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Wild bilberries – Superfood or health risk

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ABSTRACT

Wild bilberries (*Vaccinium myrtillus* L.) were collected from 23 sites in Finland, dried, ground, homogenized and pressed into pellets. The pellets were irradiated in air with a 3 MeV proton beam from the Åbo Akademi MGC-20 cyclotron. The induced X-rays were measured, and elemental concentrations were calculated from the peak areas in the spectra. Elemental concentrations for K, Ca, Mn, Fe, Cu, Zn, Rb, and Sr were found to be above the detection limit of the method. A special focus was set on the high concentration of Mn ($27 \pm 16 \mu\text{g/g}$). Already by consuming 100 g of fresh wild-grown bilberries the Recommended Dietary Allowance value for an adult male was exceeded. A child of 4–8 years old will reach the Tolerable Upper Intake Level of Mn (3 mg/day) by eating somewhat more than 100 g of bilberries in a day. In addition, the regional differences in elemental concentrations were found to be higher than earlier reported. The activity concentrations of Cs-137 in some of the bilberry samples were also measured and the equivalent dose to a person that annually consumes 2 kg of bilberries was found negligible (about 1 μSv).

1. Introduction

Blueberries are picked and eaten all over the world. They are considered to have a positive impact on human health and are in some cases consumed in considerable amounts (Vanecková, and Rollinger, 2022; Brašanac-Vukanović et al., 2019). Blueberries are often referred to as a superfood (e.g., Krishna et al., 2023). Dried berries are sold in health shops. One of the reasons for their popularity is the high concentration of manganese. By eating blueberries, it is possible to reach the recommended daily intake of manganese.

In Finland, European blueberries or bilberries (*Vaccinium myrtillus* L.) are collected in the wild, frozen and eaten in large amounts by the people (Finnish Food Authority, 2022). The berries have been used for medicinal purposes since the Middle Ages for a variety of conditions, including diarrhoea, scurvy, infections, burns, and diabetes (Smeriglio et al., 2019). Although the berries contain antioxidants as well as vitamins (C and K), the high concentration of manganese may have a harmful effect on human health especially if consumed by children

under the age of 13 (Food and Nutrition Board, National Academies, 2019a). It has been shown that the wild bilberries contain about ten times more manganese than cultivated blueberries (Karlsons et al., 2018). Manganese is a cofactor in enzyme reactions but will be neurotoxic at high concentrations. The high intake of manganese could lead to learning difficulties, behavioral problems as well as have a detrimental effect on IQ (O'Neal and Zheng, 2015).

Elemental concentrations in wild bilberries have been measured recently by applying Inductive Coupled Plasma Optical Emission Spectrometry (ICP-OES) or Inductive Coupled Plasma Mass Spectrometry (ICP-MS) to dissolved berries (e.g., Skesters et al., 2014; Karlsons et al., 2018; Drózdź et al., 2018). In most of these studies, the berries have been purchased from retail stores, supermarkets and market squares and no geographical information on sampling sites has been presented. In the present study, Particle Induced X-ray Emission (PIXE) was applied directly on dried and pressed pellets of wild bilberries from 23 sampling sites in Finland. Focus was set on the regional differences in manganese concentration.

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Fig. 1. Map of sites where bilberries were picked in the autumn of 2019.

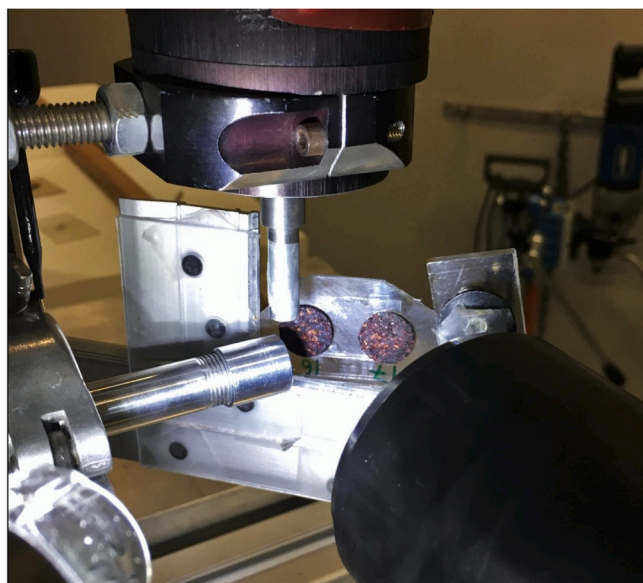


Fig. 2. PIXE set-up. Particle beam end nozzle to the left, collimator for PM-tube measuring light emission up in the middle and IGP detector end cap down in the right corner. Two pressed bilberry samples are seen in the centre.

2. Materials and methods

The bilberries were picked mainly in a region south of Vaasa in the western part of Finland (Fig. 1) in August 2019. The distance from the southern sampling site to the northern sampling site was about 23 km. The region was of special interest due to earlier findings of high manganese concentration found in pine bark samples from Ostrobothnia (Saarela et al., 2005). Two additional samples were collected from the Jakobstad region (21, 22: Fig. 1), about 100 km north of Vaasa, and one from Sjundeå in the southern part of Finland (23: Fig. 1). The fresh berries were dried in an ordinary oven at 50 °C for about 24 h. The oven door was slightly ajar to allow moisture to escape. The dry weight was 14.4 % of the fresh weight. The dried bilberry samples were crushed and homogenised in a mortar and pressed into pellets with a diameter of 13 mm.

The pellets were irradiated in air with 3 MeV protons from the MGC-20 cyclotron (Fig. 2) at Åbo Akademi University. The diameter of the particle beam was 0.5 mm, and the current was 5 nA. The current was monitored by utilizing light emission in air induced by the particle beam (Lill, 1999). Several spots on the pellets were irradiated during the acquisition of the X-ray spectra to reduce the influence of inhomogeneous elemental distributions. An IGP detector was used to measure the emitted X-rays (Fig. 3). The GUPIX software (version 2003–03–20) was used to analyse the obtained X-ray spectra and quantify the peak areas in the spectra (Campbell et al., 2000). The precision of the method depends on many factors such as element, matrix, geometry etc., but was in this study in the range of 3–16 %. The results were evaluated by analysing pressed pellets of the geological material G2 (USGS) and the biological reference material Pine Needles (SRM 1575, NIST). The relative standard deviation of the method has earlier been evaluated by repeated analyses of biological CRMs and found to be 7 % for potassium and 6 % for zinc (Lill et al., 1999).

Bilberries in Finland contain small amounts of radioactive caesium, Cs-137, that mostly originate from the Chornobyl nuclear accident in

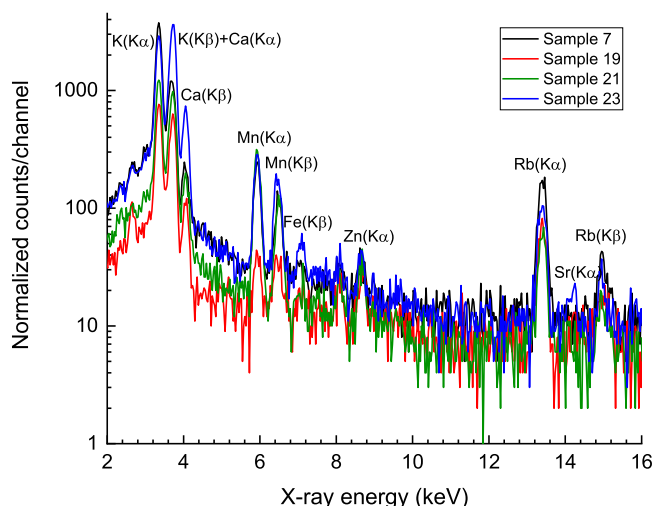


Fig. 3. Particle induced X-ray spectra from bilberry samples. Sample 19 contains overall less detectable elements for some unknown reason. Bilberries in sample 23 were picked from a site known for a calcium carbonate-rich soil. The spectra were normalized with respect to the integrated charge.

1986 (STUK, 2020). The concentration of Cs-137 was measured with an Ortec GMX HPGe detector. To optimize the measuring geometry, the samples were loaded into small Petri dishes (10 × 35 mm), which were positioned against the endcap of the detector. A lead shielding of 5 cm thickness was built around the detector to reduce the background in the spectra and thereby lower the detection limits. A calibrated Cs-137 point source (Amersham, CDR 206, Sn 2847) was applied for the efficiency calibration at 10 cm. Sample 22 was measured at 10 cm, then loaded into a Petri dish and used for calibration of the optimized geometry. The achieved detection limit of the Cs-137 activity concentration was about 0.004 Bq/g when the acquisition time was set to one day and the sample size was 1 g. The uncertainty of the Cs-137 activity measurement was then around 12 % (3–24 %).

3. Results

All elemental concentrations showed a weak positive correlation between each other and a negative correlation to the water content due to variations in the total mineral content between the samples (e.g., K-Ca 0.36, Mn-Fe 0.32, water%-K -0.27; Table 1). The variation of elemental concentrations between the bilberry samples clearly exceeded the variation expected due to the uncertainty of the analytical method or to inhomogeneous elemental distributions within the sample. The analytical precision for the lightest measurable elements (P, S, Cl) was affected by a change in geometry during irradiation and they were excluded from the results. The sample material expanded towards the proton beam, forming millimetre-sized peaks or towers (Fig. 4). This phenomenon was earlier observed upon irradiating honey to measure the elemental concentration (Saarela et al., 1998). The effect on the reported concentration is thus small (<6 %).

The elemental concentration of manganese ranges from 3 µg/g up to 63 µg/g. Large differences in the manganese concentration were observed within small regions e.g., the elemental concentration in bilberries collected from sampling site 8 was about twice as high as in bilberries collected from sampling site 9, although there was only a distance of 20 m between the sites and both sites were located in a clear-cut area (Table 1). The manganese concentration of sample 8 was 49

Table 1
Elemental concentration and corresponding uncertainties in fresh bilberries mainly sampled in Ostrobothnia at the Finnish west coast (Fig. 1). Activity concentrations of Cs-137 are also listed for some of the samples. The last row shows the part of the Recommended Dietary Intake (RDA; male > 19 years old) achieved by eating 100 g of fresh bilberries.

Sample No.	Water (%)	Elemental concentrations (µg/g)																		Cs-137 (Bq/g)		
		K	Ca	Mn	Fe	Cu	Zn	Rb	Sr													
1	85.7	897 ± 21	314 ± 8	22 ± 1	4.8 ± 0.6	0.9 ± 0.1	0.7 ± 0.1	4.8 ± 0.3	0.3 ± 0.1													
2	86.2	1121 ± 28	466 ± 12	20 ± 1	5.9 ± 0.5	1.0 ± 0.1	0.7 ± 0.1	10.1 ± 0.4	0.5 ± 0.1													
3	85.9	770 ± 22	493 ± 14	38 ± 2	6.0 ± 0.6	0.7 ± 0.1	0.9 ± 0.1	3.5 ± 0.3	0.9 ± 0.2													
4	82.8	1036 ± 31	990 ± 30	30 ± 2	27.8 ± 1.3	0.7 ± 0.2	1.5 ± 0.1	5.3 ± 0.4	1.1 ± 0.2	0.013 ± 0.003												
5	82.3	483 ± 19	264 ± 11	14 ± 1	2.8 ± 0.6	0.3 ± 0.1	0.5 ± 0.1	7.3 ± 0.4	0.3 ± 0.2													
6	87.5	695 ± 23	262 ± 9	28 ± 1	2.2 ± 0.5	0.6 ± 0.1	0.6 ± 0.1	2.7 ± 0.3	0.9 ± 0.2													
7	84.4	1273 ± 35	242 ± 9	47 ± 2	6.1 ± 0.8	0.6 ± 0.1	0.7 ± 0.1	11.2 ± 0.6	bdl	0.025 ± 0.003												
8	84.5	935 ± 16	549 ± 10	49 ± 2	6.6 ± 0.7	0.7 ± 0.1	1.2 ± 0.1	5.1 ± 0.3	0.2 ± 0.2													
9	84.0	501 ± 19	220 ± 9	22 ± 1	3.4 ± 0.7	0.4 ± 0.1	0.8 ± 0.1	3.5 ± 0.3	0.4 ± 0.2													
10	83.7	867 ± 33	465 ± 18	22 ± 2	5.1 ± 0.7	0.8 ± 0.2	1.1 ± 0.1	2.2 ± 0.3	0.4 ± 0.2													
11	85.2	216 ± 11	213 ± 10	8 ± 1	5.4 ± 0.6	0.8 ± 0.2	0.5 ± 0.1	2.5 ± 0.3	0.3 ± 0.2	0.021 ± 0.003												
12	88.1	842 ± 39	390 ± 19	15 ± 1	4.4 ± 0.6	0.4 ± 0.1	0.7 ± 0.1	3.8 ± 0.4	0.4 ± 0.2	0.007 ± 0.002												
13	86.9	401 ± 14	302 ± 11	46 ± 2	4.3 ± 0.6	0.3 ± 0.1	0.6 ± 0.1	3.3 ± 0.3	0.5 ± 0.2	0.016 ± 0.002												
14	82.0	728 ± 29	521 ± 21	44 ± 2	8.4 ± 0.9	1.2 ± 0.2	1.3 ± 0.2	4.1 ± 0.4	0.7 ± 0.2													
15	85.1	476 ± 20	496 ± 20	29 ± 2	13.6 ± 1.0	0.5 ± 0.2	0.7 ± 0.1	2.4 ± 0.3	0.5 ± 0.2													
16	91.6	109 ± 3	78 ± 2	24 ± 1	2.7 ± 0.3	0.2 ± 0.1	0.5 ± 0.0	1.3 ± 0.1	0.3 ± 0.1													
17	90.1	800 ± 24	772 ± 23	10 ± 1	9.9 ± 0.6	0.6 ± 0.1	1.3 ± 0.1	4.1 ± 0.3	1.1 ± 0.2													
18	83.1	545 ± 27	1062 ± 51	19 ± 1	13.5 ± 1.0	0.5 ± 0.1	0.8 ± 0.1	3.6 ± 0.3	0.8 ± 0.2													
19	90.1	142 ± 3	83 ± 2	3 ± 0	1.5 ± 0.2	0.3 ± 0.1	0.3 ± 0.1	2.6 ± 0.1	0.2 ± 0.1	0.005 ± 0.001												
20	88.2	300 ± 12	267 ± 10	11 ± 1	5.5 ± 0.5	0.3 ± 0.1	0.5 ± 0.1	3.4 ± 0.2	0.5 ± 0.1													
21	84.2	411 ± 19	235 ± 11	63 ± 3	5.0 ± 0.7	0.6 ± 0.1	0.8 ± 0.1	3.6 ± 0.3	bdl	0.088 ± 0.008												
22	82.4	963 ± 28	110 ± 5	8 ± 1	2.0 ± 0.5	1.0 ± 0.1	0.7 ± 0.1	8.0 ± 0.5	bdl	0.132 ± 0.007												
23	85.6	681 ± 15	705 ± 15	38 ± 1	11.2 ± 0.7	0.5 ± 0.1	0.6 ± 0.1	4.5 ± 0.3	0.4 ± 0.2	bdl												
Mean	85.6	660 ± 316	413 ± 266	27 ± 16	6.9 ± 5.7	0.6 ± 0.3	0.8 ± 0.3	4.5 ± 2.5	0.5 ± 0.3	0.036 ± 0.044												
LOD		6.4	7.4	1.3	1.2	0.2	0.2	0.2	0.2	0.004*												
RDA (%)		2	4	115	9	0.0	0.7															

The limit of detection (LOD) is calculated for a typical fresh bilberry sample.
Concentrations below the limit of detection are marked bdl (below detection limit).
*LOD for the activity of Cs-137 is presented for 1 g sample and 1 day acquisition time.

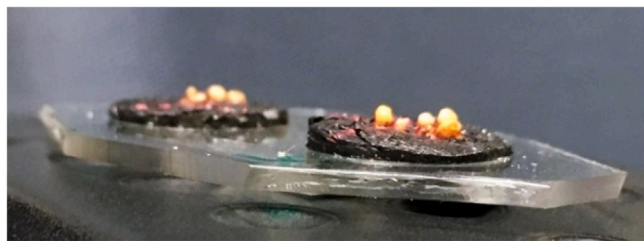


Fig. 4. Irradiated pellets of pressed bilberry powder after proton irradiation. The proton beam causes the sugar in the samples to expand, forming the small towers seen in the picture.

$\pm 2 \mu\text{g/g}$ of fresh weight and the corresponding value of sample 9 was $22 \pm 2 \mu\text{g/g}$.

The Recommended Dietary Allowance (RDA) of manganese is 2.3 mg/day for males (>19 years old) and 1.8 mg/day for females (>19 years old; Food and Nutrition Board, National Academies, 2019b). By consuming 100 g of fresh Finnish bilberries, this recommendation will be reached. Furthermore, the Tolerable Upper Intake Level (UL) is set to 11 mg/day (Food and Nutrition Board and National Academies, 2019a), which can be exceeded only in rare cases, as one has to consume more than 400 g of fresh berries in one day. These limits are, however, for healthy adults (>19 years old). In the case of children, the limits of tolerable upper intake are much lower: for 1–3 years only 2.0 mg/day, for 4–8 years 3.0 mg/day, and for 9–13 years 6.0 mg/day (Food and Nutrition Board and National Academies, 2019a). Räsänen and Nuurtamo (1981) have calculated the mean intake of manganese from ordinary food to be already 5.9 mg/day for 9–13 year old children in Finland. Taking into consideration the large regional differences in elemental concentrations, an overdose of manganese from wild bilberries is possible. Especially in the case of young consumers. The dried blueberry powder available at health shops should also be tested before being sold for human consumption in larger amounts.

The regional distribution of Cs-137 follows the activity distribution of the fallout and correlates with earlier measured activities in mushrooms (Lönnroth et al., 2011). The mean activity concentration of Cs-137 in fresh bilberries was $0.04 \pm 0.04 \text{ Bq/g}$. The large variation is due to high values in the two samples from the Jakobstad region (samples 21 and 22; Table 1), an area known from earlier studies to be contaminated with fallout from the Chernobyl accident (Lönnroth et al.,

2011; STUK, 2023). Omitting these two extreme samples would drastically lower the mean value ($0.015 \pm 0.007 \text{ Bq/g}$). Assuming an annual consumption of 2 kg of fresh blueberries containing 0.04 Bq/g will lead to an annual intake of 80 Bq of Cs-137. By using the conversion coefficients of oral intake for adults (Ministry of Social Affairs and Health, Finland, 2018), the equivalent dose was estimated to be 1 μSv . This means that from a Cs-137 radiological point of view, it is safe to eat bilberries.

The results of the PIXE analyses agreed well with the earlier reported elemental concentrations (Table 2). The mean value of manganese was 27 $\mu\text{g/g}$, which agreed well with values reported by Koivistoinen et al. (1974) and Ekholm et al. (2007) but was clearly lower than that of Tahvonen (1993). The bilberries analysed by Tahvonen were picked somewhere in the northern and eastern part of Finland - the detailed geographical information was not reported. The bilberries from Finland contain in general more manganese than the berries from Latvia, Poland, and Montenegro. The wild bilberries analysed by the Montenegro group were picked in a mountain region about 1200 m above sea level. The concentration of most elements was clearly elevated compared to concentrations in berries from the other countries in Table 2 except for the manganese concentration that was even lower than in berries from Latvia and Poland.

4. Conclusion

Particle Induced X-ray emission was applied to the elemental analysis of pressed pellets of dried bilberries. The wild-grown bilberries were picked at 23 sites in Finland, mainly at a site on the west coast near the town of Vaasa. The manganese concentration in the fresh berries was found to be high, $27 \pm 16 \mu\text{g/g}$, and already by an oral intake of 100 g of fresh bilberries, the recommended total daily intake for adults will be exceeded. Children are more sensitive and by eating the same amount of bilberries, the limit of tolerable upper intake can be reached. Already the mean intake of manganese from ordinary food will be close to the upper level.

The mean Cs-137 concentration in the fresh bilberries was $0.04 \pm 0.04 \text{ Bq/g}$. The value was higher in two samples from an area contaminated with fallout from the Chernobyl accident. The equivalent dose to an adult by an annual consumption of 2 kg of fresh bilberries was estimated to be 1 μSv . One can thereby conclude that from a Cs-137 radiological point of view, it is safe to eat wild-grown bilberries from Finland.

Table 2
Comparison with elemental concentrations in fresh wild bilberries from Europe found in literature.

Studies	Method	Country	n	Elemental mean concentrations of fresh weight ($\mu\text{g/g}$)							
				K	Ca	Mn	Fe	Cu	Zn	Rb	Sr
This study	PIXE	Finland	23	660	413	27	6.9	0.61	0.8	4.5	0.5
SD				± 316	± 266	± 15	± 5.7	± 0.26	± 0.3	± 2.5	± 0.3
Koivistoinen et al., 1974	FAAS	Finland	20	1000	170	35	4.4	0.80	1.4		
Tahvonen, 1993	FAAS	Finland	5	1010	230	68	4.2		1.4		
Ekholm et al., 2007	ICP-MS	Finland	1	768	158	33	3.5	0.57	0.9		
SD*				± 16	± 7	± 1.4	± 0.4	± 0.03	± 0.1		
Karlsons et al., 2018	FAAS	Latvia	1	1108	218	17.2	3.8	0.72	1.5		
SE				± 89	± 37	± 3	± 4	± 0.08	± 0.2		
Sketers et al., 2014	ICP-OES	Latvia	3	482	160	18.8	1.7	0.31	1.6		0.063
SD				± 16	± 10	± 0.5	± 0.06	± 0.02	± 0.2		± 0.004
Drózdź et al., 2018	ICP-OES	Poland	12	1028	186	17.6	8.6	0.31	4.8		
SD				± 24	± 4	± 0.5	± 0.2	± 0.01	± 0.2		
Brašanac-Vukanović **	ICP-MS	ME***	5	5290	651	13.61	32.5	5.11	7.24		1.32
SD				± 50	± 8	± 0.02	± 0.5	± 0.07	± 0.04		± 0.02

* Estimated from repeated analyses of CRM (n=7)

** Brašanac-Vukanović et al., 2019

*** Montenegro.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data Availability

Data will be made available on request.

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