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ORIGINAL PAPER

Degradation of the opium alkaloids in pharmaceutical relics from the eighteenth century

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Abstract

The opium alkaloids in three historical pharmaceutical relics dated to the eighteenth century were analyzed by HPLC–MS. The conditions of extraction of alkaloids from the sample and their HPLC analysis were optimized. A broad spectrum of opium alkaloids was found and degradation products of papaverine and noscapine were identified. The identity of compounds found was confirmed by MS/MS. For three compounds (methylhydrocotarnine, papaverinol, and papaveraldine) the ESI^+-MS/MS fragmentation mechanism has been proposed. The content of five selected opium alkaloids (cotarnine, meconin, morphine, papaverine, and noscapine) was determined in analyzed samples, and the concentration ratio of noscapine and cotarnine was proposed as a marker of the age of a preparation.

Graphic abstract

Keywords High-performance liquid chromatography · Decomposition · Opium · Mass spectrometry · Alkaloids

Introduction

Opium, the dried latex obtained by incision from the unripe capsules of poppy (*Papaver somniferum*), is one of the longest used drugs, notably as a potent analgesic (nowadays transformed into modern medicaments) [1, 2]. It is a rich mixture of more than 40 alkaloids, the most abundant are benzylisoquinoline alkaloids morphine, codeine, and thebaine [3, 4]. These alkaloids form about 25% of opium weight; the content of an individual alkaloid fluctuates: morphine is the most abundant (4–21%) followed by noscapine (2–8%), and papaverine (0.5–1.3%). The chemical structures of the main opium alkaloids discussed in this study are presented in Fig. 1.

The earliest archeological evidence of opium usage by mankind dates back to more than 5000 BC [5]. An organic residue analysis is employed to prove that an artefact contained opium, but the results are difficult to interpret as a result of degradation of opium alkaloids [6]. Base-ring juglets from the eastern Mediterranean, which shape resembles the capsule of the opium poppy, are the most studied objects in this context. But there are infrequent positive results for opium alkaloids in these juglets. In 1996, Koschel [7] analyzed a yellowish-brown residue from Ancient

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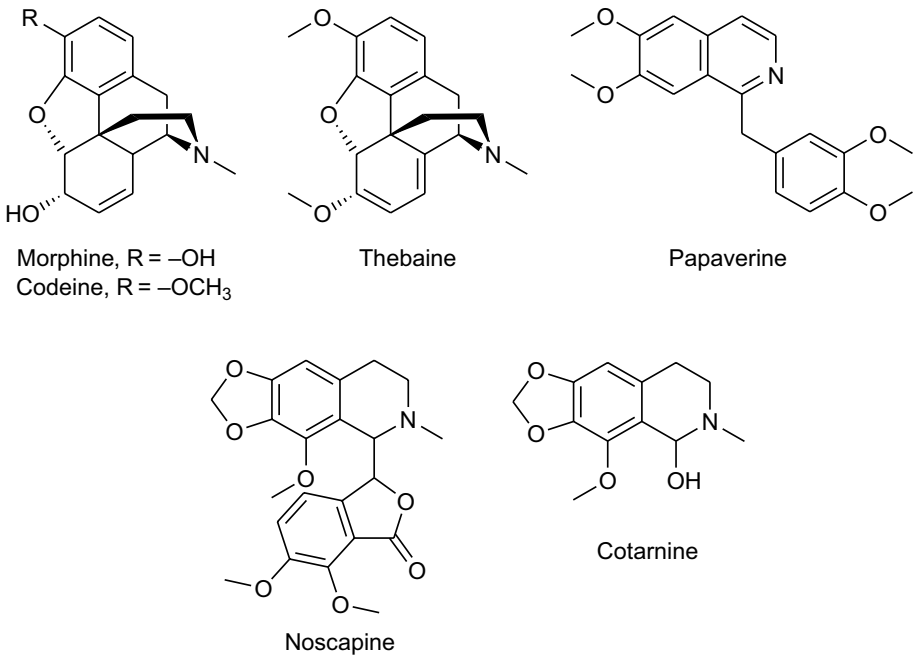

Fig. 1 Chemical structures of the main opium alkaloids discussed in this study

Egyptian juglet by TLC, GC–MS, and radioimmunoassay, and found 0.1% opium alkaloids (0.05% of morphine) and their oxidation products: oxidimorphine and apomorphine. However, the author warned that it is the result of a single analysis. On the other hand, Bisset et al. [8] using TLC, GC, or LC alone and combined with mass spectrometry, and radioimmunoassay methods found no morphine or tropane alkaloids (and hence no opium) in seven vessels from the tomb of the ancient Egyptian architect Kha (died about 1405 BC). Chovanec et al. [9] therefore proposed the model of degradation of opium alkaloids in artificially aged samples and compared results with historical relics. Using GC–MS, the authors show morphine does not preserve well. They suggest that papaverine, thebaine, and the degradation products of noscapine (first of all cotarnine) should be targeted in analyses. This suggestion is supported by extensive studies on the stability of morphine in pharmaceutical preparations, which show that degradation of morphine is accelerated in the presence of oxygen and at higher pH of the solution, whereas temperature and light have only a minor influence on the degradation rate [10]. Recently, Smith et al. [11] presented method for extracting poppy alkaloids from residues associated with archaeological ceramics based on removing the lipids by extraction with dichloromethane followed by C18 solid-phase extraction of alkaloids and subsequent HPLC–MS analysis. Only papaverine and thebaine were detected in the examined residue from a Late Bronze Age Cypriot juglet. The authors furthermore performed artificial ageing experiments with poppyseed oil under various conditions and found that morphine and codeine degraded as well. The other historical medical preparations, where opium alkaloids were analyzed, are residues of patent medicines from the end of the nineteenth century [12] and injection solutions from the first half of the twentieth century [13].

In this paper, we present the results of the analysis of three historical pharmaceutical relics of opium preparations from the eighteenth century. Analysis of opium alkaloids in artefacts older than 200 years has not been described yet. The main objection is to focus on possible degradation products of opium alkaloids and to quantitate selected opium alkaloids. The analyzed relics are from extraordinarily preserved baroque pharmacy of Capuchin order in Prague [14]. The pharmacy was established in 1680 and it was operated to the end of eighteenth century, after that it was completely untouched to the year 1895 when it was donated to National Museum in Prague. Thanks to this lucky accident the pharmacy equipment is preserved in a high baroque state, including period medical preparations. Although in the past medical preparations were not as strictly standardized as they are today, it is known that pharmacists in the mentioned pharmacy were using recipes from pharmacopoeias of that time, particularly from *Dispensatorium pharmaceuticum Austriaco-Viennense*, i.e., Austrian pharmacopoeia issued in 1729 [15].

We choose three opium-containing medical preparations from preserved equipment of the mentioned Capuchin pharmacy. All of them are older than 200 years. The first sample, further referred as 'Sample I' (Fig. 2a), is a relic of the famous 'Laudanum liquidum Sydenhamii', i.e., tincture of opium formulated by Thomas Sydenham [16–18]. It was prepared by dissolving opium in Spanish wine spiced with cinnamon (bark of *Cinnamomum verum*), cloves (dried

Fig. 2 Baroque pharmaceutical jars provided **a** Sample I, tincture of opium according Thomas Sydenham (National Museum, inv. no. H2-4372), **b** Sample II, celestial theriac (National Museum, inv. no. H2-9576), **c** Sample III, anodyne powder (National museum, inv. no. H2-9637)

flower buds of *Eugenia caryophyllata*), and saffron (*Crocus sativus*); the content of opium in the preparation is about 14%. It was used as a potent analgesic.

The second analyzed sample, further referred as 'Sample II', is a relic of preparation called 'Theriaca Caelestis' (Fig. 2b), i.e., celestial theriac. It is rich mixture of opium with numerous herbal and animal drugs (there is a great variability in the recipes [19]); the content of opium in the preparation is roughly about 5%. The preparation was used as an analgesic and diaphoretic medicine.

The third analyzed sample, further referred as 'Sample III', is a relic of preparation called 'Pulvis anodynus' (Fig. 2c), which translated means 'Anodyne powder'. It was prepared by mixing of poppy seed with numerous herbal and animal drugs and sugar [15, 19]. The powder was used as a mild analgesic.

As a reference material we used standard pharmaceutical substance of opium. As opium is hardly available, we used pharmaceutical substance of opium from 1950 with declared content of morphine 9.8%.

Analyzing these samples we have not only confirmed the authenticity of the content of historical pharmaceutical jars, but also studied degradation of opium alkaloids, and compare the results obtained with the ones described in the literature. Moreover, an optimization of HPLC method and the ESI^+-MS/MS fragmentation mechanisms of several opium alkaloids are presented.

Results and discussion

Optimization of sample preparation and HPLC separation

Acetonitrile and methanol were chosen as solvents for extraction of opium alkaloids from the samples, as the opium alkaloids are easily soluble in the organic solvents [3, 20]. Using standard pharmaceutical substance of opium with known morphine content, we tested extraction of alkaloids from the samples by acetonitrile or methanol alone, in various mutual mixtures, and by their mixtures with water. The best result is provided by treating the sample with 200-fold volume of acetonitrile–water (85:15, v/v) mixture; the yield of extraction of alkaloids is $97.2 \pm 4.5\%$.

High-performance liquid chromatography methods for separation and determination of opium alkaloids, which are performed mainly in reverse-phase mode employing various mobile phases based on aqueous buffers and acetonitrile or methanol, were reviewed several times, e.g., [21, 22]. With respect to compatibility of separation with mass spectrometric detection we have adopted the method developed by Bogusz et al. [23] based on RP C18 column and mobile phase consisting of ammonium formate aqueous buffer and acetonitrile. To improve the HPLC separation of opium alkaloids in the samples we tested and optimized: (1) replacement of acetonitrile by methanol, (2) replacement of ammonium formate aqueous buffer by ammonium acetate aqueous buffer, (3) the value of pH of buffer in the range 3.00–6.00, and (4) various profiles of gradient program. Finally, the optimized gradient elution with binary mobile phase of methanol (solvent A) and 0.01 M ammonium acetate buffer pH = 3.00 (solvent B) was used, starting with 10% A which is maintained constant for 4 min, increased to 80% within 10 min, maintained constant for 2 min, returned to 10% within 1 min, and finally maintained constant for 13 min. The total time of analysis was 30 min.

Application to historical samples

The optimized extraction and separation procedures were applied to the historical samples (Fig. 3). The identification of compounds was based on the comparison with standards (for morphine, cotarnine, meconin, papaverine, and noscapine) and simultaneously on the high-resolution tandem mass

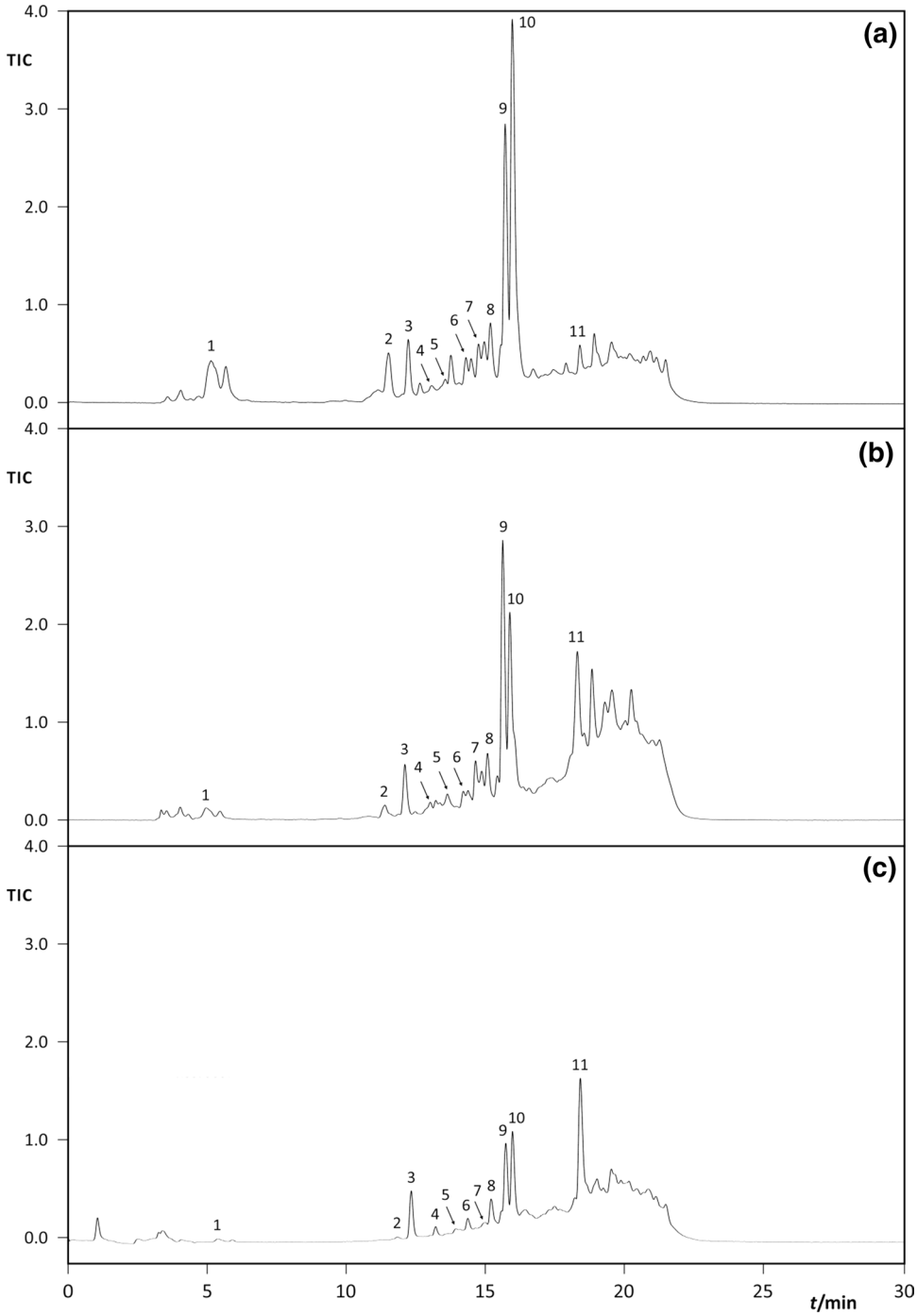

Fig. 3 HPLC–MS chromatograms of **a** Sample I, tincture of opium according Thomas Sydenham (National Museum, inv. no. H2-4372), **b** Sample II, celestial theriac (National Museum, inv. no. H2-9576), **c** Sample III, anodyne powder (National museum, inv. no. H2-9637). For peak identification see Table 1. XBridge® BEH C18 column (150 × 3.0 mm i.d., particle size 2.5 µm; Waters), mobile phase: methanol and 0.01 M ammonium acetate (pH = 3.00), gradient elution (for details see section *Optimization of sample preparation and HPLC separation*), flow rate 0.2 cm^3 min^{-1}

spectrometry (Table 1). The MS/MS was used and fragmentation spectra were compared with those published in literature or analyzed, as stated in Table 1.

The obtained results show that in addition to naturally occurring opium alkaloids, the decomposition products of these alkaloids were detected in all three analyzed samples. The majority of degradation products found originate from decomposition of papaverine and noscapine. In the case of papaverine, its oxidation products papaverinol and papaveraldine were identified, which is in accordance with the literature [24]. The degradation of noscapine leads to formation of cotarnine, hydrocotarnine, methylhydrocotarnine, and meconin, which is similar to its oxidative metabolism described by Tsunoda and Yoshimura [25]. The presence of above-mentioned degradation products of papaverine and noscapine (with the exception of meconin) was not detected in standard pharmaceutical substance of opium used as reference material.

Table 1 Identification of the opium alkaloids in the chromatograms of analyzed historical samples (retention time in HPLC, formula of $[M+H]^+$ ion and its *m/z*, diagnostic ions, identity and CASRN of the substance, and reference to mass spectrum used for confirmation of the substance

Peak number	t_r/min	Formula $[M+H]^+$	*m/z*			Diagnostic ions *m/z*	Identity, CASRN	References
			Experimental	Theoretical	Δ/ppm			
1	5.12	$C_{17}H_{20}NO_3$	286.1439	286.1437	0.1	268, 229, 201	Morphine, 57-27-2	[33, 39]
2	11.38	$C_{18}H_{22}NO_3$	300.1596	300.1594	−0.2	181, 165, 153	Codeine, 76-57-3	[33, 40]
3	12.20	$C_{12}H_{14}NO_3$	220.0968	220.0968	−0.1	205, 176, 148, 118	Dehydrogen hydrocotarnine, 57991-02-3	[41]
4	13.18	$C_{13}H_{18}NO_3$	236.1281	236.1281	−0.1	–[a]	Methylhydrocotarnine, 109506-42-5	–
5	13.50	$C_{12}H_{16}NO_3$	222.1122	222.1122	−0.1	205, 178, 163	Hydrocotarnine, 550-10-7	[26]
6	14.33	$C_{12}H_{16}NO_4$	238.1074	238.1074	−0.1	220, 205, 203	Cotarnine, 82-54-2	[26]
7	14.69	$C_{19}H_{22}NO_3$	312.1595	312.1594	−0.3	255, 266, 249	Thebaine, 115-37-7	[33, 39]
7	15.14	$C_{10}H_{12}O_4$	195.0657	195.0189	2.6	194, 177, 165, 147	Meconin, 569-31-3	[42, 43]
8	15.14	$C_{20}H_{22}NO_5$	356.1492	356.1493	0.1	–[a]	Papaverinol, 482-76-8	–
9	15.68	$C_{20}H_{22}NO_4$	340.1543	340.15433	0.1	324, 296, 202, 171	Papaverine, 58-74-2	[34]
10	15.93	$C_{22}H_{24}NO_7$	414.1550	414.1547	0.8	353, 323, 205	Noscapine, 128-62-1	[44]
11	18.33	$C_{20}H_{20}NO_5$	354.1334	354.1336	0.5	–[a]	Papaveraldine, 522-57-6	–

[a]Diagnostic ions are not described in literature, so the spectrum was analyzed in detail, see section "Mass spectrometry of selected alkaloids"

Mass spectrometry of selected alkaloids

The MS/MS was employed to confirm the identity of methylhydrocotarnine, papaverinol, and papaveraldine, as their mass spectra are inadequately described in the literature. Therefore their ESI^+-MS/MS spectra have been studied in detail and corresponding fragmentation pathways been proposed.

For methylhydrocotarnine, the ion *m/z* = 236.1284 was selected as the precursor ion, its MS^2 spectrum is given in Fig. 4a. The mechanism of methylhydrocotarnine ESI^+ fragmentation is proposed in Fig. 5. As methylhydrocotarnine is derivative of cotarnine, hydrocotarnine resp., majority of fragmentation ions of methylhydrocotarnine are analogous to fragmentation ions of these two compounds, as described in [26–28]. Nevertheless, ions *m/z* = 174.0913 and 58.0651 could be used to distinguish mass spectrum of methylhydrocotarnine from mass spectra of cotarnine or hydrocotarnine.

The structural similarity of papaverinol and papaveraldine results to parallelism in their mass spectra (Fig. 4b and c). In case of papaverinol, only *m/z* of molecular ion has been published yet [29, 30]. Thus, the molecular ion *m/z* = 356.1492 was selected as the precursor ion, its MS/MS spectrum is given in Fig. 4b and proposed fragmentation spectrum is given in Fig. 6. Similarly, in case of papaveraldine, the literature describes only molecular ion [31, 32], therefore molecular ion *m/z* = 354.1658 was fragmented (Fig. 4c) and proposed fragmentation spectrum is given in Fig. 6. For both compounds the cleavage of dimethoxyphenyl moiety leads to ion *m/z* = 165.0556, therefore this ion and its fragmentation products are common in the MS/MS of papaverinol as well as papaveraldine. Similarly the elimination of water in case of papaverinol, or elimination of oxygen in case of papaveraldine, leads to common ion *m/z* = 338.1397. This fragmentation ion is also present in MS/MS spectra of papaverine, as it is common benzylisoquinoline moiety for all papaverine derivatives. Its further fragmentation leads to ions *m/z* = 324.1210, 322.1085, 310.1423, and 307.1210. The fragmentation is in full conformity with literature [33–35]. On the other hand the localization of charge on different part of the molecular ion in case of papaverinol results in the formation of fragments *m/z* = 294.1134 and 280.0981, which could be used to distinguish mass spectrum of papaverinol from mass spectrum of papaveraldine.

Quantitation of selected alkaloids

As the alkaloid content of opium of each harvest is highly variable and depends on a number of factors during poppy cultivation [3], the content of opium alkaloids in an analyzed historical sample in the time of its preparation is naturally not available. Therefore, only representatives of all major opiate alkaloid motifs, i.e., morphine, papaverine, and noscapine, were selected for quantification. Since literature [9, 11] recommends decomposition of noscapine for the study of opium-containing samples, the contents of cotarnine and meconin were also measured. The assay was carried out by HPLC with UV detection using the calibration dependency method, and results are summarized in Table 2.

The results show that the morphine content is the highest in Sample I because it is the residue after evaporation of the ethanolic opium solution. On the other hand, in

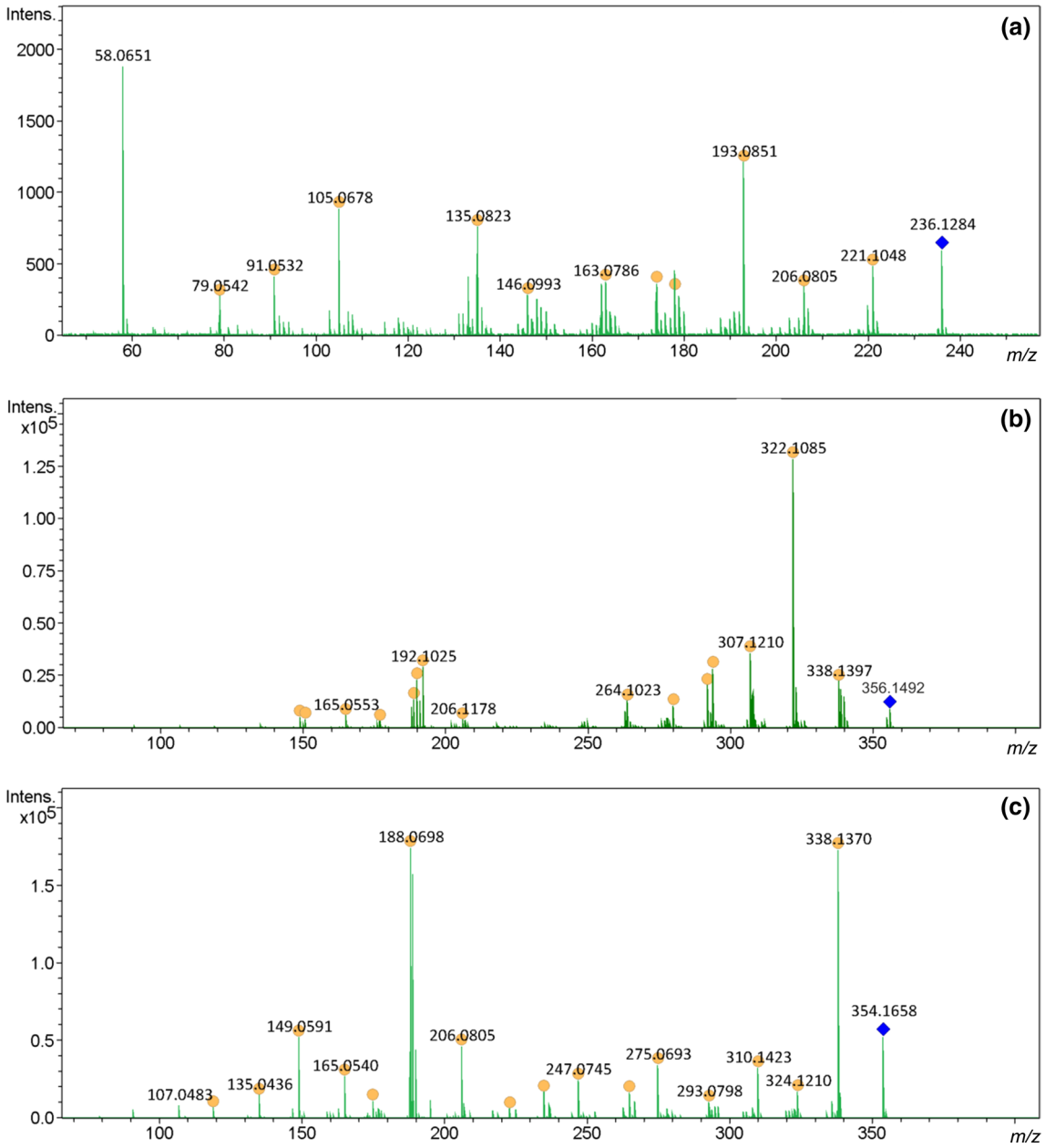

Fig. 4 ESI$^+$-MS/MS spectra of the product ions of **a** methylhydrocotarnine, **b** papaverinol, and **c** papaveraldine (precursor ion is marked by blue diamond)

Sample III, which was prepared not from opium but from poppy seeds, the morphine content is below the detection limit. As suggested [9, 11], for evidence that the sample contains opium, it is appropriate to focus on the content of papaverine, noscapine, and its decomposition product cotarnine. The papaverine and noscapine content in all samples analyzed is within the range of literature [3, 36], so it cannot be used to evaluate the age of the product even if the concentration of the corresponding alkaloid could be reduced by its degradation. As meconin is already contained in crude opium [37], its content can also not be taken into account when characterizing the age of a historical sample.

On the other hand, the concentration of the cotarnine is significantly higher in all analyzed historical samples in comparison with the reference pharmaceutical substance of opium. However, estimating the age of an object according to the content of individual alkaloids content is difficult because, as noted, the values can vary widely in opium of each harvest. Therefore, we propose to take into account the ratio of concentration of noscapine and its direct disintegration product cotarnine as an applicable marker of

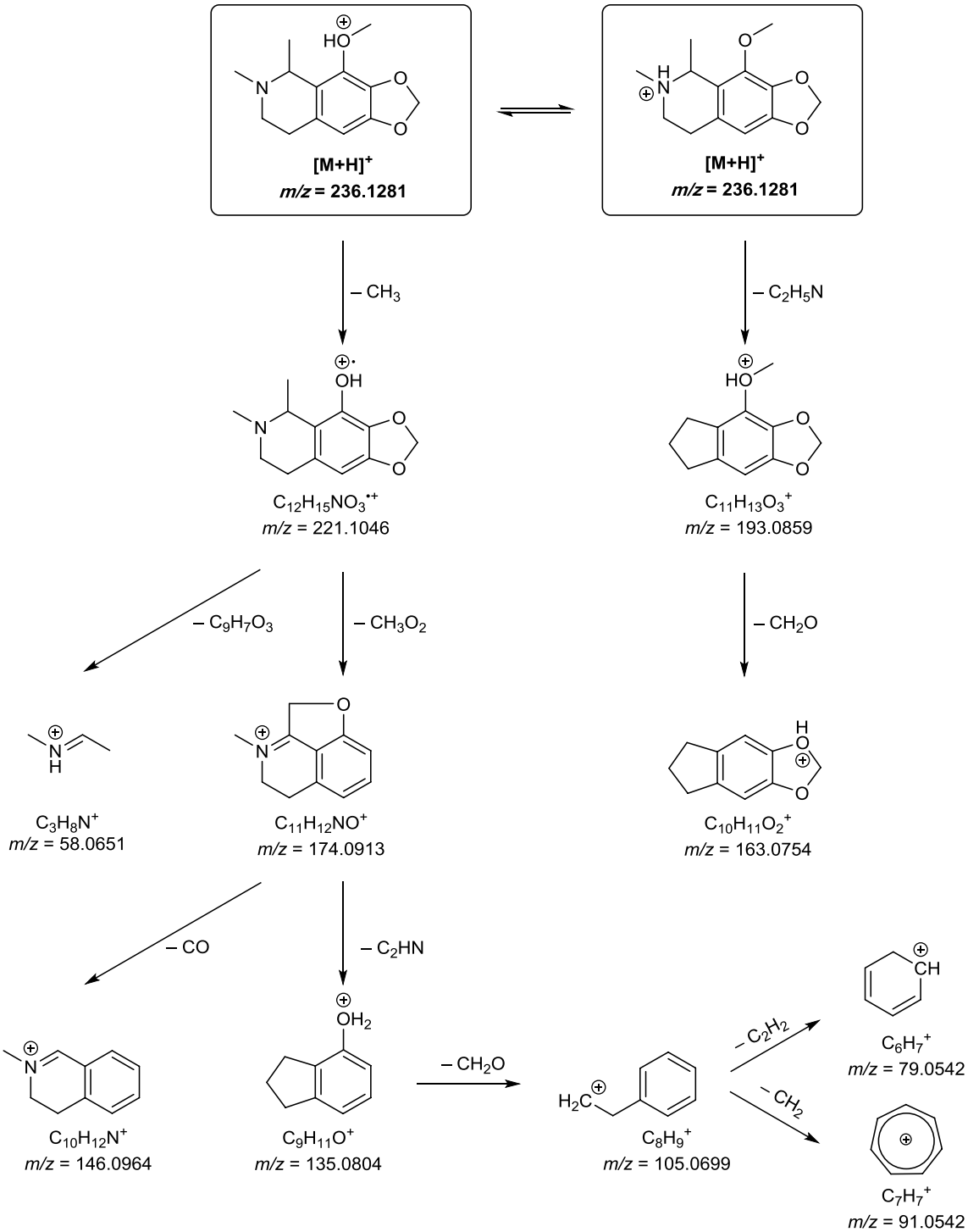

Fig. 5 Proposed ESI$^+$-MS/MS fragmentation of protonated methylhydrocotarnine (the *m/z* values are calculated)

the age of analyzed sample. As shown in Table 2, the ratio for historical samples is about 50 times lower than in the reference pharmaceutical substance of opium. However, a wider study with multiple samples of different age is needed to examine this hypothesis.

Conclusion

Three samples of historical relics of pharmaceutical preparations, more than 200-years old, were analyzed using

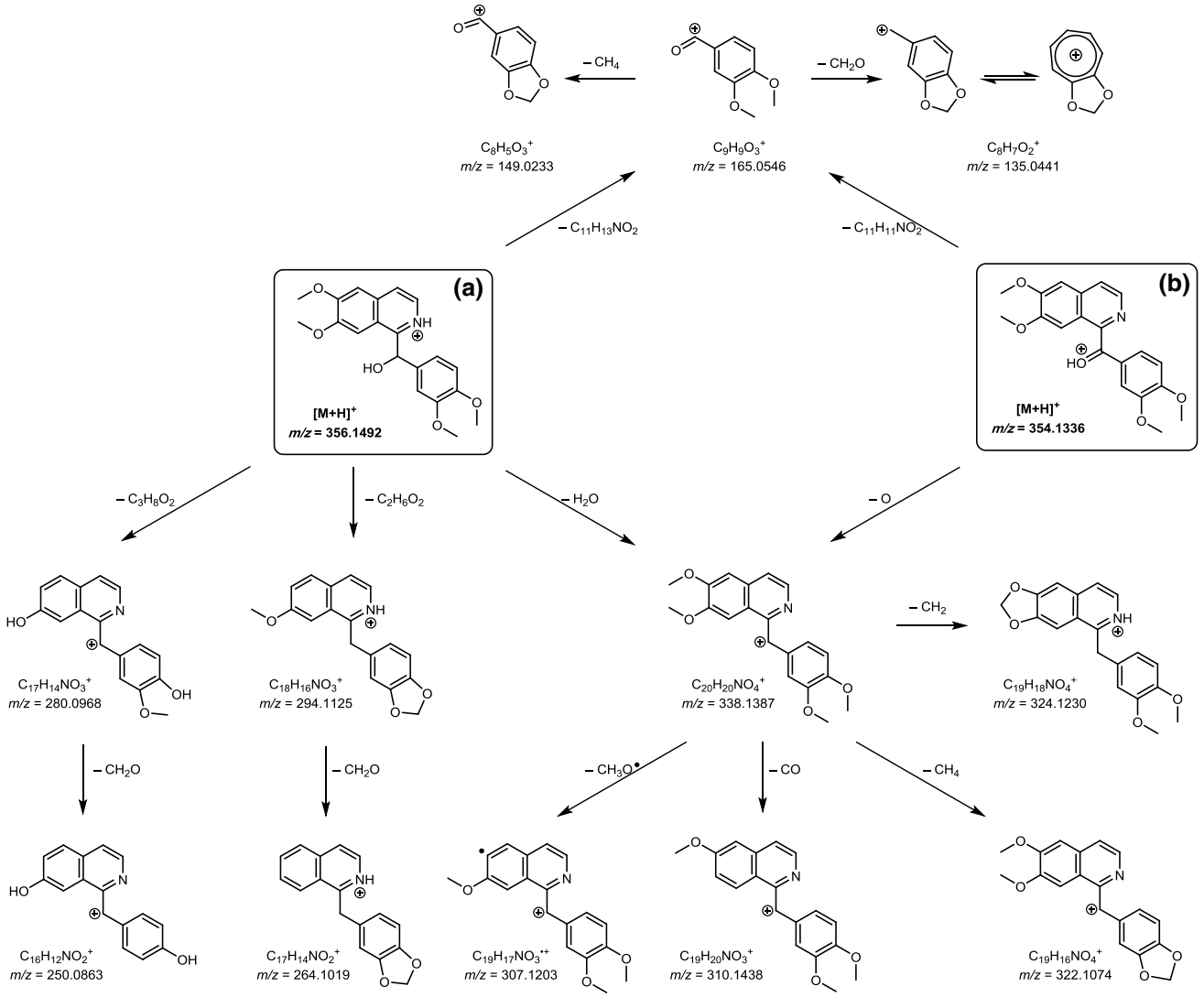

Fig. 6 Proposed ESI$^+$-MS/MS fragmentation of protonated **a** papaverinol and **b** papaveraldine (the *m/z* values are calculated)

Table 2 Results of determination of selected alkaloids in the analyzed historical samples and reference pharmaceutical substance of opium (expressed as median ± SD from three measurements) and the concentration ratio of noscapine and cotarnine

Alkaloid	Measured concentration/%			
	Sample I	Sample II	Sample III	Pharmaceutical substance of opium
Cotarnine	1.55 ± 0.012	0.25 ± 0.017	0.048 ± 0.0022	0.019 ± 0.0054
Morphine	21 ± 1.1	0.7 ± 0.15	< LOD	10 ± 3.6
Meconin	0.496 ± 0.0061	0.081 ± 0.001	< LOQ	0.141 ± 0.0041
Papaverine	2.76 ± 0.024	0.537 ± 0.008	0.021 ± 0.0013	0.885 ± 0.0068
Noscapine	5.83 ± 0.030	0.32 ± 0.020	0.021 ± 0.0016	2.617 ± 0.0014
Ratio of concentrations noscapine: cotarnine	3.76	1.28	0.44	137.8

HPLC–MS/MS. All analyzed samples showed opium alkaloid content, which is the first case at the samples older than 200 years. Degradation products of papaverine and noscapine were detected and the ESI$^+$-MS/MS fragmentation mechanisms of methylhydrocotarnine, papaverinol, and papaveraldine were proposed, which have not been

described in the literature yet. Finally, the concentrations of five selected alkaloids (cotarnine, meconin, morphine, papaverine, and noscapine) in analyzed relics were determined. The concentration ratio of noscapine and cotarnine is suggested as possible marker of the age of analyzed sample.

Experimental

Samples, sample preparation

The analyzed samples are coming from Collection of Old Czech History, National Museum (Prague, The Czech Republic). The baroque pharmaceutical glass jars are dated to the eighteenth century [14].

The first jar (inv. no. H2-4372; herein referred as Sample I) is made from so-called forest glass in a shape of drop of height 14.0 cm and diameter 6.0 cm, and is labelled in Latin 'Laud. Liqvid. Syd.' (Fig. 2a). The lid of the jar is missing and only dry residuum of the content was found caked on the wall of the jar and was collected using the spatula.

The second jar (inv. no. H2-9576, herein referred as Sample II) is made from clear glass in a shape of a cup of height 12.0 cm and diameter 5.0 cm. The jar is painted with pale brown finishing, and is labelled in Latin 'Theriac: Cœleſt:' (Fig. 2b). The original leathern lid, fastened by a string, was gently opened and using glass spoon three samples of the content were collected; one from the center of the jar content, and two from the opposite sides located at the wall of the jar.

The third jar (inv. no. H2-9637, herein referred as Sample III) is made from clear glass in a form of a cylinder of height 14.0 cm and diameter 6.0 cm, and is labelled in Latin '♃ Anodinus' (Fig. 2c). The lid of the jar is missing and the content is freely accessible. The sample was collected using the spatula.

The collected samples were stored in glass containers in a dark. Prior to the analysis the sample was homogenized in a porcelain mortar and the powder was placed in a desiccator with phosphorus pentoxide as a desiccant on 24 h.

For extraction of alkaloids, the analyzed powdered sample was weighed about 5 mg to the tube. An amount of 1 cm^3 of a mixture of acetonitrile–water (85:15, v/v) was added and extraction took 30 min under vigorous shaking. The solution was filtered using 0.2 µm filter (Whatman) and appropriately diluted by mobile phase before HPLC analysis.

Chemicals

The pharmaceutical substance of opium with declared content of opium 9.8% dated to 1950 is from collections of Department of Analytical chemistry, Faculty of Pharmacy, Charles University.

The analytical standards of cotarnine, morphine, meconin, papaverine, and noscapine are from collections of Department of Analytical Chemistry, Faculty of Pharmacy, Charles University. Their identity was confirmed by mass spectrometry and purity was checked by potentiometric titration with 0.1 M perchloric acid in glacial acetic acid medium [38].

The other chemicals employed: acetic acid, acetonitrile, ammonium acetate, ammonium formate, formic acid, methanol, and sodium formate were of HPLC or p.a. grade, and purchased from Sigma-Aldrich.

Instrumentation

A liquid chromatograph UHPLC Nexera XR (Shimadzu, Japan) with an internal diode-array detector was used, followed by a Compact QTOF Bruker mass spectrometer (Bruker, Germany) with ESI ionization. The XBridge® BEH C18 (150 × 3.0 mm i.d., particle size 2.5 µm; Waters) was used. The column temperature was maintained at 40 °C. The mobile phase was composed of methanol and 0.01 M acetate buffer ($CH_3COOH + CH_3COONH_4$) of pH = 3.00 (adjusted by diluted acetic acid). The flow rate of mobile phase was 0.2 cm^3 min^{-1}. The volume of injected sample

Table 3 Figures of merit of HPLC–UV determination of selected opium alkaloids based on area of the peak

Compound	Cotarnine	Morphine	Meconin	Papaverine	Noscapine
Wavelength of detection/nm	250	250	308	250	240
Linear dynamic range/mg dm^{-3}	5–40	40–320	2–35	5–40	10–140
Slope of calibration/mAU min mg^{-1} dm^3	14.114 ± 0.014	0.620 ± 0.014	6.170 ± 0.036	46.52 ± 0.56	9.301 ± 0.094
Intercept/mAU min	0.25 ± 0.31	3.4 ± 2.5	0.89 ± 0.67	6.6 ± 1.3	5.6 ± 7.0
R^2	1.000	0.9985	0.9999	0.9997	0.9996
Limit of quantification/mg dm^{-3}	0.25	4.72	1.59	3.19	10.6
Limit of detection/mg dm^{-3}	0.08	1.43	0.48	0.97	3.2

was 2 mm^3. The compounds were detected using diode-array detector and mass spectrometer. Ionization of the analytes was performed in the positive ion mode at capillary voltage 4.5 kV. The pressure of the nitrogen (nebulizing gas) was set to 0.40 bar. Nitrogen (4.0 dm^3/min) also served as drying gas at 250 °C. Sodium formate was used as a standard for calibration of mass spectrometer.

Selected opium alkaloids were quantified using calibration dependences of the standards. The calibration was based on the area of peak of HPLC–UV chromatograms, and respective figures of merit of the calibration dependences are given in Table 3.

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References

1. Nesmerak K (2016) Bolest 19:103
2. Sneader W (2005) Drug discovery: a history. Wiley, New York
3. Evans WC (2009) Trease and Evans pharmacognosy, 16th edn. Elsevier, Edinburgh
4. Beaudoin GAW, Facchini PJ (2014) Planta 240:19
5. Merlin MD (2003) Econ Bot 57:295
6. Nesměrák K, Kudláček K, Babica J (2017) Monatsh Chem 148:1557
7. Koschel K (1996) Ägypten und Levante/Egypt Levant 6:159
8. Bisset NG, Bruhn JG, Curto S, Holmstedt B, Nyman U, Zenk MH (1994) J Ethnopharmacol 41:99
9. Chovanec Z, Rafferty SM, Swiny S (2012) Ethnoarchaeology 4:5
10. Vermeire A, Remon JP (1999) Int J Pharm 187:17
11. Smith RK, Stacey RJ, Bergstrom E, Thomas-Oates J (2018) Analyst (Cambridge, UK) 143:5127
12. Maurice S, Diefenbach A, Garshott D, McDonald E, Sanday T, Fahey M, Benvenuto MA (2015) ACS Symp Ser 1189:181
13. Nesmerak K, Sticha M, Cvancarova M (2010) Anal Lett 43:2572
14. Nesměrák K, Kunešová J (2015) Ces Slov Farm 64:79
15. Collegium Pharmaceuticum (1729) Dispensatorium pharmaceuticum Austriaco-Viennense. Kürner, Vienna
16. Macht DI (1915) J Am Med Assoc 64:477
17. Warolin C (2010) Rev Hist Pharm (Paris) 58:81
18. Sigerist HE (1941) Bull Hist Med 9:530
19. Triller D (1764) Dispensatorium pharmaceuticum universale: tomus secundus. Varrentrapp, Francofurtum
20. Baggesgaard-Rasmussen H, Reimers F (1935) Arch Pharm Ber Dtsch Pharm Ges 273:129
21. Kursinszki L, Hank H, Kery A, Szoke E (2011) Chromatogr Sci Ser 102:769
22. Bosch ME, Sanchez AR, Rojas FS, Ojeda CB (2007) J Pharm Biomed Anal 43:799
23. Bogusz MJ, Maier RD, Kruger KD, Kohls U (1998) J Anal Toxicol 22:549
24. Hermann TW, Girreser U, Michalski P, Piotrowska K (2002) Arch Pharm (Weinheim, Ger) 335:167
25. Tsunoda N, Yoshimura H (1979) Xenobiotica 9:181
26. Habermehl G, Schunck J, Schaden G (1970) Justus Liebigs Ann Chem 742:138
27. Göeber B, Bauer G, Pfeifer S, Dube G, Engelhardt G, Jancke H (1973) Pharmazie 28:221
28. Göeber B, Pfeifer S, Pankow K, Kraft R (1979) Pharmazie 34:830
29. Melzer B, Bracher F (2015) Org Biomol Chem 13:7664
30. Rideau M, Morard P, Gansser C, Chenieux JJ, Viel C (1988) Pharmazie 43:332
31. Choe S, Kim S, Lee C, Yang W, Park Y, Choi H, Chung H, Lee D, Hwang BY (2011) Forensic Sci Int 211:51
32. Liu C, Hua Z, Bai Y (2015) Forensic Sci Int 257:196
33. Zhang Z, Yan B, Liu K, Bo T, Liao Y, Liu H (2008) Rapid Commun Mass Spectrom 22:2851
34. Peng Z, Song W, Han F, Chen H, Zhu M, Chen Y (2007) Int J Mass Spectrom 266:114
35. Wood GW, Mak N, Hogg AM (1976) Anal Chem 48:981
36. Mohana M, Reddy K, Jayshanker G, Suresh V, Sarin RK, Sashidhar RB (2005) J Sep Sci 28:1558
37. Neumann H (1984) J Chromatogr 315:404
38. Šafařík L, Stránský Z (1986) Titrimetric analysis in organic solvents. Elsevier, Amsterdam
39. Raith K, Neubert R, Poeaknapo C, Boettcher C, Zenk MH, Schmidt J (2003) J Am Soc Mass Spectrom 14:1262
40. Bijlsma L, Sancho JV, Hernández F, Niessen WMA (2011) J Mass Spectrom 46:865
41. Goeber B, Pankow K, Pfeifer S, Kraft R (1975) Pharmazie 30:754
42. Tsunoda N, Yoshimura H (1981) Xenobiotica 11:23
43. Ashour A, Hegazy MAM, Moustafa AA, Kelani KO, Fattah LEA (2009) Drug Test Anal 1:327
44. Fang ZZ, Krausz KW, Li F, Cheng J, Tanaka N, Gonzalez FJ (2012) Br J Pharmacol 167:1271