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DETERMINATION OF VANADIUM IN DIETARY SUPPLEMENTS

Urška Repinc and Ljudmila Benedik

Department of Environmental Sciences, Jožef Stefan Institute, 1000 Ljubljana, Slovenia;
e-mail: urska.repinc@ijs.si

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Abstract

In our present work 10 sports dietary supplements and 6 daily multivitamin/mineral supplements were analysed for vanadium content. Radiochemical neutron activation analysis was used to determine whether vanadium had been added to some popular dietary supplements used to increase muscle mass. After irradiation and rapid dissolution of the irradiated sample, solvent extraction was used for selective separation of vanadium. The chemical yield of the radiochemical procedure for each sample aliquot was determined by spectrophotometry. The daily intakes of supplemental vanadium were compared to the median intake of supplemental vanadium and Tolerable Upper Intake Level (UL).

Key words: vanadium, supplements, neutron activation analysis

Introduction

Vanadium is a trace element present in normal human diet only in minor quantities. Normal vanadium intake from food is up to 20 μg/day, while the median intake of supplemental vanadium by adults is approximately 9 μg/day.[1] The essentiality of vanadium has not been proved beyond doubt for humans and limited information is available about vanadium toxicity. During the last decade, vanadium has been found to act in an insulin-like manner in all three main target tissues of the hormone insulin, namely skeletal muscle, adipose tissue and liver.[2] Because insulin has an anabolic effect on skeletal muscles, the finding that vanadium mimics insulin was quickly exploited by seller of supplements as support for the claim that vanadium has a anabolic effect, and can thus be used to enhance muscle building, strength and performance. However, a double-blind placebo-controlled study involving 31 weight-trained athletes found no benefit at a dosage more than 1000 times the nutritional dose.[3]

Dietary supplements are products intended for ingestion as a supplement to the diet. They include vitamins, minerals, herbs, botanicals, and other plant-derived substances, amino acids (the individual building blocks of protein) and concentrates, metabolites, constituents and extracts of these substances. Dietary supplements are some

of the hottest selling items on the market today. But even with all the business they generate, consumers still ask questions: Can their claims be trusted? Are they safe? Because of the widespread use of high-dose supplemental vanadium by athletes to improve performance, with consumption of up to 60 mg of vanadyl sulphate (or 18.6 mg elemental vanadium) per day, there is increased concern about its long term toxicity and further reasearch should be performed on the efficacy and safety of the use of vanadium as a nutritional supplement.[1]

A number of analytical techniques have been used to determine mg/kg to μg/kg levels of vanadium in biological materials. Owing to its sensitivity, radioanalytical neutron activation analysis (RNAA) is superior for this purpose, but problems arise in practice due to the very short half-life of the neutron-induced radionuclide ^{52}V ($t_{1/2}$ = 3.75 min). Post-irradiation radiochemical separation retains the unique advantage of NAA as a "blank-free" technique, but it requires a skilled rapid and selective separation of ^{52}V from the matrix.

In our present work 10 sports dietary supplements and 6 daily multivitamin/mineral supplements were analysed for vanadium content using RNAA. The daily intakes of supplemental vanadium were compared to the Tolerable Upper Intake Level (UL) for vanadium. The Tolerable Upper Intake Level (UL) is the highest level of daily nutrient that is likely to pose no risk of adverse health effects for almost all individuals. The UL for vanadium for the adult population (>19 years) is significantly higher compared to normal levels found in food, and was derived to give the value of 1800 mg/day.[1]

Experimental

Reagents

N-benzoyl-N-phenyl-hydroxylamine (BPHA, Merck) was dissolved in toluene to give a 0.2% w/v solution, and stored in the dark. A 5 μg V/g irradiation standard solution was prepared by dilution of a vanadium standard solution of 1 mg V/g (Merck, Lot.No.K24914066). A carrier solution of 200 μg V/g was prepared from ammonium vanadate (Merck).

Irradiations and instruments

Samples were irradiated in the pneumatic transfer system (rabbit facility) of the TRIGA MK II reactor in Ljubljana up to 60 s, at a neutron fluence rate of 4×10^{12} n cm^{-2} s^{-1} to induce ^{52}V by the reaction $^{51}V(n,\gamma)^{52}V$ ($t_{1/2}$ = 3.75 min). Irradiation standards consisted of about 200 mg of solution (5 μg V/g) sealed in a polyethene tube 2 mm i.d. and taped alongside the sample. Both sample and standard were encapsulated in polythene foil.

Separation procedure

The irradiated sample (up to 0.1 g) was weighed after irradiation. and quickly transferred to a long-necked silica Kjeldahl flask and using a large gas flame, wet-ashed vigorously in 3 mL of conc. sulphuric acid containing 100 μg V-carrier by repeated additions of nitric acid, until a pale yellow-green solution was obtained. Then 2 mL of 70% perchloric acid was added and the mixture heated until perchloric acid was fumed off. The flask was cooled, 8 mL of water and 3 mL of 10 M HCl was added to transfer the content to a 50 mL separatory funnel. Then 6 mL of BPHA reagent were added and the separator shaken for 20-30 s to extract vanadium. The aqueous phase was discarded, the organic phase scrubbed for 15 s with 5 mL of 5 M HCl, and 5 mL of organic phase pipetted into a counting vial for ^{52}V gamma activity measurement at 1434.2 keV. The time taken from the end of irradiation , to the measurement of ^{52}V takes about 8 min or with practice, even less.

Measurement of ^{52}V

Samples and standards were counted on an HP Ge well-type detector connected to a Canberra MCA by Genie 2000 software in a 5 mL measuring vial with 300 s counting time.

Chemical yield determination

Absorbances of the organic phase after extraction without dilution and of vanadium calibration solutions for spectrophotometry were measured on an ISKRA MA 9525–SPEKOL 210 visible spectrophotometer, single beam instrument, at a monochromator slit width of 3 nm at λ = 525 nm, calibrated with a reference cell containing toluene.

Results and discussion

Radiochemical NAA for determination of vanadium, based on rapid wet ashing of the sample and solvent extraction of V into BPHA, gives reliable results and its accuracy has been checked by analysis of reference materials (Table 1).

Table 1. Results for determination of vanadium in reference materials (Uncertainties were calculated based on EUROCHEM guide combined uncertainty budget table for 2 replicates).[4]

Samples	V(ng/g)	
	Literature values	This work
IAEA Wheat Flour V-2/1	43.8 ± 3.5	42.8 ± 2.4
IAEA A-8, Milk Powder	19.5 ± 2	19.9 ± 1.4
NBS SRM-1567a, Wheat Flour	(NC 11)	11.7 ± 0.8
NIST SRM 1570a, Spinach Leaves	(C 570 ± 29)	548 ± 31

NC - non certified, C – certified.

The post-irradiation separation procedure demands manipulation of a rather radioactive sample to perform selective decontamination of few ng of vanadium from matrix elements present at μg concentration range (Na, Cl, Fe, Mn, Cu, etc.). As far as radiochemical purity of the gamma spectra is concerned, most of the interferences are removed in the selective solvent extraction step and spectral purity is excellent (Figure 1a). As noted earlier by Byrne,[5] ^{101}Mo -^{101}Tc peaks were observed in samples rich in molybdenum (Figure 1b), which is added as a supplement in the form of natural molybdate. Molybdenum is nearly quantitatively co-extracted with vanadium into BPHA and can be simultaneously determined for very little extra effort in the same sample aliquot. For samples with adequate concentrations this procedure using a single short irradiation suffices on the basis of the induced radionuclides ^{52}V, ^{101}Mo ($t_{1/2}$ =14.6 min, E_γ = 191.9 keV (18.8%), 590.9 keV (16.4%), 1012.5 keV (12.8%), etc.). Interfering peaks of induced ^{51}Ti ($t_{1/2}$ =5.79 min) formed by the reaction ^{50}Ti(n, γ) at 320 keV (93.0%), 608.6 keV (1.18%) and 928.64 kev (6.9%) were observed in sample Centrum A-Z, *Whitehall*. Titanium extraction efficiency into BPHA should be evaluated in further reasearch. Interferences due to the ^{128}I ($t_{1/2}$ = 24.99 min, E_γ = 442.9 keV(17.5%)) and ^{56}Mn ($t_{1/2}$ = 2.58 h, E_γ = 846.6 keV (99%)) were observed only in samples with high iodine and manganese content. The evaluation of the extraction yield has shown that less than 4% of iodine and less than 1% of manganese is co-extracted with vanadium into

BPHA. ^{52}V peak is well resolved from interfering peaks and was quantitatively evaluated using Genie 2000 software.

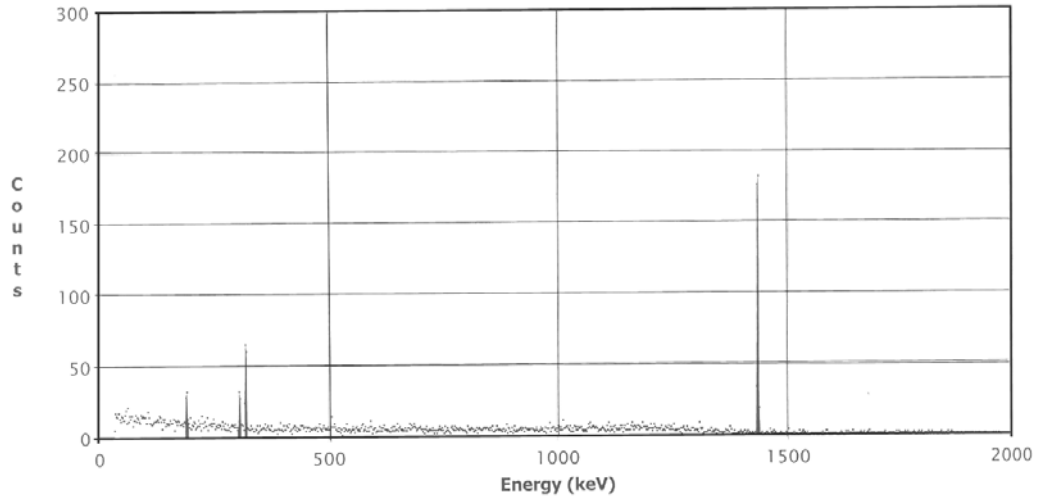

Figure 1a. Spectrum of V-BPHA complex after separation, sample "Myoplex Nutrition Shake, *EAS*".

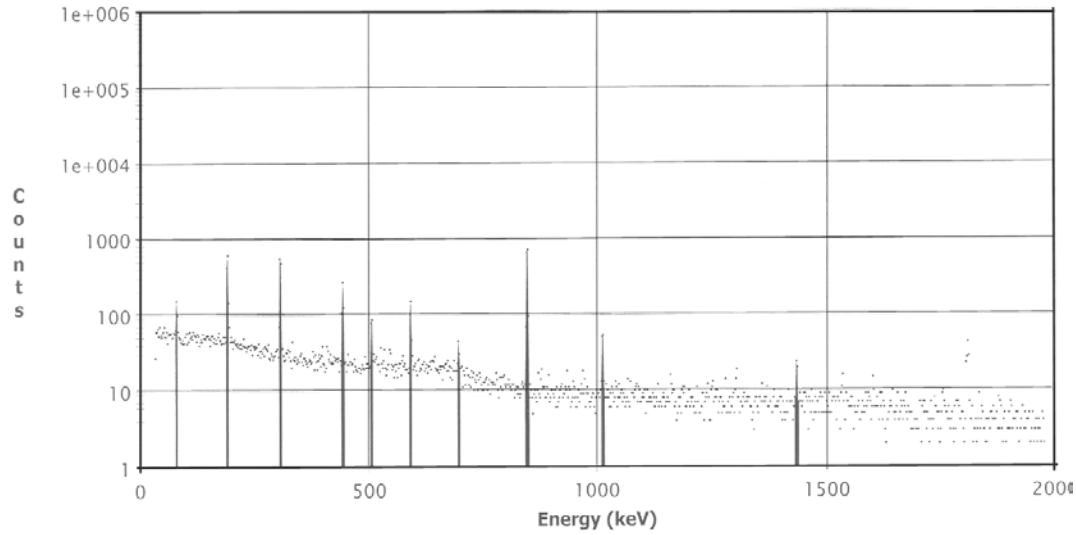

Figure 1b. Spectrum of V-BPHA complex after separation, sample "Daily One, *Twin Lab*" (log scale), with interferences from ^{128}I, ^{56}Mn and ^{101}Mo -^{101}Tc.

The results of 10 sports dietary supplements and 6 daily multivitamin/mineral supplements analysed for vanadium content using RNAA are presented in Table 2 and 3. The daily intakes for vanadium were calculated on the basis of the highest recommended serving amount and compared to the median intake of supplemental vanadium as reported by Pennington and Jones in 1987.[1]

Table 2. Concentrations, recommended servings and calculated daily intakes (CDIs) of vanadium in sport supplements.

	Sample, *Producer*	Recommended serving (g/day)	Concentration of V (ng/g)	CDI (μg)	(CDI)/(MI) %
1	Myoplex, *EAS*	228	186 ± 11	43 ± 2.6	478
2	Simply Whey Protein, *EAS*	120	27 ± 2	3.2 ± 0.2	36
3	Promax, *Scitec Nutrition*	75	60 ± 4	4.5 ± 0.3	50
4	Scipro, *Scitec Nutrition*	75	4.0 ± 0.4	0.3 ± 0.03	3.3
5	Myoplex Nutrition Shake, *EAS*	304	216 ± 13	66 ± 4	733
6	Iso Pro, *Prolab*	102	61 ± 4	6.3 ± 0.4	70
7	Volumas 35, *Scitec Nutrition*	300	4.9 ± 0.7	1.5 ± 0.2	17
8	HMB, *Scitec Nutrition*	9	6.6 ± 0.7	0.1 ± 0.01	1.1
9	Daily One, *Twin Lab*	1	598 ± 40	0.6 ± 0.04	6.7
10	Sport Fuel, *Twin Lab*	6	107 ± 7	0.6 ± 0.04	6.7

CDI – calculated daily intake, MI – median intake of supplemental vanadium.

Table 3. Concentrations, recommended servings and calculated daily intakes (CDIs) of vanadium in vitamin/mineral supplements.

	Sample, *Producer*	Recommended serving (tablet/day)	Concentration of V (ng/g)	CDI (μg)	(CDI)/ (MI) %
1	Centrum A-Z, *Whitehall*	1	11.5 ± 1.5(10*)	0.016 ± 0.002	0.17
2	Full Spectrum, *Perfect Nutrition*	1-3	805 ± 50	3.2 ± 0.2	35
3	Fe shower tablets, *Krüger*	1	209 ± 14	0.94 ± 0.06	10.4
4	PEZ multivitamin shower tablets, *PEZ Hungaria Kft.*	1	1.9 ± 0.5	0.008 ± 0.002	0.09
5	Tema basic vitamin C, *Amos Vital GmbH*	1	40.0 ± 2.5	0.160 ± 0.01	1.8
6	UNICAP T, *Pharmacia & Upjohn*	1	1950 ± 300	2.0 ± 0.3	21.6

CDI – calculated daily intake, MI – median intake of supplemental vanadium, *reported value.

Conclusions

The presented results of the calculated daily intakes for vanadium in 10 sports dietary supplements and 6 daily multivitamin/mineral supplements showed that the supplements do not significantly increase normal vanadium intake levels. The results for sports supplements selected in this study indicate that vanadium is present only as a natural constituent of whey or wheat powder and has not been added in supplemental form as vanadyl sulphate or sodium metavanadate. Values obtained were significantly lower compared to the Tolerable Upper Intake Level (UL) of 1800 mg/day, showing that the risks of adverse effects resulting from excesss consumption of these products is very unlikely.

Acknowledgements

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Povzetek

Radiokemijska nevtronska aktivacija (RNAA) je zelo občutljiva in uporabna metoda za določanje vanadija v bioloških vzorcih. Vzorec po obsevanju razkrojimo in ločimo vanadij s selektivno ekstrakcijo z N-benzoil-N-phenil-hidroxilaminom v toluenu ter merimo aktivnost ^{52}V (E_γ = 1434.2 keV) na gama detektorju. Izkoristek postopka določimo spektrofotometrično z merjenjem absorbance V-BPHA kompleksa pri E_{max} = 525 nm. V tej študiji smo v 10 prehranskih dodatkih za športnike in 6 multivitaminskih/mineralnih prehranskih dodatkih določili vsebnost vanadija, izračunali dnevni vnos vanadija in primerjali rezultate z vrednostjo iz literature.