

## **Supplementary material**

**Table S1.** Oregano food alerts reported on the Food and Feed Safety Alerts (RASFF) portal related to the presence of pyrrolizidine alkaloids (PAs) and their oxidized forms (pyrrolizidine alkaloids *N*-oxides, PANOS) during 2019-2020. Data obtained online from: <https://webgate.ec.europa.eu/rasff-window/portal/?event=SearchForm&cleanSearch=1> (accessed on 28 May 2020)

**Search criteria** | **Hazard category** natural toxins (other) | **Risk decision** serious

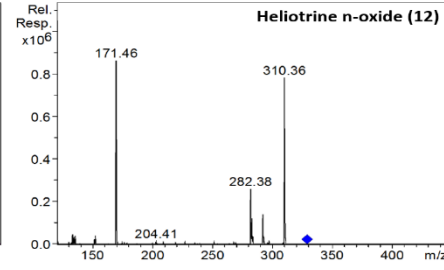
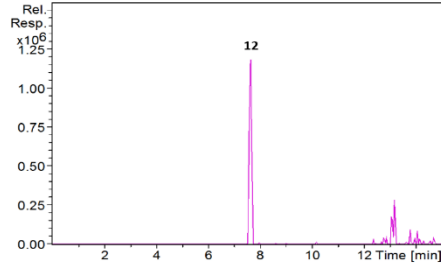
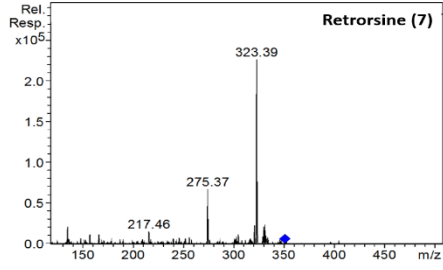
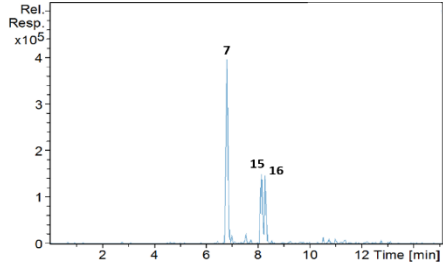
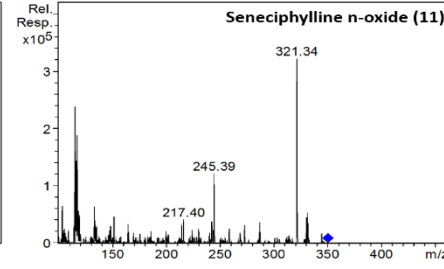
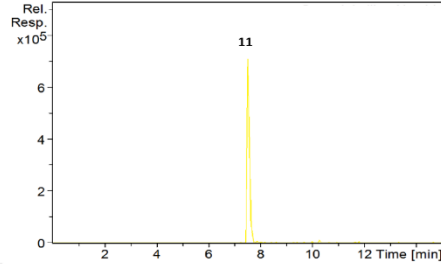
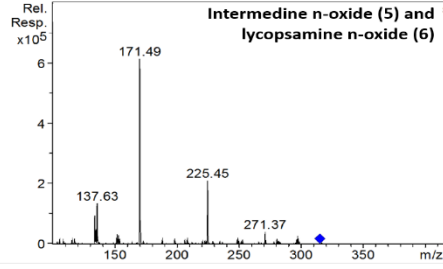
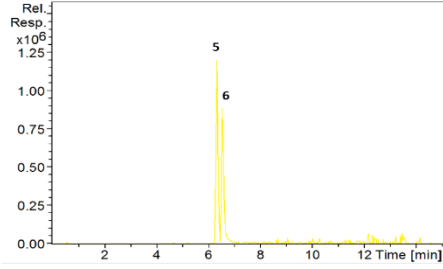
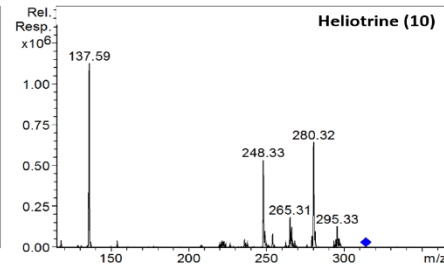
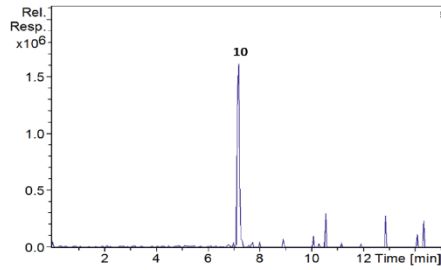
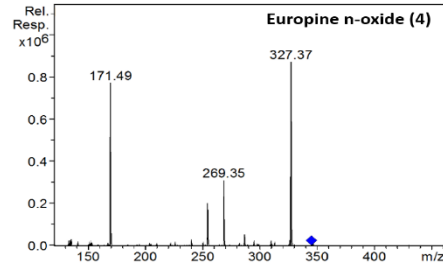
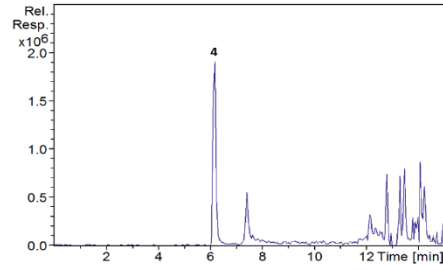
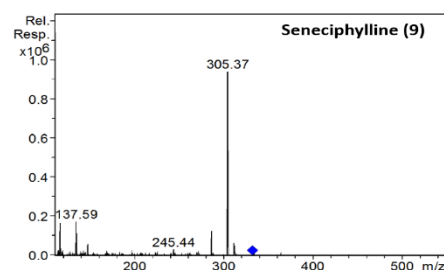
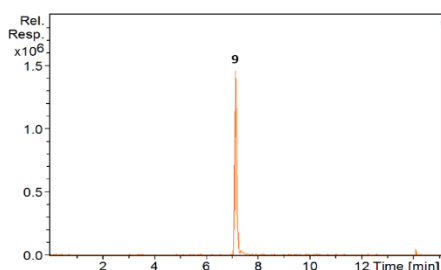
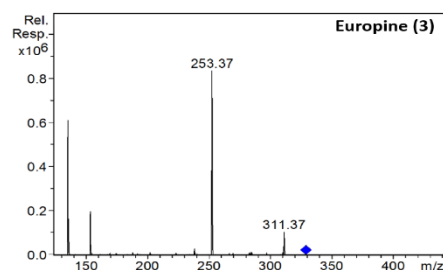
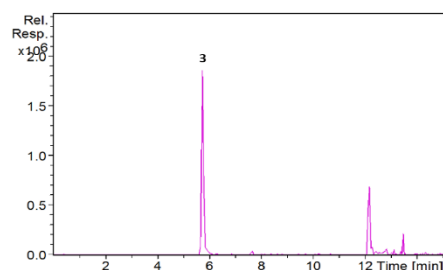
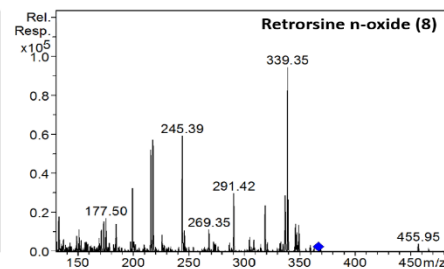
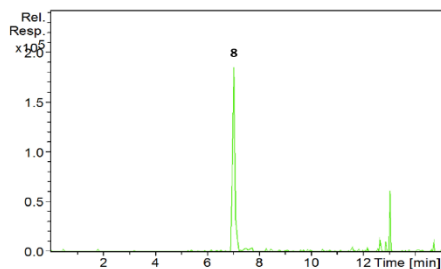
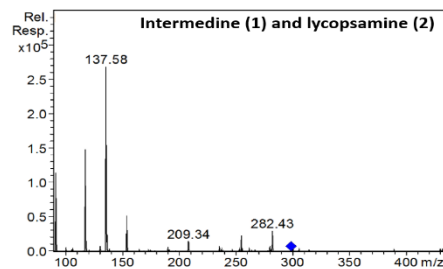
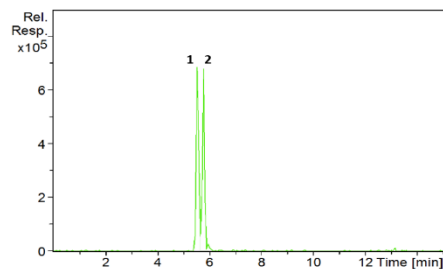
product category	date	reference	product type	notification type	notification basis	notified by	countries concerned	subject	action taken	distribution status	risk decision
herbs and spices	28/01/2019	2019.0305	food	alert	company's own check	Germany	Austria (O), Belgium (D), Czech Republic (D), France (D), Germany (D/O), INFOSAN, Netherlands (D), Slovakia (D), Turkey (O), Ukraine (D)	pyrrolizidine alkaloids (35447; 42724 µg/kg - ppb) in oregano from Austria and Turkey, processed in Germany	withdrawal from the market	distribution to other member countries	serious
herbs and spices	12/02/2019	2019.0504	food	alert	official control on the market	Germany	Austria (D), Commission Services, France (D), Germany (D), INFOSAN, Italy (D), Turkey (O)	pyrrolizidine alkaloids (6660 µg/kg - ppb) in oregano from Turkey	withdrawal from the market	distribution to other member countries	serious
herbs and spices	25/03/2019	2019.1121	food	information for attention	company's own check	Germany	Germany (D), INFOSAN, Turkey (O)	pyrrolizidine alkaloids (16896 µg/kg - ppb) in dried oregano from Turkey	detained by operator	distribution restricted to notifying country	serious
herbs and spices	09/05/2019	2019.1736	food	alert	official control on the market	Austria	Austria (D), France (O), Germany (D)	pyrrolizidine alkaloids (14871 µg/kg - ppb) in oregano from France	withdrawal from the market	distribution to other member countries	serious
herbs and spices	09/05/2019	2019.1739	food	alert	official control on the market	Austria	Austria (D/O), Germany (D)	pyrrolizidine alkaloids (14366 µg/kg - ppb) in oregano from Austria	detained by operator	distribution to other member countries	serious
herbs and spices	09/05/2019	2019.1741	food	alert	official control on the market	Austria	Austria (D/O), Germany (D)	pyrrolizidine alkaloids (21011 µg/kg - ppb) in rubbed oregano from Austria	detained by operator	distribution to other member countries	serious
herbs and spices	31/05/2019	2019.1997	food	information for attention	official control on the market	Germany	Germany (D), INFOSAN, Turkey (O)	pyrrolizidine alkaloids (9870 µg/kg - ppb) in oregano from Turkey	withdrawal from the market	distribution restricted to notifying country	serious
herbs and spices	03/06/2019	2019.2012	food	information for attention	official control on the market	Germany	Germany (D), INFOSAN, Turkey (O)	pyrrolizidine alkaloids (13100 µg/kg - ppb) in rubbed oregano from Turkey	withdrawal from the market	distribution restricted to notifying country	serious
herbs and spices	07/06/2019	2019.2099	food	alert	official control on the market	Germany	Austria (O), France (D), Germany (D), Italy (D)	pyrrolizidine alkaloids (11600 µg/kg - ppb) in oregano from Austria	withdrawal from recipient(s)	distribution to other member countries	serious
herbs and spices	20/06/2019	2019.2235	food	alert	official control on the market	Germany	Austria (D), Belgium (D), Cyprus, France (D), Germany (D), INFOSAN, Netherlands (D), Turkey (O)	pyrrolizidine alkaloids (11700 µg/kg - ppb) in oregano from Turkey	withdrawal from recipient(s)	distribution to other member countries	serious
herbs and spices	08/07/2019	2019.2458	food	information for attention	official control on the market	Germany	Germany (D), INFOSAN, Turkey (O)	pyrrolizidine alkaloids (10526 µg/kg - ppb) in oregano from Turkey	detained by operator	distribution restricted to notifying country	serious
herbs and spices	10/07/2019	2019.2504	food	alert	official control on the market	Austria	Austria (D), Germany (D), INFOSAN, Turkey (O)	pyrrolizidine alkaloids (15852 µg/kg - ppb) in oregano from Turkey	withdrawal from the market	distribution to other member countries	serious
herbs and spices	19/07/2019	2019.2635	food	information for attention	official control on the market	Germany	Germany (D), INFOSAN, Turkey (O)	pyrrolizidine alkaloids (13641 µg/kg - ppb) in oregano from Turkey	recall from consumers	distribution restricted to notifying country	serious
herbs and spices	30/07/2019	2019.2763	food	alert	official control on the market	Germany	Belgium (D), France (D), Germany (D), INFOSAN, Malta (D), Netherlands (D), Turkey (O), unknown origin	pyrrolizidine alkaloids (18496 µg/kg - ppb) in oregano from Turkey	recall from consumers	distribution to other member countries	serious
herbs and spices	07/08/2019	2019.2881	food	information for attention	official control on the market	Germany	Germany, INFOSAN, Turkey (O)	pyrrolizidine alkaloids (7590 µg/kg - ppb) in oregano from Turkey	return to consignor	product not (yet) placed on the market	serious
herbs and spices	26/09/2019	2019.3393	food	alert	company's own check	Germany	Austria (D), Belgium (D), Commission Services, Denmark (D), Germany, INFOSAN, Netherlands (D), Norway (D), Sweden (D), Turkey (O)	pyrrolizidine alkaloids (22167.4 µg/kg - ppb) in oregano from Turkey	informing recipient(s)	distribution to other member countries	serious
herbs and spices	29/01/2020	2020.0453	food	alert	official control on the market	Germany	Belgium (D), Denmark (D), France (D), Germany (D/O), Hungary (D), Luxembourg (D), Netherlands (D), Poland (D), Portugal (D), Slovenia (D), Spain (D), United Kingdom (D)	pyrrolizidine alkaloids (133870 µg/kg - ppb) in rubbed oregano from Germany	no stock left	distribution to other member countries	serious
herbs and spices	04/02/2020	2020.0568	food	alert	company's own check	Germany	Austria (D), Germany (D), INFOSAN, Turkey (O)	pyrrolizidine alkaloids (8836 µg/kg - ppb) in rubbed oregano from Turkey	withdrawal from the market	distribution to other member countries	serious
herbs and spices	05/02/2020	2020.0581	food	alert	official control on the market	Germany	France (D), Germany (D), INFOSAN, Turkey (O)	pyrrolizidine alkaloids (16962 µg/kg - ppb) in oregano from Turkey	withdrawal from the market	distribution to other member countries	serious

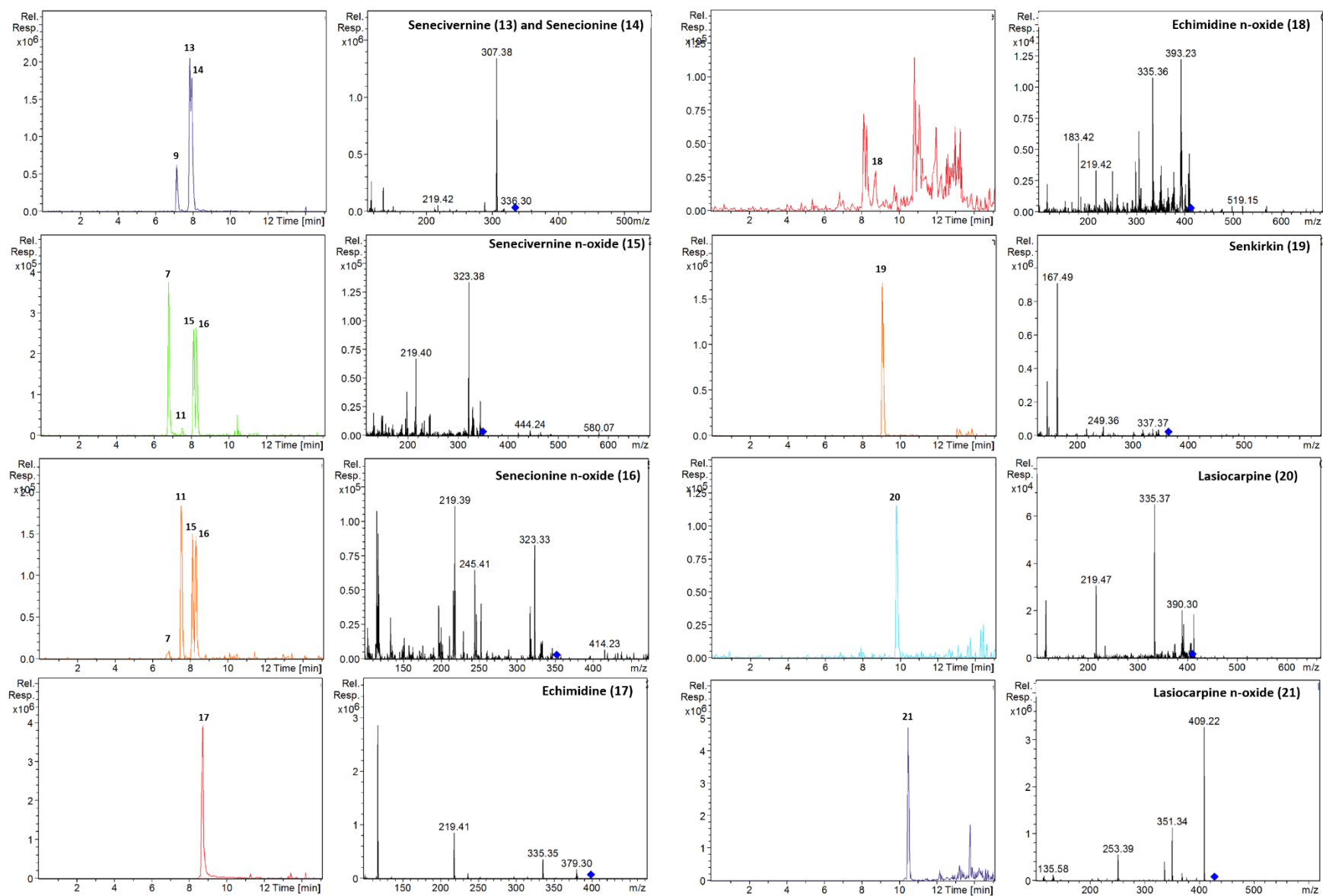
**Table S2.** Information of the oregano samples analyzed.

Oregano Sample	Product origin	Raw material origin	Description
C-S1	Spain	Unknown	Milled oregano leaf
C-S2	Italy	Unknown	Lyophilized oregano
C-S3	Spain	Spain	Milled oregano leaf
C-S4	Spain	Peru	Milled oregano leaf
C-S5	Spain	Peru	Milled oregano leaf
C-S6	Spain	Unknown	Milled oregano leaf
C-S7	Spain	Unknown	Milled oregano leaf
C-S8	Spain	Spain	Milled oregano leaf
C-S9	Spain	Spain	Milled oregano leaf
C-S10	Spain	Unknown	Milled oregano leaf
C-S11	Spain	Unknown	Milled oregano leaf
C-S12	Spain	Unknown	Milled oregano leaf
C-S13	France	Turkey	Milled oregano leaf
C-S14-A	Spain	France	Milled oregano leaf
C-S14-B	Spain	France	Milled oregano leaf
C-S15-A	Germany	Turkey	Milled oregano leaf
C-S15-B	Germany	Turkey	Milled oregano leaf
C-S16-A	Spain	Spain	Milled oregano leaf
C-S16-B	Spain	Spain	Milled oregano leaf
O-S17	Spain	Unknown	Milled oregano leaf from organic farming
O-S18	Spain	Spain	Milled oregano leaf from organic farming
O-S19	Spain	Unknown	Milled oregano leaf from organic farming
W-S20	Spain	Spain	Wild oregano on branch

**Table S3.** Solvent-based standard and matrix-matched calibration curves (expressed at µg/L)

Analyte	Solvent-based standard calibration, R <sup>2</sup>	Matrix-matched calibration, R <sup>2</sup>
Intermedine	y = 27944 x – 53949 0.999	y = 6513 x + 5771 0.999
Europine	y = 61607 x – 49511 0.999	y = 14342 x + 58782 0.999
Lycopsamine	y = 27348 x - 25302 0.999	y = 4990 x + 8916 0.999
Europine <i>N</i> -oxide	y = 138491 x – 357700 0.998	y = 21811 x + 43628 0.999
Intermedine <i>N</i> -oxide	y = 40240 x + 144383 0.999	y = 6754 x + 75308 0.999
Lycopsamine <i>N</i> -oxide	y = 48020 x – 282150 0.999	y = 6454x + 29990 0.999
Retrorsine	y = 26655 x + 2816 0.999	y = 3493 x + 12358 0.999
Retrorsine <i>N</i> -oxide	y = 9764 x + 40585 0.999	y = 1739 x + 13424 0.999
Seneciphylline	y = 61799 x – 12213 0.998	y = 7181 x + 25457 0.999
Heliotrine	y = 88524 x + 33860 0.998	y = 15268 x + 9413 0.999
Seneciphylline <i>N</i> -oxide	y = 27270 x + 27109 0.998	y = 9139 x - 29835 0.999
Heliotrine <i>N</i> -oxide	y = 68052 x – 183194 0.999	y = 2189 x + 13923 0.999
Senecivernine	y = 100194 x – 46983 0.998	y = 24972 x + 480780 0.999
Senecionine	y = 143096 x + 2237 0.998	y = 25056 x + 339046 0.999
Senecivernine <i>N</i> -oxide	y = 18958 x + 42888 0.999	y = 7319 x + 4421 0.999
Senecionine <i>N</i> -oxide	y = 13974 x + 34238 0.999	y = 3865 x + 146398 0.999
Echimidine	y = 88932 x – 223173 0.998	y = 15054 x + 167863 0.999
Echimidine <i>N</i> -oxide	y = 2911 x + 64931 0.999	y = 3217 x - 48988 0.999
Senkirkin	y = 54118 x + 412125 0.998	y = 4247 x + 17984 0.999
Lasiocarpine	y = 5028 x – 24107 0.998	y = 902 x - 4928 0.999
Lasiocarpine <i>N</i> -oxide	y = 150969 x + 113710 0.999	y = 20006 x + 2254 0.999





**Figure S1.** Extracted ion chromatograms and mass spectra (MS<sup>2</sup>) of: intermedine (1), lycopsamine (2), europine (3), europine *N*-oxide (4) intermedine *N*-oxide (5), lycopsamine *N*-oxide (6), retrorsine (7), retrorsine *N*-oxide (8), seneciphylline (9), heliotrine (10), seneciphylline *N*-oxide (11), heliotrine *N*-oxide (12), senecivernine (13), senecionine (14), senecivernine *N*-oxide (15), senecionine *N*-oxide (16), echimidine (17), echimidine *N*-oxide (18), senkirkin (19), lasiocarpine (20) and lasiocarpine *N*-oxide (21) in a standard solution containing 100 µg/L of each analyte.