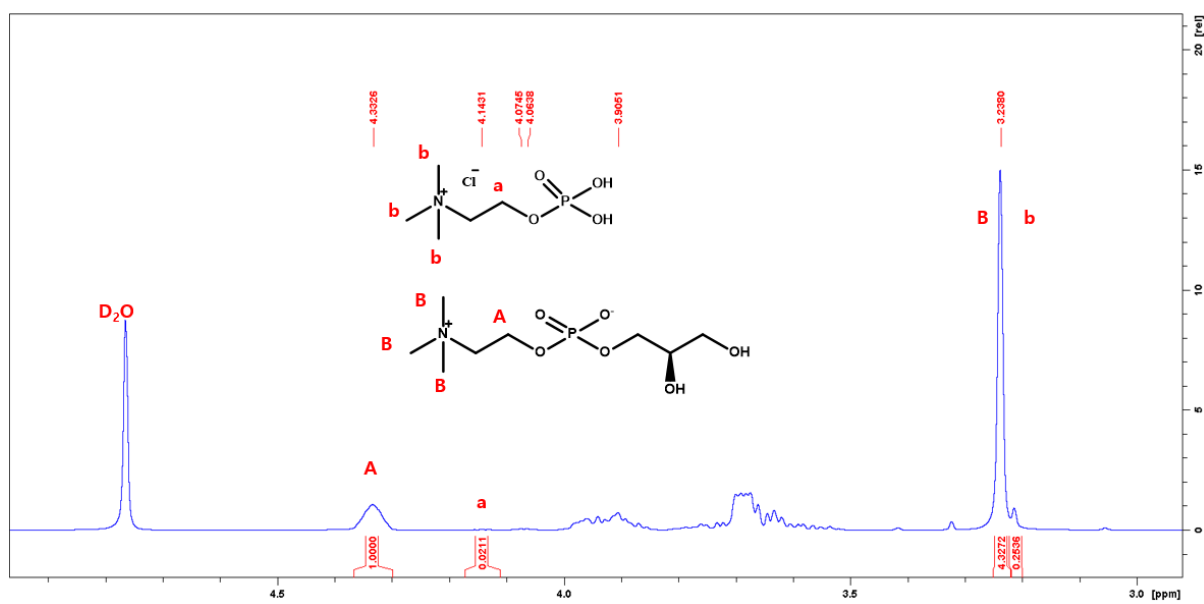


**Table S3.** Reaction conditions and conversions of second step reactions in a batch reactor.

entry	solvent	CPD/ phosphorylcholine	temperature (°C)	reaction time	conversion (%)
3-1	ethanol	1.14	75	24hr	90
3-2	H <sub>2</sub> O + ethanol	1.14	75	24hr	98



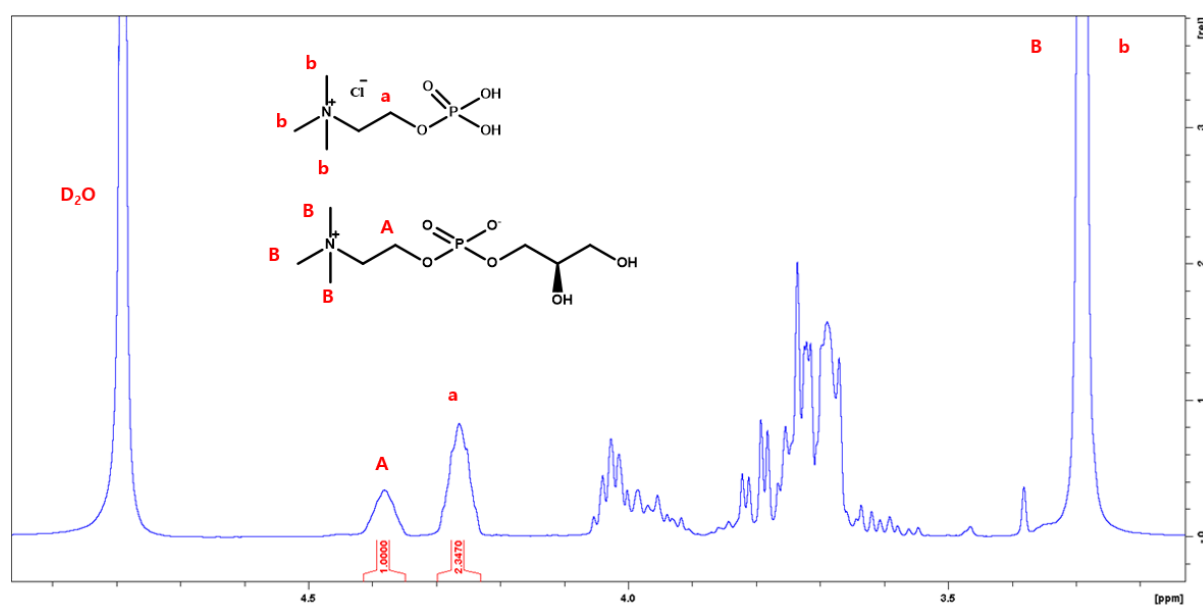
**Figure S11.** <sup>1</sup>H NMR spectrum of entry 3-1.



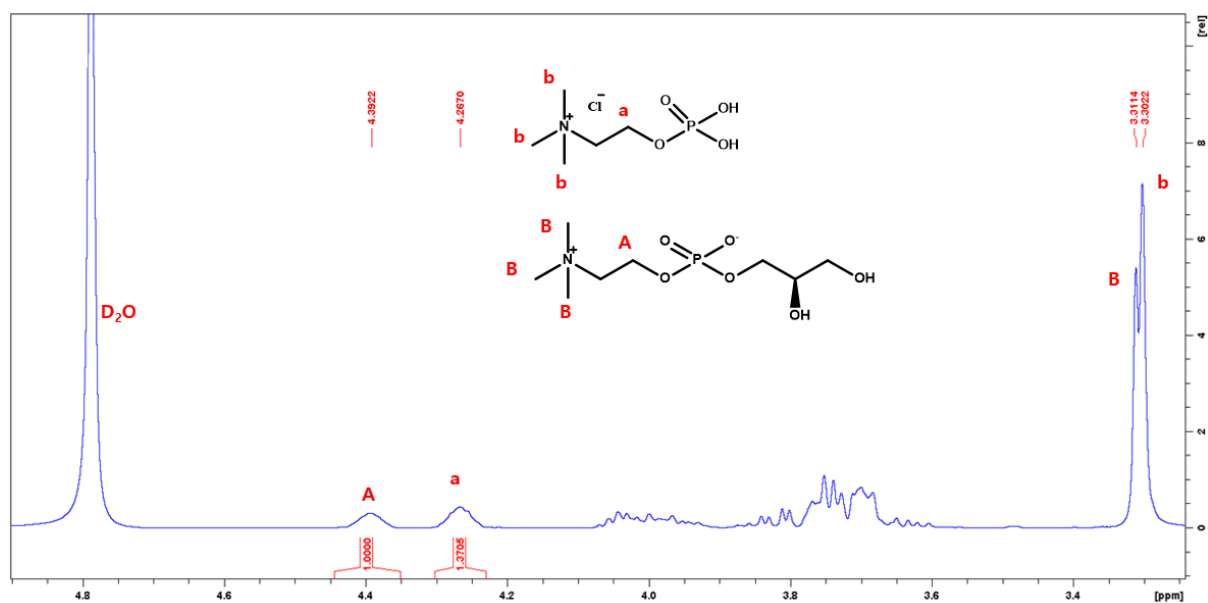
**Figure S12.** <sup>1</sup>H NMR spectrum of entry 3-2.

**Table S4.** Reaction conditions and conversions of second step reactions in a flow reactor.

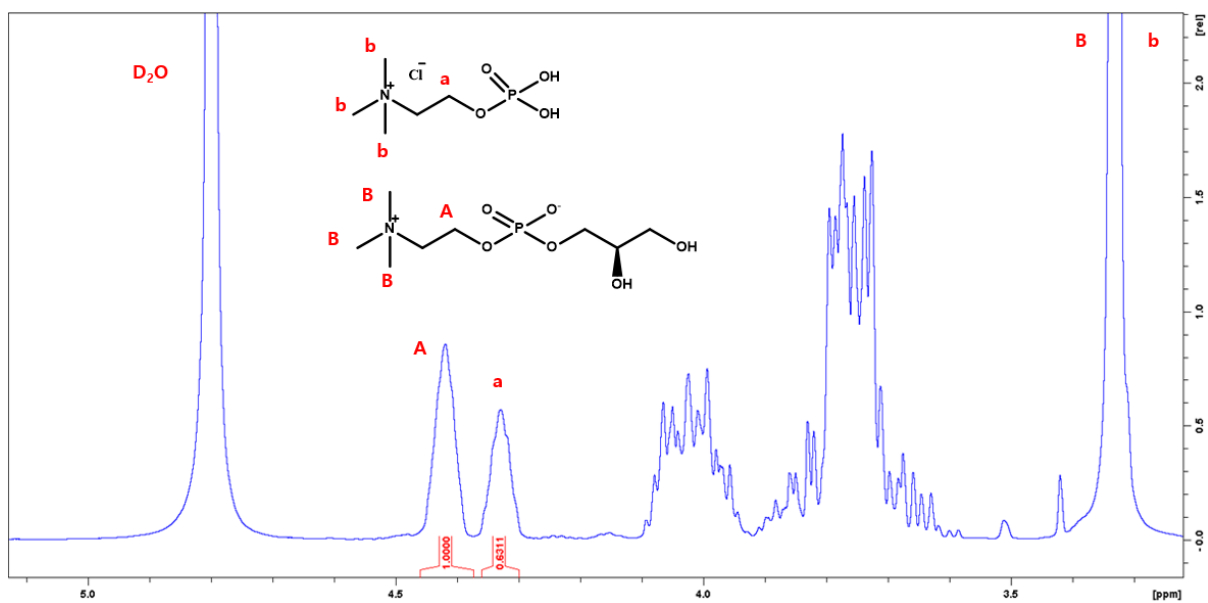
entry	H <sub>2</sub> O (mL)	reactor volume (mL)	temperature (°C)	reaction time	Conversion (%)
4-1	15	12	75	6hr	30
4-2	12	12	75	6hr	42
4-3	10	12	75	6hr	61
4-4	10	12	80	6hr	79



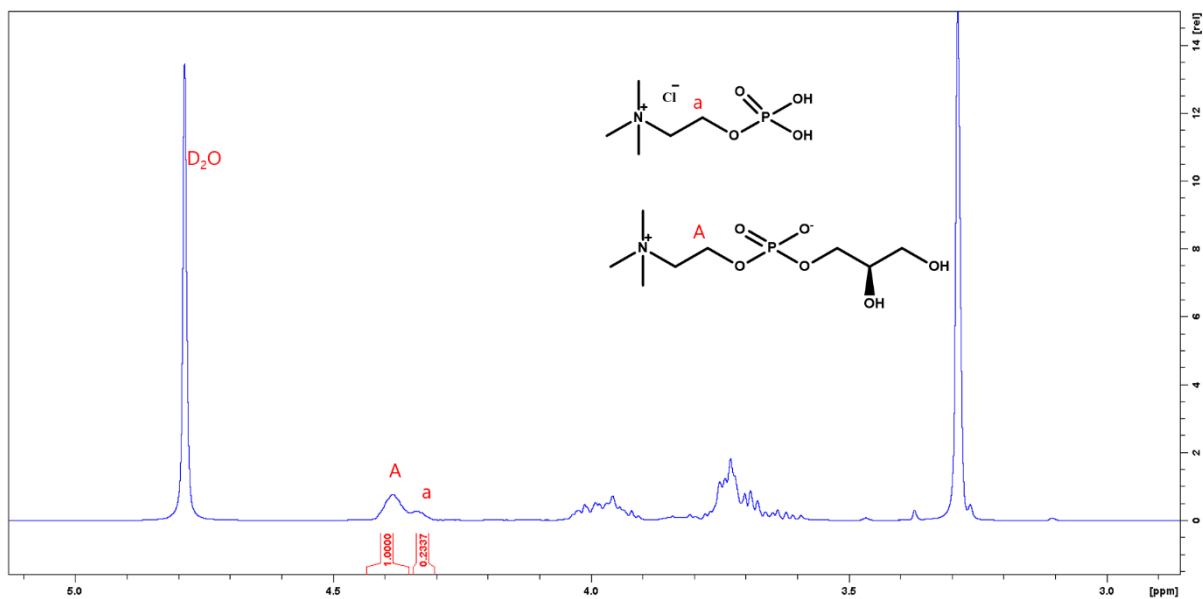
**Figure S13.** <sup>1</sup>H NMR spectrum of entry 4-1.



**Figure S14.**  $^1\text{H}$  NMR spectrum of entry 4-2.



**Figure S15.**  $^1\text{H}$  NMR spectrum of entry 4-3.



**Figure S16.**  $^1\text{H}$  NMR spectrum of entry 4-4.

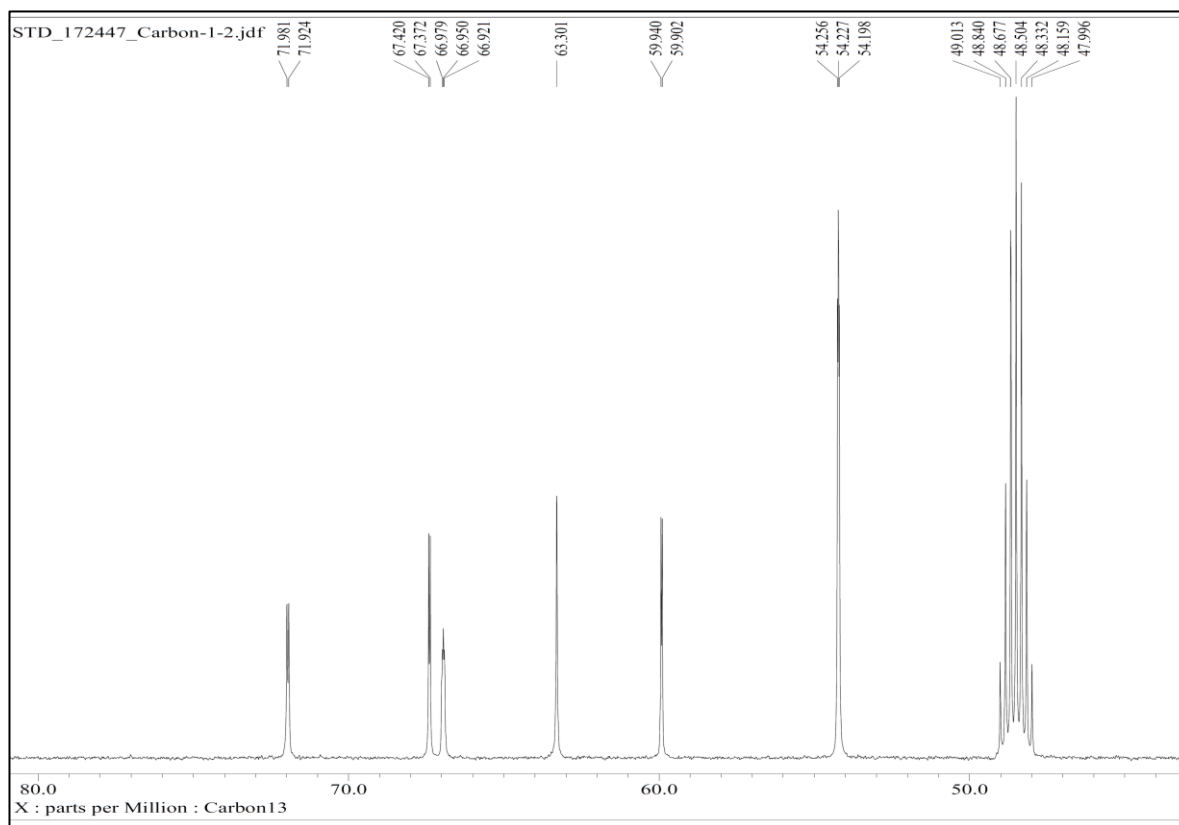
**Table S5.** Reproducibility experiment of first step reactions in a flow reactor.

entry	$\text{H}_2\text{O}$ <sup>a</sup> (mL)	$\text{POCl}_3$ / choline chloride <sup>b</sup>	reactor volume (mL)	temperature ( $^\circ\text{C}$ )	flow rate ( $\mu\text{L}/\text{min}$ )	reactn time	Conversion <sup>c</sup> (%)
1	0.15	6	30	25	31.25	3hr 25min	97
2	0.15	6	30	25	31.25	3hr 13min	95.2
3	0.15	6	30	25	31.25	3hr 26min	97.3

**Table S6.** Reproducibility experiment of second step reactions in a flow reactor.

entry	$\text{H}_2\text{O}$ (mL)	reactor volume (mL)	temperature ( $^\circ\text{C}$ )	reaction time	Conversion (%)
1	10	12	80	6hr	79
2	10	12	80	6hr	76
3	10	12	80	6hr	78

# <sup>13</sup>C NMR



**Figure S17.** <sup>13</sup>C NMR spectrum of choline alfoscerate.

## Mass spectrum

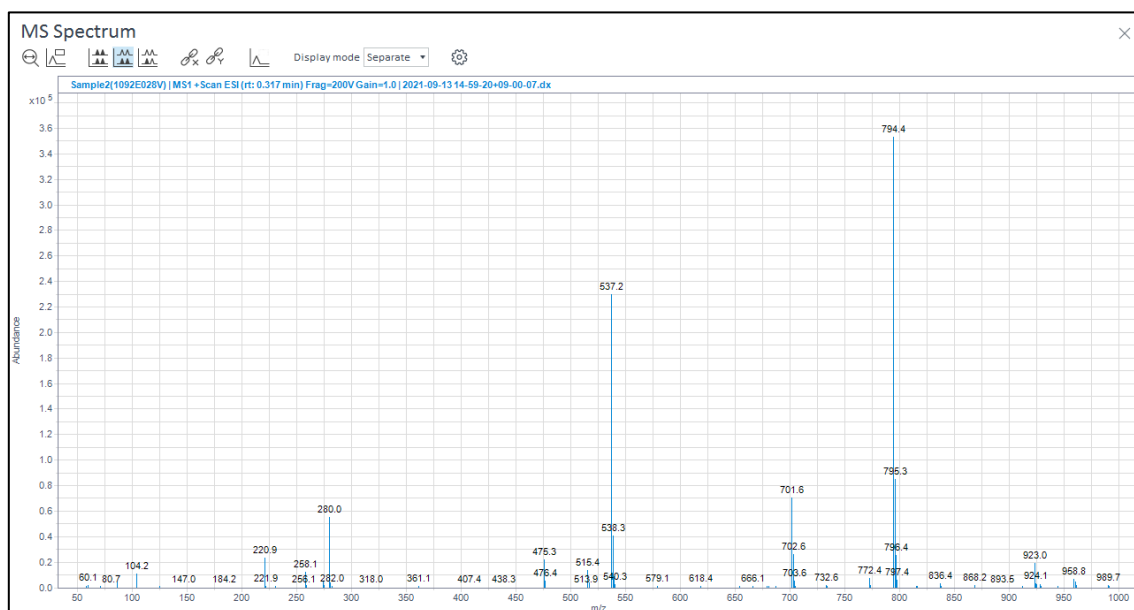


Figure S18. Mass spectrum of choline alfoscerate.

## IR spectrum

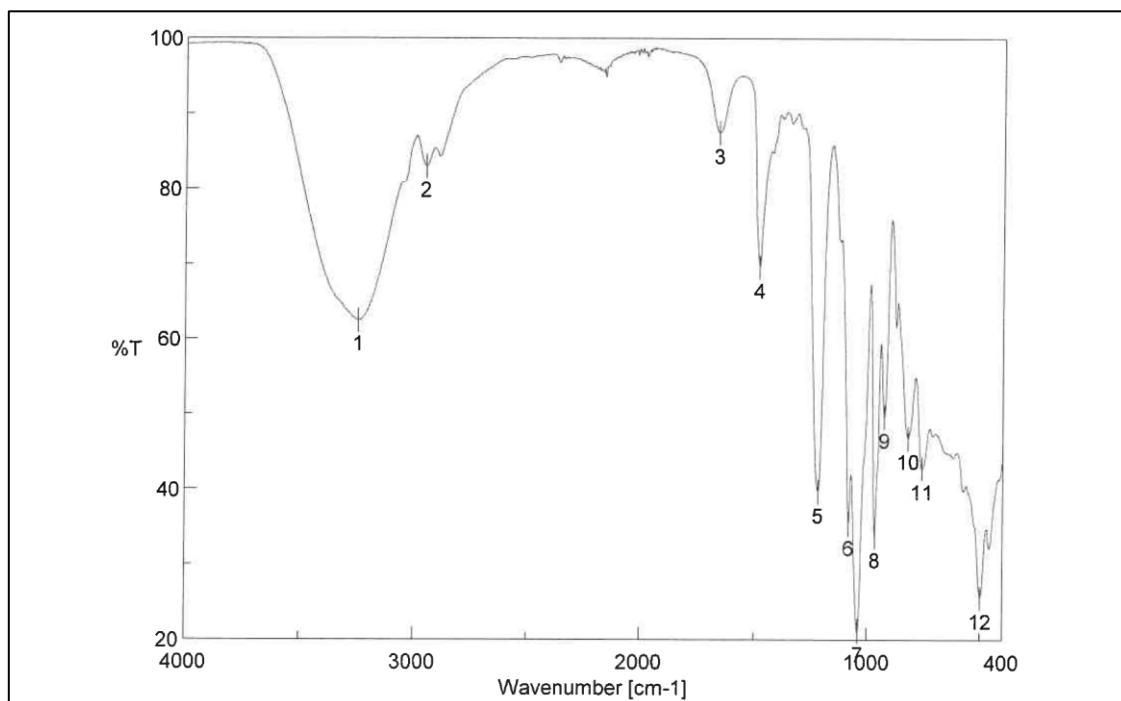


Figure S19. IR spectrum of choline alfoscerate.

## HPLC data

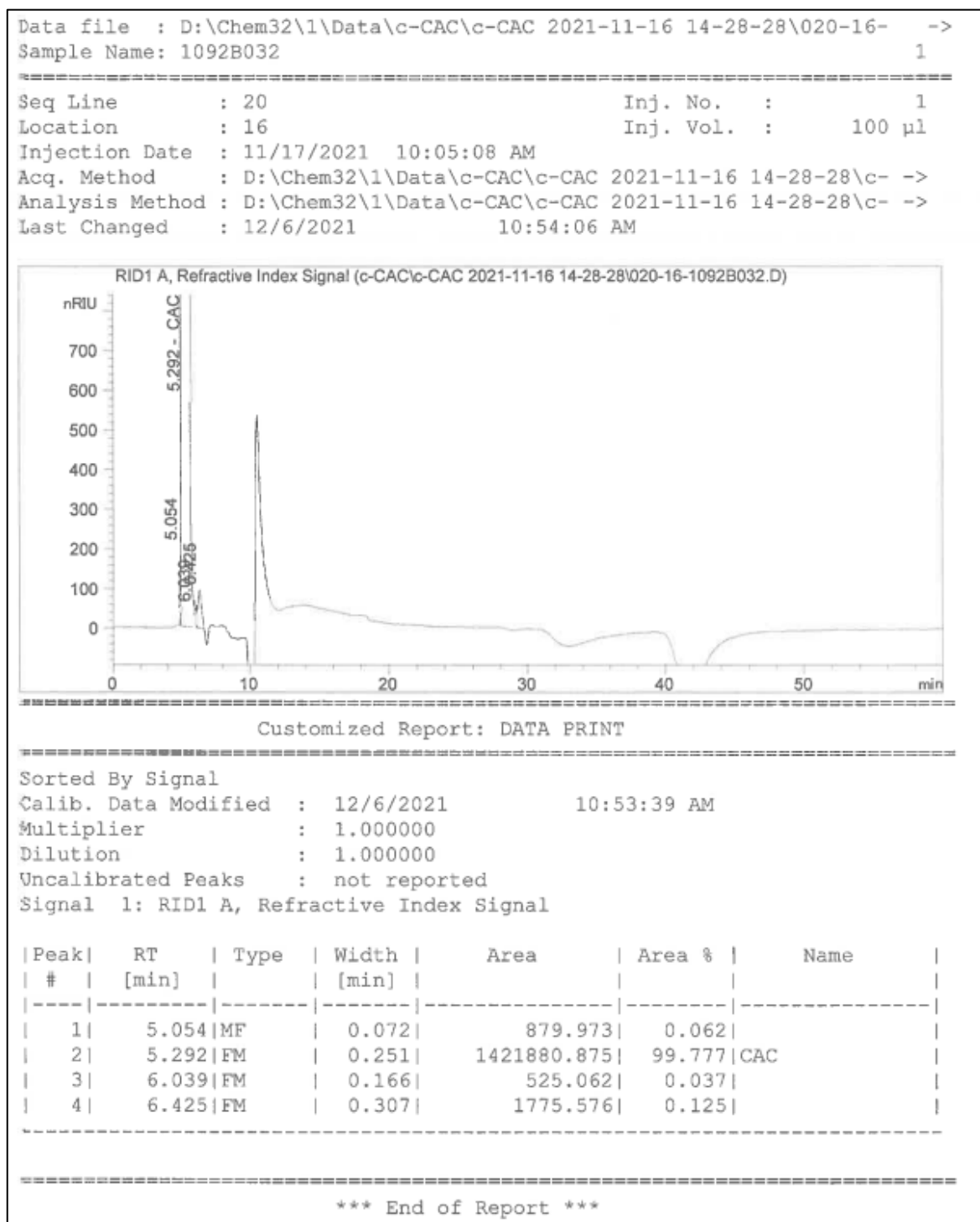


Figure S20. HPLC graph of choline alfoscerate.