Pyrrolizidine alkaloids: toxic plant metabolites in our diet

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Background

Pyrrolizidine alkaloids (PAs) are a large class of toxic metabolites produced by many plants (Figure 1). Nowadays, acute intoxication due to dietary consumption of PAs is rare, but chronic, low level consumption of PAs may still present a health risk (EFSA 2011 Scientific Opinion on PAs, EFSA J, 9:2406). PAs are regarded as genotoxic carcinogens for which a lifelong exposure of only 7 ng PAs/kg b.w. per day would correspond to a Margin of Exposure (MOE) of 1:10.000, the threshold of what EFSA considers a low risk.

There are many potential food products that could be contaminated by PAs, but honey and (herbal) tea are often regarded as important sources by which PAs could enter the food chain. 24-h duplicate diets can give a good indication of the actual levels of contaminants that people are exposed to and can also provide information on the probable causative food items.

Objectives

- Insight on the actual exposure of PAs through the diet.
- Insight on possible food items that can be linked with PA contamination.

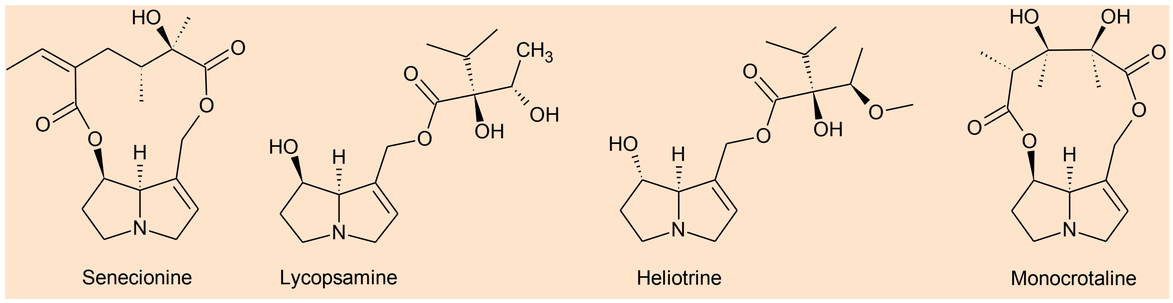

Figure 1. PAs representing the major types of PAs.

Method

- A set of 62 24-h freeze-dried duplicate diets of adults (25-65 years), collected in spring 2011, was used for the analysis of PAs.
- Samples were extracted under acidified aqueous conditions. Extracts were cleaned by SPE (Phenomenex Strata-X), followed by LLE with ethyl acetate/dichloromethane.
- PAs were separated on a Waters UPLC BEH C18 column (150 x 2.1 mm) and analyzed by positive ESI using a Waters Xevo TQ-S LC-MS/MS.
- Samples were screened for 32 PAs. Quantification was performed by standard addition of a set of 18 reference PAs. LOQs were around 0.1 µg/kg.

Results

In Table 1 and Figures 2 and 4 the results are shown for the diets analysed. In 23 samples between 1 and 6 different PAs were detected. In total 15 different PAs were found; integerrimine (10 x), senecionine (8 x), retrorsine (6 x) and heliotrine (6 x) were most often detected.

Senecionine- and lycopsamine-type PAs contributed about equally to the contamination of the diets, while the contribution of heliotrine-type PAs was somewhat lower. Monocrotaline-type PAs were not detected.

The incidence of contamination was significantly higher in the diets of women than that of men. In both groups the incidence and overall contamination also increased with age, being the highest for women of 45 years and older.

The data suggest a correlation between PA incidence and tea or honey consumption. Of the 23 PA-positive diets, 16 contained tea or honey. Of the diets of 32 tea/honey users, 50% was positive.

Table 1. Results obtained for PAs in 24-h duplicate diets

	All	Men	Women	Men 25-44 yrs	Men 45-65 yrs	Women 25-44 yrs	Women 45-65 yrs
No of samples	62	30	32	15	15	15	17
Positive samples	23	6	17	2	4	5	12
% of samples positive	37%	19%	53%	13%	27%	33%	71%
Avg. content (µg/kg)	0.29	0.07	0.49	0.04	0.11	0.17	0.78
Max. content (µg/kg)	5.30	1.11	5.30	0.35	1.11	0.64	5.30
Avg. exposure (ng/kg bw/day)	1.28	0.27	2.26	0.12	0.43	0.94	3.42
Tea/honey consumers	32	10	22	5	5	9	13

Conclusions

- A method for the determination at low level of 32 PAs in freeze-dried 24-h duplicate diets has been developed.
- Of the 62 diets analyzed, 23 contained 1 up to 6 PAs (0.1 – 5.3 µg/kg).
- Women are exposed to higher levels of PAs than men and exposure increases with age for both men and women diets.
- In 2 cases exposure was higher (22.3 and 9.9 ng/kg bw/day) than the safe level proposed by EFSA.
- In 16 of the 23 positive findings respondents had consumed honey and/or tea.

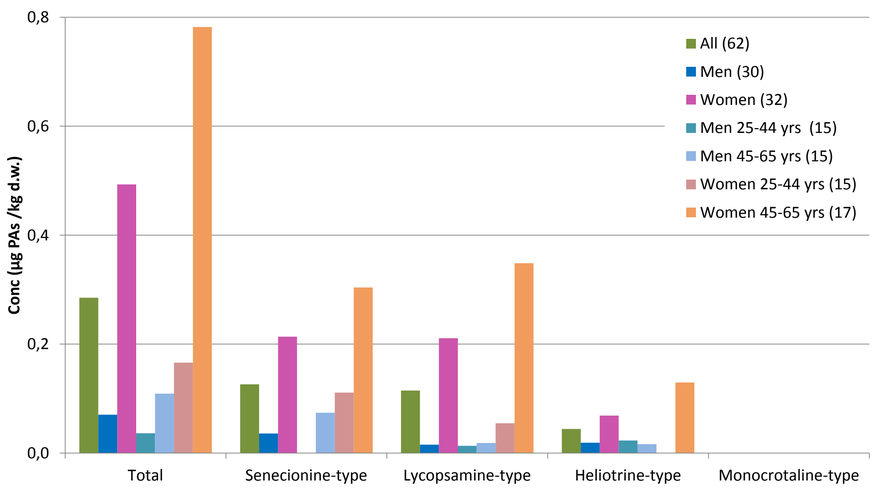

Figure 2. Average concentration of PA-types in 24-h diets of different (sub-)populations.

Figure 3. Our diet, a complex mixture of food items.

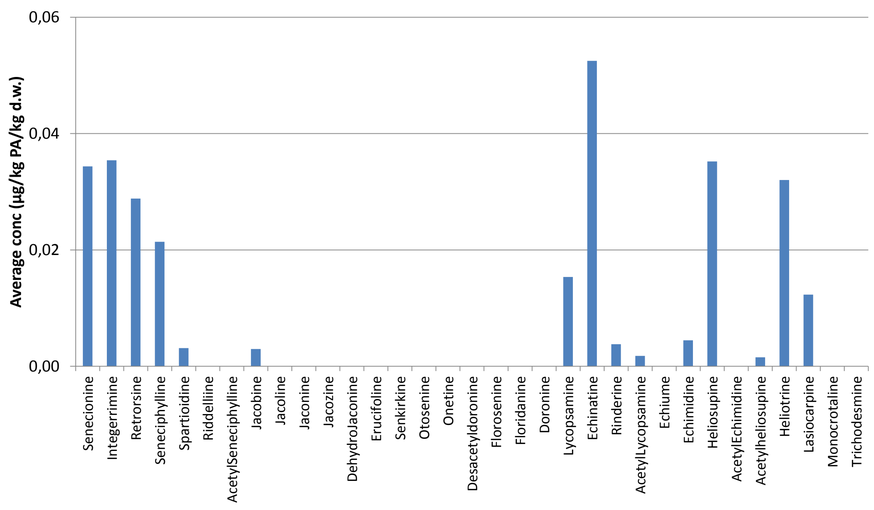

Figure 4. Average concentration of individual PAs in 24-h diets.

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