

ORGANIC ACIDS

Occurrence in food:

**biochemical processes, acidulants, stabilizers,
preservation substances, microbial activity**

Typical sensory characteristics (taste and flavour):

sour, hot, saltybitter, buttery etc.

Typical chemical characteristics:

**chain length, cyclic and aromatic structures
number of carboxylic groups, other groups (-OH, =O)
(un-) and saturated - cis, trans izomers etc.
volatility, dissociation constant**

Classification of organic acids (I)

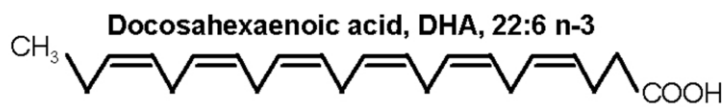
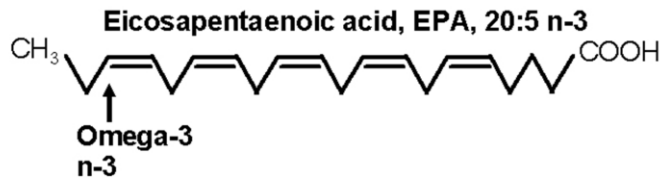
1. Alifatic saturated monocarboxylic acids

H - COOH	methanoic	<i>formic</i>	methanoyl <i>formyl</i>	methanoate <i>formate</i>
CH ₃ - COOH	ethanoic	<i>acetic</i>	ethanoyl <i>acetyl</i>	ethanoate <i>acetate</i>
C ₂ H ₅ - COOH	propanoic	<i>propionic</i>	propanoyl <i>propionyl</i>	propanoate <i>propionate</i>
CH ₃ [CH ₂] ₂ - COOH	butanoic	<i>butyric</i>	butanoyl <i>butyryl</i>	butanoate <i>butyrate</i>
CH ₃ [CH ₂] ₃ - COOH	pentanoic	valeric	<i>pentanoyl</i>	<i>pentanoate</i>
CH ₃ [CH ₂] ₄ - COOH	hexanoic	kaproic	<i>hexanoyl</i>	<i>hexanoate</i>
CH ₃ [CH ₂] ₁₀ - COOH	dodekanoic	lauric	<i>dodekanoyl</i>	<i>dodekanoate</i>
CH ₃ [CH ₂] ₁₄ - COOH	hexadekanoic	palmitic	<i>palmitoyl</i>	<i>palmitoate</i>
CH ₃ (CH ₂) ₁₆ - COOH	oktadekanoic	stearic	<i>stearoyl</i>	<i>stearoate</i>

Classification of organic acids (II)

2. Alifatic unsaturated monocarboxylic acids

$C_{17}H_{33} - COOH$	octadec-9-enoic	<i>oleic</i>	<i>oleoyl</i>	<i>oleate</i>
$C_{17}H_{31} - COOH$	octadeca-9,12-dienoic	<i>linoleic</i>	<i>linoloyl</i>	<i>linolate</i>
$C_{17}H_{29} - COOH$	octadeca-9,12,15-trienoic	<i>linolenic</i>	<i>linolenoyl</i>	<i>linolenate</i>



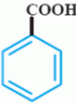
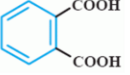
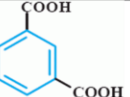

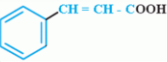
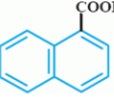
Classification of organic acids (III)

3. Alifatic dicarboxylic acids

HOOC - COOH	ethanedioic	<i>oxalic</i>	<i>oxalyl</i>	<i>oxalate</i>
			ethanedioyl	ethanedioate
HOOC - CH ₂ - COOH	propanedioic	<i>malonic</i>	<i>malonyl</i>	<i>malonát</i>
			propanedioyl	propandioate
HOOC - (CH ₂) ₂ - COOH	butanedioic	<i>succinic</i>	<i>succinyl</i>	<i>succinate</i>
			butanedioyl	butanedioate
HOOC - (CH ₂) ₃ - COOH	pentanedioic	<i>glutaric</i>	<i>glutaryl</i>	<i>glutarate</i>
			pentanedioyl	butandioate
HOOC - (CH ₂) ₄ - COOH	hexanedioic	<i>adipic</i>	<i>adipoyl</i>	<i>adipate</i>
			hexanedioyl	hexanedioate
	(Z) -butenedioic	<i>maleinic</i>	<i>maleinyl</i>	<i>maleinate</i>
			butenedioyl	butenedioate
	(E) -butenedioic	<i>fumaric</i>	<i>fumaroyl</i>	<i>fumarate</i>
			butenedioyl	butenedioate

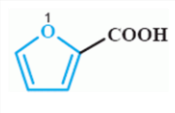
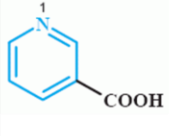
Classification of organic acids (IV)

4a. Aromatic carboxylic acids

	benzene-carboxylic	<i>benzoic</i>	<i>benzoyl</i>	<i>benzoate</i>
	benzene-1,2-dicarboxylic	<i>phthalic</i>	<i>phthalyl</i>	<i>phthalate</i>
	benzene-1,3-dicarboxylic	<i>iso-phthalic</i>	<i>isophthalyl</i>	<i>isophthalate</i>
	benzene-1,4-dicarboxylic	<i>terephthalic</i>	<i>terephthalyl</i>	<i>terephthalate</i>
	3-phenylpropenic	<i>cinnamic</i>	<i>3-phenylpropenyl</i>	<i>3-phenyl-propenoate</i>
	naphthalene-1-carboxylic	<i>1-naphtoic</i>	<i>1-naphtyl</i>	<i>1-naphtoate</i>

Classification of organic acids (V)

5. Heterocyclic carboxylic acids

	furane-2-carboxylic	<i>2-furoic</i>	<i>2-furoyl</i>	<i>2-furoate</i>
	pyridine-3-carboxylic	<i>nicotinic</i>	<i>nicotinoyl</i>	<i>nicotinate</i>

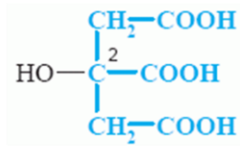
Classification of organic acids (VIa)

6a. Hydroxy acids

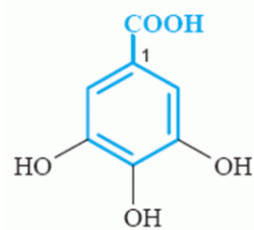
$\text{HO}-\text{CH}_2-\text{COOH}$	hydroxyethanoic hydroxyacetic	<i>glycolic</i>	<i>glycoloyl</i>
$\begin{array}{c} \text{CH}_3-\text{CH}-\text{COOH} \\ \\ \text{OH} \end{array}$	2-hydroxypropanoic	<i>lactic</i>	<i>lactoyl</i>
$\begin{array}{c} \text{CH}_2-\text{CH}-\text{COOH} \\ \quad \\ \text{OH} \quad \text{OH} \end{array}$	2,3-dihydroxypropanoic	<i>glyceric</i>	<i>glyceroyl</i>
$\begin{array}{c} \text{HOOC}-\text{CH}-\text{CH}_2-\text{COOH} \\ \\ \text{OH} \end{array}$	2-hydroxybutandioic hydroxysuccinic	<i>malic</i>	<i>maloyl</i>
$\begin{array}{c} \text{HO}-\text{CH}-\text{COOH} \\ \\ \text{HO}-\text{CH}-\text{COOH} \end{array}$	2,3-dihydroxybutandioic 2,3-dihydroxysuccinic	<i>tartaric</i>	<i>tartaroyl</i>

Classification of organic acids (VIb)

6b. Hydroxy acids



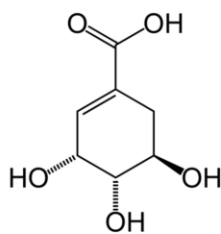
2-hydroxypropane-1,2,3-tricarboxylic *citric*



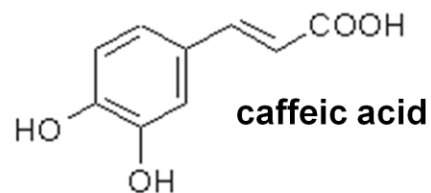
3,4,5-trihydroxybenzoic

gallic

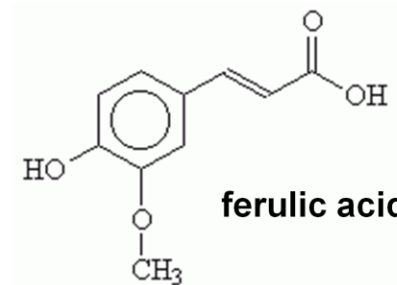
galloyl



shikimic acid



caffeic acid



ferulic acid

Classification of organic acids (VII)

7. Oxo acids

OHC - COOH

oxoethanoic

glyoxylic
oxalaldehydic

CH₃ - CO - COOH

2-oxopropanoic

pyruvic

Methods of analysis of organic acids (I)

Total acidity: titration – NaOH or KOH solution
indication – phenolphthalein or potentiometric
pH 8.2 - AOAC, pH 7.0 - EU

Expression: various according to matrix type - usually
consumption of titration solution of appropriate concentration
related to sample weight, reported as a dominant acid content

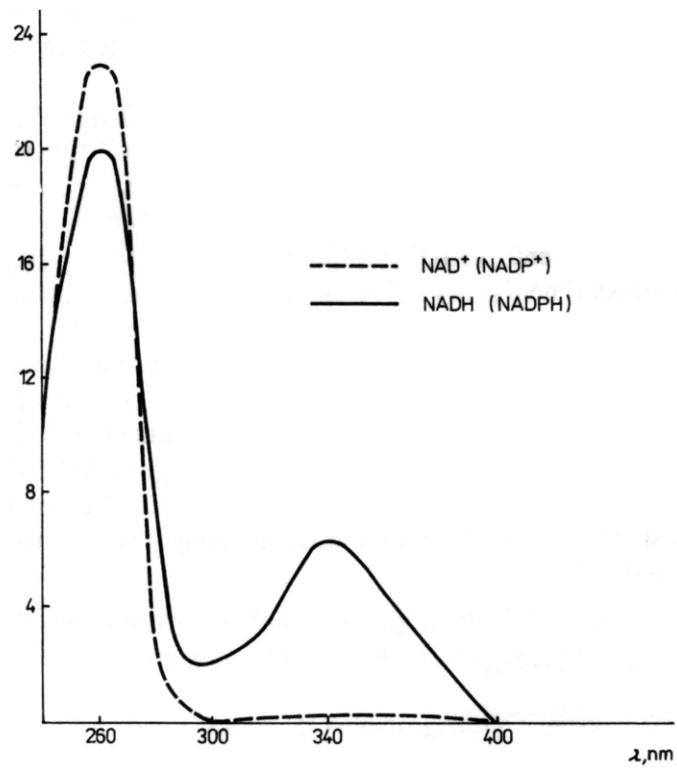
Volatile acids: distillation (possibly with water steam)

SPFM: directly (UV) or after reaction with agent (UV/VIS)
as a groups (unsaturated FA) or individually

Enzymatically: use of ratio NAD⁺/NADH or NADP⁺/NADPH
- measurement at 340 nm

Electroanalytically: redox titration or polarography (ascorbic a.)
possibly combination with enzyme electrode

Absorption spectra of various forms of pyridine dinucleotides



Methods of analysis of organic acids (II)

Separation methods

- 1. Chromatographic: gas chromatography – direct or
after derivatization
liquid chromatography - RP, IEC**
- 2. Elektromigration: electrophoresis, isotachopheresis**

Group or individual analysis

Separation of wide spectrum including minority components

Possibility of profile investigation

Methods of analysis of organic acids (III)

Analytical methods - examples

Analyte, matrix	Isolation	Determination
Lactic acid: wines	$\text{CuSO}_4 + \text{Ca(OH)}_2$ + H_2SO_4	SFPM - VIS 560 nm (reaction with p-hydroxydiphenyl)
dairy products	deproteination extraction - Et_2O oxidation to CH_3CHO	SPFM - VIS 570 nm (reaktion with piperidine)
Citric acid: cheeses	extraction with hot water (60-70°C) + deproteination CCl_3COOH	SPFM - VIS 428 nm (reaction with pyridine)

Methods of analysis of organic acids (IV)

Analytical methods - examples

<i>Analyte, matrix</i>	<i>Izolation</i>	<i>Determination</i>
Succinate: wines, fruit juices	dilution, pH 8.0 decolorization - 1% polyvinylpyrrolidone or activated carbon	SFPM - UV 340 nm (NADH)
meat, vegetable, cheeses	hot water extraction, deffating, cooling, deproteination, HClO₄	SFPM - UV 340 nm (NADH)

Methods of analysis of organic acids (V)

Analytical methods - examples

<i>Analyte, matrix</i>	<i>Izolation</i>	<i>Determination</i>
<i>Fumaric acid:</i> fruit juices	agitation with Celite + filtration	Polarographic method Peak potential: -1.15V
solid food	MeOH extraction + filtration	

Methods of analysis of organic acids (VI)

Analytical methods - examples

GC/FID,MS: stationary phase - volatility, functional groups

(derivatization: BF_3/MeOH , HMDS, TMCS)

- polysiloxanes, carbowax

- special: FFAP = polyethyleneglycol-2-nitroterephthalate

Sample treatment:

according to matrix type:

some drinks - dilution

some extracts, drinks - filtration, clarification, centrifugation

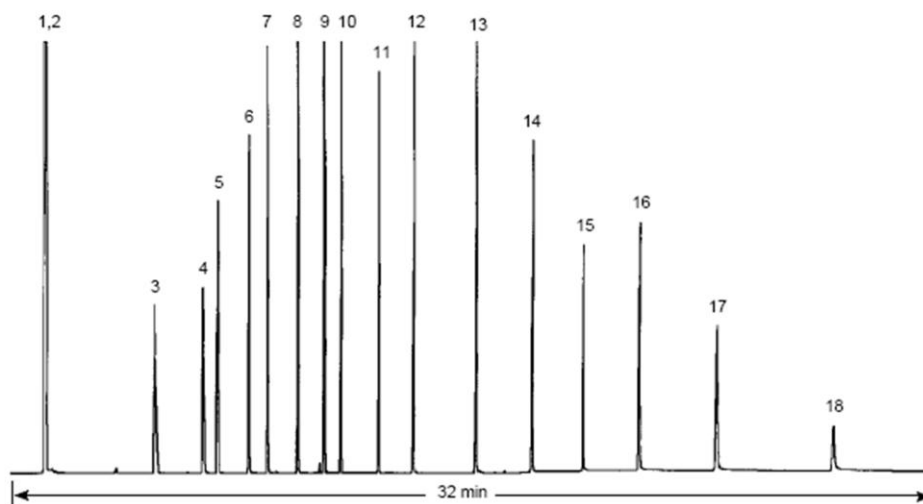
complex matrices, extracts - column chromatography, SPE etc.

Methods of analysis of organic acids (VIa)

Organic Acids

Column: DB-FFAP
30 m x 0.25 mm I.D., 0.25 μ m
J&W P/N: 122-3232
Carrier: Helium at 40 cm/sec, measured at 100°C
Oven: 100°C for 5 min
100-250°C at 10°/min
250°C for 12 min
Injector: Split 1:50, 250°C
Detector: FID, 300°C
Nitrogen makeup gas at 30 mL/min

- | | |
|----------------------------------|---|
| 1. Acetone | 11. Heptanoic acid |
| 2. Formic acid | 12. Octanoic acid |
| 3. Acetic acid | 13. Decanoic acid |
| 4. Propionic acid | 14. Dodecanoic acid |
| 5. Isobutyric acid | 15. Tetradecanoic acid |
| 6. Butyric acid | 16. Hexadecanoic acid |
| 7. Isovaleric acid | 17. Octadecanoic acid |
| 8. Valeric acid (pentanoic acid) | 18. Arachidic acid
(eicosanoic acid) |
| 9. Isocaproic acid | |
| 10. Caproic acid (hexanoic acid) | |



Methods of analysis of organic acids (VII)

Analytical methods - examples

HPLC/RID,EC,UV,MS

stationary phase: RP (C8, C18 silikagel), IEC (anex)

mobile phase: water, organic solvents, acids (H₂SO₄, HCOOH),
acidic buffers

Sample treatment: *(generally simpler compare to GC):*

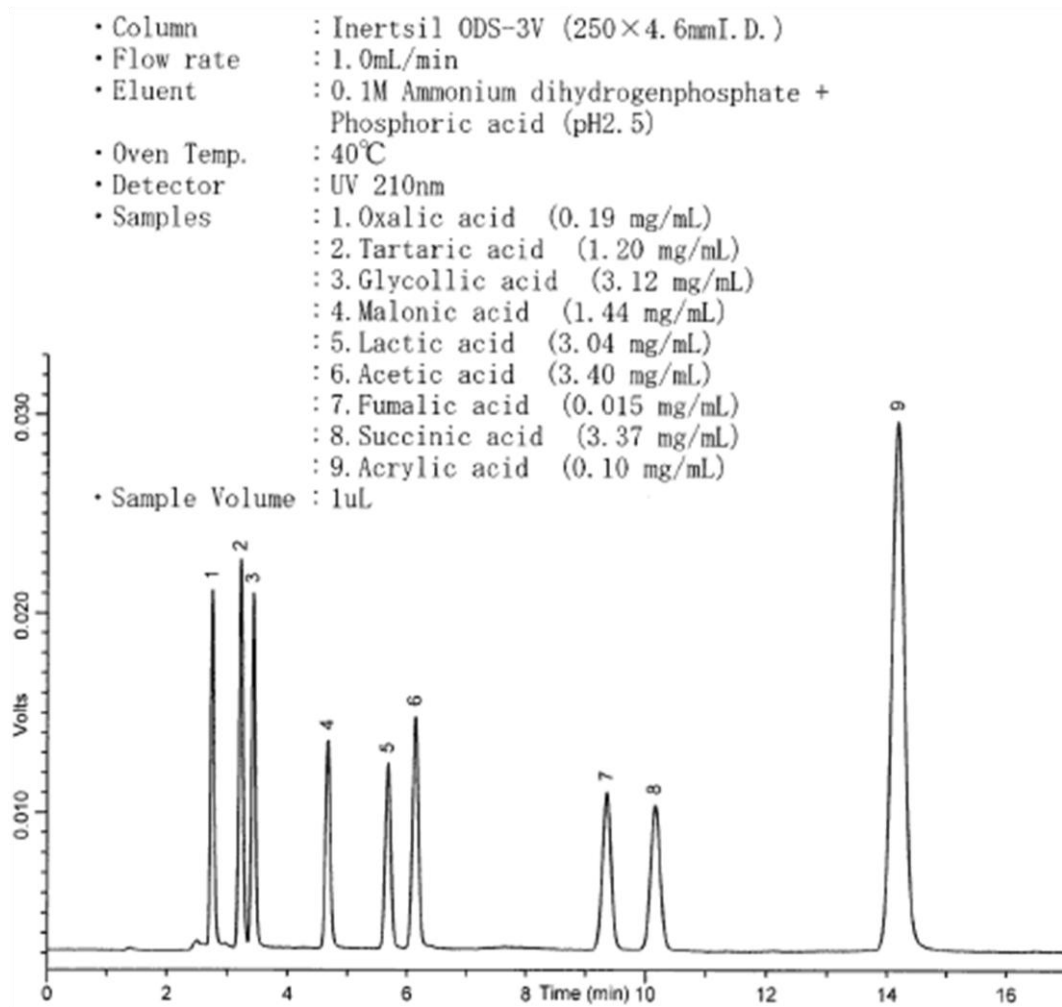
according to matrix type:

some drinks - dilution

some extracts, drinks - filtration, clarification, centrifugation

complex matrices, extracts - column chromatography, SPE etc.

Methods of analysis of organic acids (VIIa)

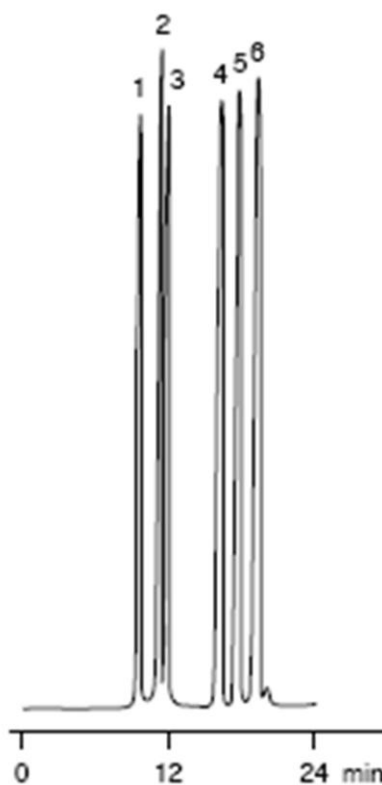


Methods of analysis of organic acids (VIIb)

Organic Acids

Column: Rezex ROA-Organic Acid
Dimensions: 300 x 7.8 mm
Part No.: 00H-0138-K0
Mobile Phase: 0.005 N Sulfuric Acid
Flow Rate: 0.5 mL/min
Detection: UV @ 210 nm
Temperature: 55 °C
Sample:

1. Oxalic
2. Citric
3. Tartaric
4. Succinic
5. Formic
6. Acetic



Methods of analysis of organic acids (VIII)

Analytical methods - examples

CZE, ITP/EC,UV

Sample treatment:

according to matrix type:

- dilution, fractionation, removal of interferences, filtration, clarification, centrifugation, column chromatography, SPE etc.

Methods of analysis of organic acids

(Villa)

Capillary: uncoated fused-silica (50 μm i.d. x 100 cm total length)

Cartridge and carousel temp: 25°C

Polarity: negative to positive

Run voltage: 25 kV

Run time: 30 min

Detector: 220 nm

Sample injection: 5 μs

Purge cycles (3): 0.1 M HCl (120s), BGE (240s)

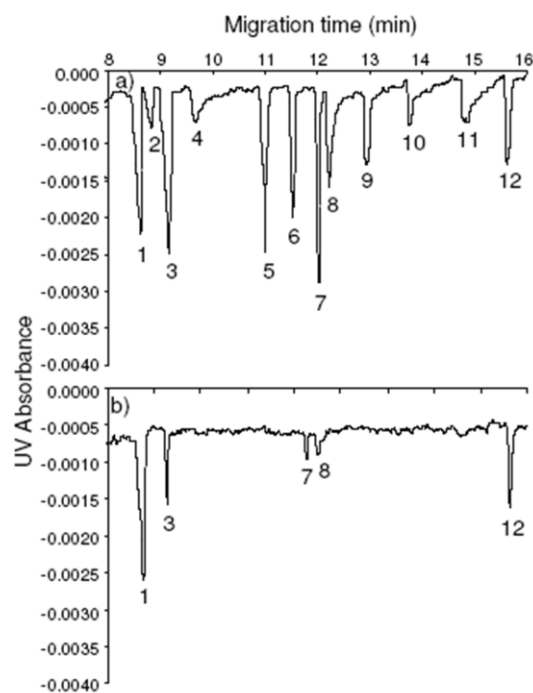
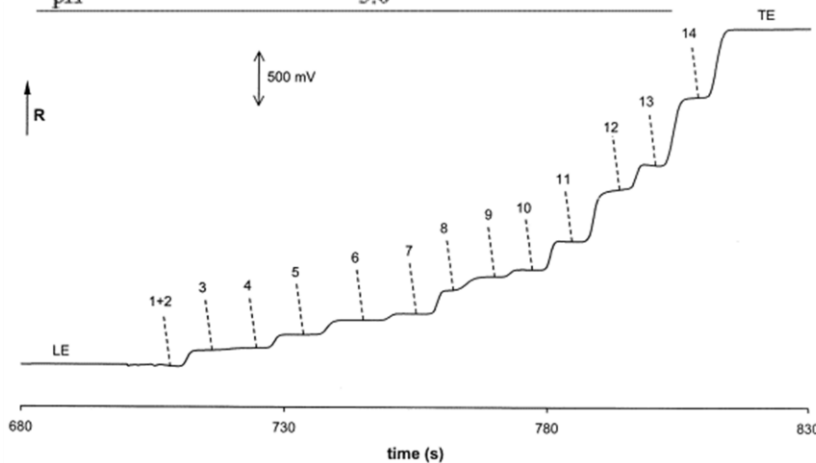


Figure 1 Inorganic anions and organic acids in (a) standard solution with each analyte at 1 mM, and (b) hot water extract of foliage from *P. pinaster*. Peaks are: 1, chloride; 2, nitrate; 3, sulphate; 4, oxalate; 5, formate; 6, tartarate; 7, malate; 8, citrate; 9, succinate; 10, pyruvate; 11, acetate; 12, phosphate. CE conditions as described in the text.

Methods of analysis of organic acids (VIIIb)

Parameter	
Solvent	Water
Leading ion	Chloride
Concentration (mM)	10
Counter-ion	β -Alanine
pH	2.9
Suppressor of electroosmotic flow	Methylhydroxyethylcellulose
Concentration (% w/v)	0.1
Terminating ion	Glutamate or capronate ^a
Concentration (mM)	5.0
Counter-ion	Histidine
pH	5.0



Isotachopheretic separation of organic acids and inorganic anions present in a model mixture on the CC chip. Zone assignments: LE, leading anion (chloride); 1*, migration position of sulphate; 2, sulphite; 3, phosphate; 4, malonate; 5, tartrate; 6, citrate; 7, malate; 8, lactate; 9, gluconate; 10, aspartate; 11, succinate; 12, ascorbate; 13, acetate; 14, sorbate; TE, terminating anion (capronate). The driving current was 10 μ A. The concentrations of the analytes in the injected model sample were 12–42 mg/l. R, increasing resistance.