ARSENIC SPECIATION IN RICE AND FISH USING HPLC-ICP-MS

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INTRODUCTION

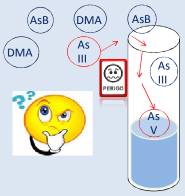
Chemical speciation in foodstuffs is of uttermost importance since it is nowadays recognized that both toxicity and bioavailability of an element depend on the chemical form in which the element is present (1). Regarding arsenic, inorganic species are classified as carcinogenic while organic arsenic, such as arsenobetaine (AsB) or arsenocholine (AsC), are considered less toxic or even non-toxic (2). By coupling a High Performance Liquid Chromatography instrument (HPLC) with an Inductively Coupled Plasma Mass Spectrometer (ICP-MS), it is possible to associate the power of separation of the first with the selectivity and sensitivity of the second. The objective of the present work is to develop a method, using HPLC-ICP-MS technique, to identify and quantify the chemical species of arsenic present in two food matrices, rice and fish.

MATERIAL AND METHODS

Two extraction methods, ultrasound and microwave, and several conditions were evaluated. The best method was chosen based on recovery percentages.

To ensure that no interconversion of species was occurring, individual spikes of each species of arsenic were made on both matrices and recovery rates between 80 and 120% were accepted.

Reference material BCR-627 TUNA FISH, containing certified values for AsB and DMA, was analyzed to guarantee accurate results.



Chromatographic separation was achieved using an anion exchange column, HAMILTON-PRP X-100, which allowed to separate the four arsenic species for which standards were available (AsB, dimethylarsenic (DMA), arsenite (AsIII), arsenate (AsV). The mobile phase was chosen based on scientific literature and adjusted to laboratory conditions. Different gradients were studied.

Table 1 – *Extraction Method - Rice*

Sample weight		0.3 g
H_3PO_4		10 mL
Microwave extraction		
Step	Time (min)	Temperature (°C)
1	20	0-80
2	20	80
Centrifugation		3500 rpm 30 min
Filtration		0.2 µm

Table 2 – *Extraction Method - Fish*

Sample weight	0.25 g
MeOH/H_2O (1:1 v/v)	10 mL
Sonication	60 min
Centrifugation	3500 rpm 30 min
Filtration	0.2 µm

Table 3 – *Operating conditions ICP-MS (Thermo X series II)*

Forward Power (W)	1400
Cool gas flow rate (l/min)	13.0
Nebulizer flow rate (l/min)	0.87
Aux gas flow rate (l/min)	0.90
Pole Bias	-0.1
Hexapole Bias	-0.3

Table 4 – *Operating conditions HPLC (Finnigan Surveyor)*

Column	HAMILTON PRP-X100
Mobile phase	10-50 mM $(NH_4)_2CO_3$; 3% MeOH; pH=9.2
Injection volume	25 µL – fish 100 µL – rice
Flow	1 ml/min

Figure 1 – *Clean room facilities at INSA/DAN*

RESULTS

QUALITY CONTROL

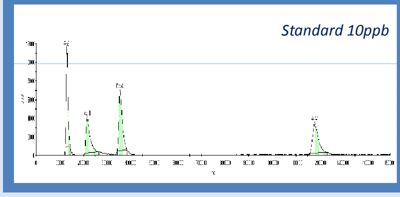


Figure 2 – Arsenic species (AsB, As^{III}, DMA , As^{V})

Table 5 – BCR 627 Tuna Fish

	Laboratory value (mg/kg)	Reference value (mg/kg)
AB	3.5	3.68 – 4.12
DMA	0.17	0.13 - 0.17

FISH

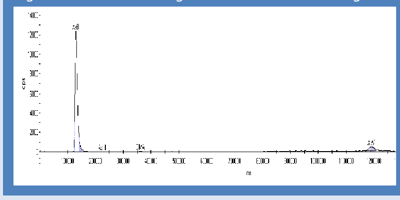


Figure 3 – Anion exchange HPLC-ICP-MS chromatogram

Table 6 – Arsenic species and total arsenic concentration

Total As : 786 µg/kg			Total As (%)
Arsenic species	AsB	597 µg/kg	76 %
	DMA	<LQ	-
	As (V)	5 µg/kg	-

RICE

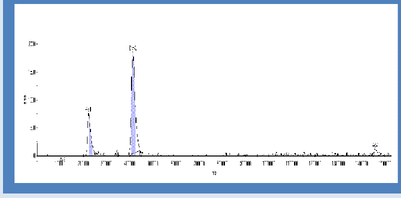


Figure 4 – Anion exchange HPLC-ICP-MS chromatogram

Table 7– Arsenic species and total arsenic concentration

Total As : 277 µg/kg			Total As (%)
Arsenic species	As (III)	84 µg/kg	30 %
	DMA	164 µg/kg	59 %

CONCLUSIONS

Different arsenic species were identified in the foodstuffs under study. While in fish 90% of the arsenic present was in the form of AsB, in rice 80% of arsenic was present as DMA and 20% as inorganic arsenic (As III). By determining only the total arsenic concentration, results suggest that fish consumption represent a bigger risk to consumers than rice. However, speciation studies reveal that the species present in rice, even thought at lower concentrations, are more toxic and therefore represent a higher threat to consumers. Thus the obtained results show that determining only the total concentration of arsenic may be misleading.

Our results demonstrate that HPLC coupled to ICP-MS is a crucial tool to identify arsenic chemical species present in foodstuffs.

References

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