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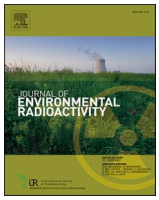
Journal of Environmental Radioactivity 261 (2023) 107134

Contents lists available at ScienceDirect

Journal of Environmental Radioactivity

journal homepage: www.elsevier.com/locate/jenvrad



Experimental determination of concentration factors of Mn, Zn and I in the phytoplankton species Phaeodactylum Tricornutum

K. Insulander Björk [a,*], R. Thomas [a], S. Holgersson [b], M. Isaksson [a]

[a] Department of Medical Radiation Sciences, Institute of Clinical Sciences, Sahlgrenska Academy, University of Gothenburg, Sweden
[b] Division of Energy and Materials, Department of Chemistry, Chalmers University of Technology, Sweden

ARTICLE INFO

Handling Editor: Dr S.C. Sheppard

ABSTRACT

Anthropogenic radionuclides released into the environment cause a radiation dose to wildlife and humans which must be quantified, both to assess the effect of normal releases, and to predict the consequences of a larger, unplanned release. To estimate the spread of the radioactive elements, the ecosystem around release points is modelled, and element uptake is usually quantified by concentration factors (CF), which relates the concentration of an element in an organism to the concentration of the same element in a medium under equilibrium conditions. In this work, we experimentally determine some phytoplankton CF that are needed for improved modelling of the marine ecosystems around nuclear facilities and release points. CFs that require better determination have been identified through literature search. Sensitivity studies, using the currently used ecosystem modelling software PREDO, show that for most studied groups, the dose committed by the respective radionuclides is almost proportional to the corresponding phytoplankton CFs. In the present work, CFs are determined through laboratory experiments with cultured phytoplankton and radionuclides of the concerned elements, assessing the element uptake by the phytoplankton through detection of the emitted radiation. The three CF assessed in this work were those for manganese, zinc and iodine in phytoplankton. Conservative estimates of these CF based on the present data are 40 000 L/kg for manganese, 50 000 L/kg for zinc and 180 L/kg for iodine with the phytoplankton masses referring to their dry weight.

1. Introduction

Nuclear power plants (NPPs) and other nuclear facilities during normal operation release small amounts of radioisotopes, e.g. activation products such as Mn-54 and Co-60, and fission products such as Cs-137 and Sb-124. The resulting radiation dose to the public is assessed to ensure that it falls below regulatory limits. This is done through modelling of the ecosystem and the transport of radionuclides through the food web and other possible exposure pathways. Concentration factors (CFs) are important model parameters (IAEA 2014), relating the concentration of a certain element in a living organism to the corresponding concentration in the surrounding medium under equilibrium conditions. The Swedish nuclear facilities routinely perform these dose assessments using the software PREDO (deWith et al., 2015) which implements a box model for the marine ecosystem.

The International Atomic Energy Agency (IAEA) and the International Commission on Radiological Protection (ICRP) have published several reports (IAEA 2004; IAEA 2014; ICRP 2009) with recommended values of CFs for a wide range of combinations of elements and organisms. The Wildlife Transfer Database (Copplestone et al., 2013) continuously collects published CF data. However, it is recognized that these recommended values still suffer from a large degree of uncertainty. The large variability in CF values reported in the literature is often due to insufficiently documented experimental conditions (IAEA 2014; ICRP 2009). In many cases the recommended CF values are estimations, based on assumptions of biochemical similarity between elements and/or organisms (Beresford et al., 2016). It is also clear that uncertainties in the CFs have a significant impact on the modelling results (Periáñez et al., 2019). For example, the experimental data on phytoplankton CFs for Mn, reported in (IAEA 2014) comprises six samples from two different studies with a spread in CF from 20 L/kg to 5000 L/kg fresh weight, and the same CF cited in (IAEA 2004) is 50 000 L/kg fresh weight.

Furthermore, CFs are to some extent affected by several environmental factors, such as daylight, temperature and bioavailability of the element, which in turn depend on the element's speciation and hence the chemical composition of the local environment (Masood Khan et al.,

* Corresponding author.
E-mail address: klara.insulander.bjork@gu.se (K. Insulander Björk).

https://doi.org/10.1016/j.jenvrad.2023.107134
Received 10 March 2022; Received in revised form 16 September 2022; Accepted 3 February 2023
Available online 17 February 2023

2017). The presence of nutrients also affects the uptake of elements in biota (Wang and Dei, 2001). Thus, CFs determined for an element and organism in one environment may have limited applicability in a different environment, which is why we seek to mimic the conditions in temperate waters and use authentic seawater samples, including both saline and brackish water to identify any significant differences. For ecosystem modelling, it should be checked that the experimental CF data used is representative for the modelled environment, which may lead to more conservative dose estimates as compared to using, for instance, the mean value of all published CF values.

According to a screening performed during the development of PREDO only the 15 most dose-relevant nuclides (11 elements) contribute more than 99% of the calculated dose (deWith et al., 2015). These are ^{108m}Ag, ^{110m}Ag, ^{243}Am, ^{58}Co, ^{60}Co, ^{134}Cs, ^{137}Cs, ^{59}Fe, ^{3}H, ^{54}Mn, ^{238}Pu, ^{124}Sb, ^{125}Sb, ^{90}Sr and ^{65}Zn. The screening included several different nuclear facilities, including NPPs, a nuclear fuel factory and a nuclear waste handling facility, and is thus probably representative of most nuclear facilities worldwide. The reliability of the published phytoplankton CFs for the relevant elements was assessed based on 1) the IAEA technical documents 422 (IAEA 2004) and 479 (IAEA 2014), 2) the current version of the Wildlife Transfer Database, and 3) a literature search. In addition, the CF data for iodine was assessed in the same way, since this is a volatile element released in nuclear reactor accidents and nuclear weapon detonations, with radionuclides ^{129}I and ^{131}I which have a significant impact on dose calculations in emergency situations.

Based on these investigations, it was concluded that the CFs in phytoplankton for Mn, Zn and I are both dose-relevant and determined to a dissatisfying extent. Therefore, the aim of this work is to experimentally determine CFs for phytoplankton under controlled conditions for Mn, Zn and I. The published CF data for these elements is summarized in Table 1 and the entries are discussed in further detail below.

1.1. Manganese

CF data for Mn is reported in IAEA TECDOC 479 for six samples (Ancellin et al., 1979; Polikarpov 1966), with determined CFs ranging from 20 L/kg fresh weight to 5000 L/kg fresh weight whereas IAEA TECDOC 422 reports a much higher value of 50 000 L/kg. Fisher (1985) reports an experimentally determined vol/vol CF of 1000 for Mn in the cyanobacterium *Synechococcus* and Lee and Fisher (1993) report a vol/vol CF of 9000 in *T. Pseudonana*. Smythers et al. (2019) found the Mn CF in phytoplankton to be concentration dependent, decreasing with increasing Mn concentration. At the lowest investigated Mn concentration (70 μM), the CF that can be calculated from their data is approximately 40 L/kg fresh weight.

Table 1
Literature values of phytoplankton concentration factors for Mn, Zn and I. All CFs are given in L/kg fresh weight. For the data from the IAEA TECDOCs, primary sources are stated although the samples are listed jointly. For each source, the arithmetic mean value, the standard deviation (STD) and the number of samples (N) are listed if stated in the source.

	Mean	STD	N
Manganese			
IAEA TECDOC 422 (IAEA 2004) (Eisler 1981)	$5{\cdot}10^4$		
IAEA TECDOC 479 (IAEA 2014) (Ancellin et al., 1979; Polikarpov 1966)	$3.5{\cdot}10^3$	$3.5{\cdot}10^3$	6
Fisher (1985) (*Synechococcus* sp., vol/vol CF)	$1{\cdot}10^3$		
Smythers et al., (2019) (at 70 μM Mn)	40		4
Lee and Fisher (1993) (*T. Pseudonana*, vol/vol CF)	$9.0{\cdot}10^3$		3
Zinc			
IAEA TECDOC 422 (IAEA 2004) (Fisher and Reinfelder, 1991)	$1{\cdot}10^4$		5
Chouvelon et al., (2019)	$1.3{\cdot}10^2$		
Fisher et al., (1984) (*T. Pseudonana*, vol/vol CF)	$12{\cdot}10^3$		
Fisher (1985) (*Synechococcus* sp., vol/vol CF)	$3{\cdot}10^4$		
Iodine			
IAEA TECDOC 479 (IAEA 2014) (Bowen 1979)	$9.5{\cdot}10^2$		1
Wilson et al. (2007)	$1.4{\cdot}10^2$		
Coughtrey et al. (1984) and Kuenzler (1967)	$1.2{\cdot}10^3$		

1.2. Zinc

IAEA TECDOC 479 does not contain any information about CF for Zn. The earlier, similar, TECDOC 422 (IAEA, 2004) recommends a value of 50 000 L/kg fresh weight, referring to Bowen (1979) and Eisler (1981). Chouvelon et al. (2019) report Zn concentrations in seawater and mixed plankton in different size fractions, from which a CF of approximately 130 L/kg fresh weight can be calculated. Fisher et al. (1984) report an experimentally determined vol/vol CF for Zn in the diatom *T. Pseudonana* of 12 000 L/kg fresh weight.

1.3. Iodine

The CF data for I is very scarce. IAEA TECDOC 479 (IAEA 2014) refers only to one analysed sample (Bowen 1979), in which the CF was found to be 950 L/kg fresh weight. Wilson et al. (2007) determined the CF to 141 L/kg, although it is not clear whether the number refers to fresh weight or dry weight. Coughtrey et al. (1984) and Kuenzler (1967) report values with a mean of 1200 L/kg fresh weight.

2. Method

2.1. Computational sensitivity studies

The software PREDO (deWith et al., 2015) was used to assess to which extent the committed dose from each of the investigated radionuclides is a result of exposure through the food chain, relative to other exposure pathways. If other exposure pathways would dominate, the phytoplankton CFs would be of less importance.

The default PREDO calculation gives the yearly dose after 100 years of release of 1 Bq/year, for later normalization to the actual releases. This dose was calculated for five different human population groups (fishing, hunting, farming, vegetarian and dairy farming families). In particular, the PREDO model for the aquatic environment surrounding the Ringhals NPP at the west-coast of Sweden, was used. For each population group, the dose was calculated for the adult, child and infant subcategories. The calculations were made with the currently implemented concentration factors, and with the same concentration factors increased and decreased by one order of magnitude, respectively. The calculations were made for only one nuclide of each of the studied elements: I-131, Mn-54 and Zn-65, since the same CF applies to all radionuclides.

2.2. Seawater sampling and analysis

Seawater samples were collected by the Swedish Meteorological and Hydrological Institute (SMHI). The sampling was performed in February 2022, at the sampling stations Anholt E (Kattegat – saline water, water depth 63 m, sampling depth 50 m) and Karlsödjupet (Baltic sea – brackish water, water depth 110 m, sampling depth 80 m), see Fig. 1. The seawater salinity was 33‰ at Anholt E and 10‰ at Karlsödjupet, according to the characterization made by SMHI.

The natural concentrations of the investigated elements were measured by the standard addition method (Christian and O'Reilly, 1986) using ICP-MS (Thermo iCAP Q) and corrected for instrumental background. The samples were diluted 1.7 × with 0.5 M HNO_3 (Suprapur Merck), containing internal standards of 2 ppb Sc, In and Bi. Standards of 0, 1, 5 and 10 ppb were prepared from 10 ppm stock solutions (UltraScientific, CPA Chem, VGA Labs). Mass intensity readings

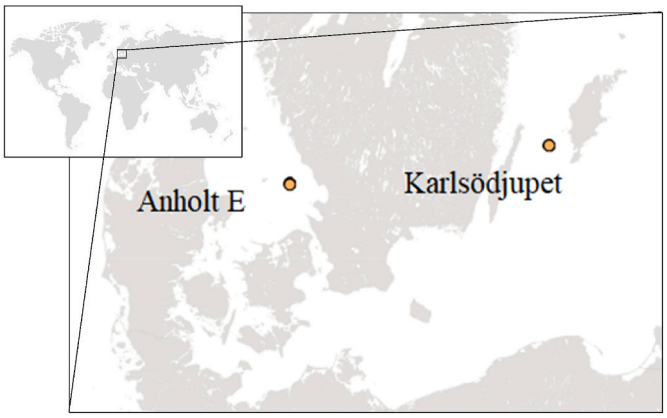


Fig. 1. The geographical location of the sampling stations Anholt E (in Kattegat – saline water) and Karlsödjupet (in the Baltic Sea – brackish water).

for Mn, Zn and I at respective masses 55, 66 and 127 were taken five times to generate average value and standard deviation.

2.3. Phytoplankton cultures

Diatoms are by far the most common phytoplankton in the seas around Sweden (SIME - Swedish Institute for the Marine Environment, 2019). A non-axenic strain of the diatom *Phaeodactylum Tricornutum* was procured from the algal bank at the University of Gothenburg.[1] The bacteria were enumerated by counting under the microscope and found to be approximately one attached bacterial cell per 10 algal cells in a stationary culture, and no free-living bacteria were observed. Given the small size of the bacterial cells compared with the algal cells, it was assumed that their contribution to the total biomass was negligible, which was further supported by the measured algal cell dry weight. *P. Tricornutum* is an atypical diatom species in that it has a very lightly silicified frustule (Fisher et al., 1991), which is not, however, necessarily important for radionuclide accumulation characteristics.

Immediately before inoculation with phytoplankton cultures, the seawater was filtered through a 0.2 μm polycarbonate membrane filter (Cytiva Whatman™ Cyclopore™) to remove debris and micro-organisms. The phytoplankton were cultured in 50 mL polystyrene cell culture flasks with vented caps (Thermo Scientific™ Nunc™ EasY-Flask™, Nunclon™ Delta surface plasma treated), containing 29 mL of the seawater and 1 mL of stock phytoplankton culture. Nutrients ($8.8\cdot10^{-4}$ M $NaNO_3$, $3.6\cdot10^{-5}$ M NaH_2PO_4 and $1.1\cdot10^{-4}$ M Na_2SiO_3) were added in proportions according to Guillard (1975), however no vitamins or trace elements were added, since initial trials showed that this was not necessary for phytoplankton growth. EDTA was not added.

Radionuclides were added as the dissociated salts of $MnCl_2$, $ZnCl_2$ and NaI respectively. The Mn-54 and Zn-65 (Eckert & Ziegler Strahlen- und Medizintechnik AG, Germany) sources had a carrier concentration of 10 μg/mL of stable isotopes of the respective element. I-131 was in a sodium iodide solution intended for injection for medical use (GE Healthcare Buchler GmbH & Co. KG, Germany) with no stable carrier. The added concentrations of radionuclides and stable carrier atoms are listed in Table 2. The added amount of stable carrier increased the concentrations of Mn and Zn significantly above the natural levels (see Table 3). All three radionuclides were added in combinations to the same samples and were analysed in parallel. Initial trials with separate samples for each radionuclide confirmed that this would not affect the results, i.e. that the presence of a high concentration of one element did not affect the uptake of the other elements.

The concentration of the considered radionuclides was also varied, to investigate whether the radioactivity as such would have any effect on the phytoplankton growth or on the uptake of the considered elements.

Identical control samples with the same radionuclide and nutrient concentrations, but with 30 mL of seawater and no phytoplankton, were prepared for checking that only negligible amounts of radioactivity were sorbed onto filters when no phytoplankton were present in the solution.

To assess the amount of the radionuclides passively adsorbed on the phytoplankton, samples with phytoplankton and nutrients were prepared with no radionuclides. When the phytoplankton concentration had reached approximately 10^6 cells/mL, the samples were refrigerated to 1 ± 1 °C in the dark for 24 h, which stopped the phytoplankton growth and metabolism. After this, radionuclides were added and after 24 h of equilibration in the same cold and dark conditions, the samples were analysed in the same manner as the samples with growing phytoplankton.

Triplicate samples of each type were prepared for statistical purposes.

In this work, light- and temperature conditions similar to those found in temperate regions were considered. The samples were placed under controlled light and temperature conditions. Light was provided 12 h/day with cold-white (6000 K) LED illumination at an intensity of 200 $\mu Em^{-2}s^{-1}$, similar to the conditions used by e.g. Heldal et al. (2001) and Fisher and Reinfelder (1991). The temperature varied slightly with the light, increasing from 20 °C at the beginning of the light period, reaching 25 °C towards the end and then decreasing down to 20 °C again during the dark period. Preliminary trials were also made at lower temperatures (5°C–15 °C) and with constant gentle shaking, however, neither of these conditions did affect the results.

Phytoplankton concentration and growth was assessed using an Automated Cell Counter (Countess, ThermoFisher Scientific). A 100 μL aliquot from each culture flask was collected for phytoplankton counting at approximately two-day intervals, until a phytoplankton concentration of 10^6 cells/mL was reached, which generally occurred within 6–8 days. The samples were fixated with 2 μL of acidic Lugol's solution before counting.

The pH of the cultures was monitored throughout the experiment by the use of pH indicator sticks (Fisherbrand®, pH Indicator Paper Sticks, pHix 0–14) and remained constantly at 8 ± 0.5.

Table 2
Added radionuclide and stable carrier concentrations.

	Initial radionuclide concentration [MBq/L]	Initial radionuclide concentration [pM]	Added stable carrier concentration [nM]
Manganese	0.1	0.65	18 000
Zinc	0.1	0.51	15 000
Iodine	1	0.017	0

Table 3
Measured concentrations of the considered elements. The given uncertainties are one standard deviation.

Element	Mn	Zn	I
Measured element concentration at Anholt E [nM]	9.1 ± 1.8	84 ± 5	350 ± 20
Measured element concentration at Karlsödjupet [nM]	15 ± 4	52 ± 2	248 ± 4

2.4. Phytoplankton dry weight determination

Phytoplankton dry weight was assessed using the method of Fisher and Schwarzenbach (1978) with the modification that polycarbonate (Cytiva Whatman™ Cyclopore™, pore size 1 μm, diameter 25 mm) instead of glass-fibre filters were used (Heldal et al. 2001) to reduce the adsorption of sea salts. The filters were pre-soaked in NH_4HCO_2

[1] https://www.gu.se/en/marina-vetenskaper/about-us/algal-bank-gumacc.

(Ammonium formate, Acros Organics, 99%) solution and dried. After filtering (transmembrane pressure 17 kPa), the filters (and deposited phytoplankton) were rinsed twice with 5 mL of NH_4HCO_2 solution, isotonic with the sea water.

The filters were weighed immediately after drying overnight at 60 °C (ensuring a complete evaporation of the ammonium formate), the total phytoplankton dry weight ($m_{plankton}$) was calculated as the mass gain of the filters and the dry weight per cell was calculated based on the phytoplankton concentration at the last count before filtration. The mass gain of the filters used for filtering the control samples was negligible.

2.5. Concentration factor determination

For determination of concentration factors, separate phytoplankton cultures were grown and then treated similarly to the ones used for dry weight determination, however, the filters were now rinsed with sterile filtered seawater using the method described by Fisher et al. (1983). The CF were determined at the final time point by filtering of the entire 30 mL batch of phytoplankton culture.

After rinsing, the filters were immediately placed in plastic containers and the activity on each filter ($A_{plankton}$) was measured with gamma spectrometry. The detector system consisted of a high-purity germanium (HPGe) coaxial detector (Ortec GEM 50P4) with a relative efficiency of 52%, connected to a digital signal processor (DSPEC jr 2.0) providing an energy resolution of 1.65 keV at 1.33 MeV (Ametek, Inc., Oak Ridge, TN, USA). The phytoplankton-free control samples were filtered, rinsed and measured in an identical manner, and the average activity adsorbed on these filters ($A_{control}$) was subtracted from the total activity on the corresponding phytoplankton sample filters. The activity of the filtrate (A_{water}) was also measured for cross-checking the radionuclide concentration in the medium. The volume of the filtrate was measured to ensure that correct geometrical efficiency and dilution corrections were made. The volume of the filtrate before rinsing of the filter (V_{water}) was also measured for the CF determination. The filtrates were contained in cylindrical plastic containers and their activity measured in the same manner as that of the filters.

The gamma counting time was chosen to ensure that the counting uncertainty was less than 3%. Geometric efficiency corrections were made using efficiency transfer from a calibration source measured in a similar container, however with a different fill height. The geometric efficiency corrections for the filters were made assuming a cylindrical plastic beaker geometry as for the filtrates, however with a 20 mm diameter (i.e. the filter area upon which the phytoplankton were deposited) and a filter thickness ("fill height") of 0.5 mm. Variations of the filter thickness below 1 mm, or variations in the filter composition assumed for the efficiency transfer calculations had an impact on the efficiency smaller than 1% for the energies of interest. The EFFTRAN v. 4.2 computer code (Vidmar 2005) was used for the efficiency transfer calculations.

Concentration factors were calculated as

$$CF = \frac{A_{plankton} - A_{control}}{m_{plankton}} \left(\frac{A_{water}}{V_{water}} \right)^{-1} \quad (1)$$

where the notation used is that described in the text above.

The activity of radionuclides sorbed onto the culture flasks was measured using the same HPGe detector. The activity determination precision was low due to the difficulty of making adequate geometrical efficiency corrections for the non-cylindrical culture flasks. However, this activity was found to be more than two orders of magnitude smaller that the activity in the filtrate, i.e. the fraction of the radioisotopes sorbed on the culture flask walls was negligible. The flasks were not rinsed, since that would release radionuclides adsorbed onto the surfaces which would not have been available to the suspended phytoplankton.

The filtrate was checked for any remaining cells which may have passed around filter edges, but none were observed in any of the five investigated filtrate samples. However, a slight colouring of the filtrate was observed, indicating that some cell walls may have broken, possibly releasing their content into the filtrate. No clear connection was found between the measured activity of the cells and the observed colouring of the filtrate.

3. Results and discussion

3.1. Computational sensitivity studies

Radiation doses were calculated for the five representative population groups (fishing, hunting, farming, vegetarian and dairy farming families) and for the three age subcategories (adult, child and infant). Given that the calculations were made for a hypothetical release of 1 Bq/year, the numbers are only relevant for intercomparison. The results are shown in Fig. 2. The results for the farming family and the dairy farming family were identical and are therefore presented jointly as "farming". For all studied elements (I, Mn and Zn) and population groups, the infant age category received the highest dose, and for all studied elements and age categories, the fishing family received the highest dose.

In general, the calculated dose is almost directly proportional to the used value of the CF, indicating that a major part of the dose is committed via the marine food chain, presumably via intake of seafood which are in turn directly or indirectly dependent on phytoplankton as a food source. The vegetarian family is an exception – for this family, the dose is completely independent on the concentration factors, indicating that they only receive dose through dwelling in or at the sea, presumably since their diet does not include fish. Since dose is strongly coupled to the diet of a population it is clear that phytoplankton CFs for the investigated elements are essential for the calculation of doses to most population groups. The habitat and diet of each model family is described in deWith et al. (2015).

3.2. Element concentrations in seawater samples

The concentrations of Mn, Zn and I, as determined by ICP-MS analysis, are listed in Table 3. The measured concentrations of iodine are similar to previous measurements in Skagerrak (Truesdale et al., 2003) and the Baltic Sea (Truesdale et al., 2001). The measured concentrations of manganese were in the range of previous measurements at different depths at Bornholm, situated approximately halfway between the water sampling sites (Kremling and Petersen, 1978), and at 6 m depth at different locations within the Baltic Sea (Kremling and Streu, 2000). The measured concentrations of zinc were a few times higher than previously measured values (Kremling and Streu, 2000), possibly due to contamination during sampling.

3.3. Phytoplankton dry weight

The average dry weight of phytoplankton in all 25 analysed samples was 23 pg. It did not differ significantly (by more than two standard deviations) between the water from the two different sampling sites, and the variation in phytoplankton mass between sample sets with different additions of Mn, Zn and I respectively was also not statistically significant, (the measured dry masses from the different sample sets differed by less than one standard deviation). The measured dry mass is slightly lower than the 35 pg/cell that can be calculated from the data reported by Fábregas et al. (1996), but virtually identical to the 22.6 pg/cell measured by Fisher et al. (1991). The organic mass according to Cresswell (2010) is 23 pg/cell for *P. Tricornutum*, which seems to imply that the results with respect to the phytoplankton dry weight are reasonable. This measured value also supports our assumption that the contribution of the bacterial cells in the culture is negligible compared with the algal cells.

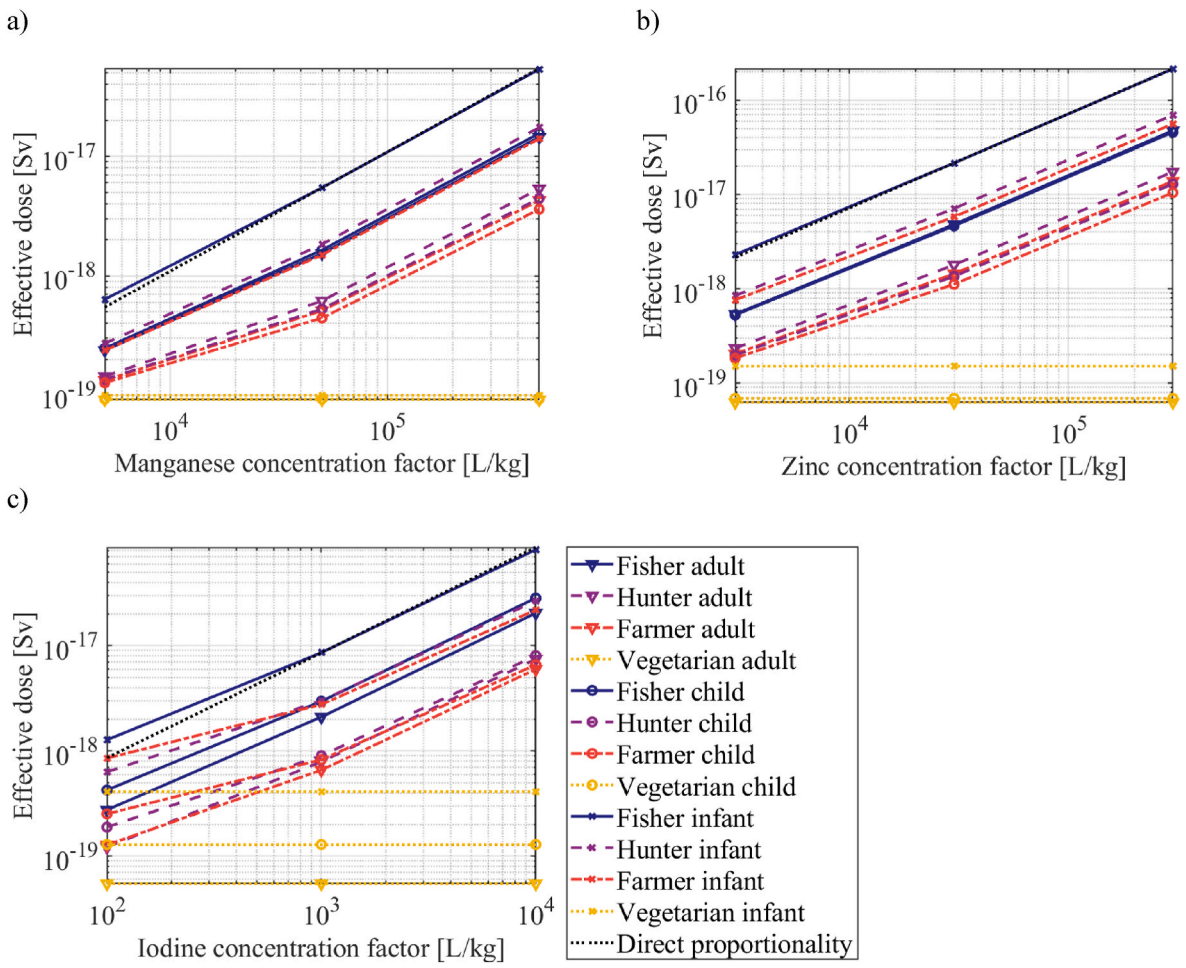


Fig. 2. Effective dose committed to each of the representative families listed in the plot legends (fishing, hunting, farming and vegetarian) and each age category (adult ▼, child ● and infant ×), calculated for a) Mn-54, b) Zn-65 and c) I-131. In each plot, a black dashed line representing direct proportionality has been added for reference.

3.4. Concentration factors

Concentration factors were calculated separately for each sample, using equation (1). Mean values and standard deviations are listed in Table 4, and discussions follow below. For Mn and Zn, the average activity measured on control filters was less than 0.9% of the average activity measured on the filters with growing as well as senescent phytoplankton. For I, the activity measured on control filters was 27% and 1.4% for growing cells in the Anholt and Karlsödjupet water respectively, and the corresponding numbers for senescent cells were 196% and 36% respectively, which is the reason why no CF value is given for senescent cells in the Anholt water in Table 4.

3.4.1. Manganese

The calculated manganese CF in phytoplankton is 7200 L/kg fresh weight for the cells grown in water from the Anholt sampling station. This lies within the wide range of previously published values (20–50 000 L/kg fresh weight) and is much lower than the currently implemented 50 000 L/kg fresh weight in PREDO. The adsorption on senescent cells is very small in comparison with the active uptake. The Mn CF for phytoplankton grown in the water from Karlsödjupet sampling station was significantly lower: 1800 L/kg (more than two times the standard deviation).

3.4.2. Zinc

The calculated zinc CF in phytoplankton is about 3200 L/kg fresh weight for active uptake in water samples from the Anholt sampling station. This is lower than most of the published data (60 000–300 000 L/kg by Bowen (1979); Eisler (1981); Fisher et al. (1984)), but higher than the 700 L/kg dry weight found by Chouvelon et al. (2019) for mixed plankton. The adsorption on senescent cells is once again much lower than the active uptake. Finally, the accumulation is similar at in the water samples from Karlsödjupet: 9100 L/kg, i.e. the difference is less than two standard deviations.

3.4.3. Iodine

The calculated iodine CF in phytoplankton is about 33 L/kg fresh weight for active uptake in water samples from the Karlsödjupet sampling station. This is much lower than the previously reported literature data, where most results are about 1000 L/kg fresh weight, except for the result by Wilson et al. (2007) of 141 L/kg (probably referring to fresh weight). The passive uptake is approximately 80% lower than the active uptake. For the Anholt E sampling station, where the natural iodine concentration is about double that at Karlsödjupet, the CF is only about 20% (5 L/kg) of the CF for Karlsödjupet. No previous results imply that this should be due to the varying iodine concentrations, but other factors could also affect the uptake.

In general, the measured CFs are lower than previously published literature data. This could be due to *P. Tricornutum* generally accumulating less Mn, Zn and I than previously studied phytoplankton species. Another possible reason for this could be that cell walls broke during the filtration process, releasing their contents into the filtrate, which would

Table 4
Calculated concentration factors for *P. Tricornutum* and the investigated elements, calculated as the average over the three analysed samples. The given uncertainties are one standard deviation. CFs with standard deviations are given in L/kg dry weight (dw), rounded to two significant digits. Relative standard deviations (RSD) are given in % calculated from non-rounded figures. CFs for growing cells are also given in L/kg fresh weight (fw) for comparison with literature values below, where the conversion factor of 0.18 dry weight to fresh weight recommended by IAEA (IAEA 2014) has been used.

Element	State of cells	Water origin	CF (dw)	Standard deviation (dw)	RSD	CF (fw)
Manganese	Growing	Anholt E	40 000	14 000	36%	7200
		Karlsödjupet	10 000	4000	39%	1800
	Senescent	Anholt E	1000	300	29%	
		Karlsödjupet	1000	300	26%	
Zinc	Growing	Anholt E	18 000	4000	25%	3200
		Karlsödjupet	50 000	23 000	45%	9100
	Senescent	Anholt E	3900	2700	69%	
		Karlsödjupet	6600	5100	78%	
Iodine	Growing	Anholt E	27	13	49%	4.9
		Karlsödjupet	180	69	38%	33
	Senescent	Anholt E	[a]			
		Karlsödjupet	4.3	2.1	49%	

[a] The activity on the filters with the senescent cells was similar to the activity on the control filters, so no CF could be calculated.

lead to some of the activity originally associated with the cells being measured in the filtrate and not on the filter.

4. Conclusions

Computational sensitivity studies using the aquatic PREDO model for the Ringhals NPP show that phytoplankton CFs are important for calculation of the dose committed to most exposed groups through release to water of radioisotopes of iodine, manganese and zinc. This is assumed to be due to the fact that the exposure to these elements is dominated by intake via the food chain where phytoplankton are the primary producers.

Phytoplankton dry weights are in good agreement with literature data on the investigated phytoplankton species; the diatom *P. Tricornutum*, indicating that the used method is working well. Phytoplankton CFs for iodine, manganese and zinc were investigated and conservative estimates of the CFs for the investigated elements, i.e. the highest values measured for the respective elements (mean of three samples) were: 7200 L/kg for manganese, 9100 L/kg for manganese and 33 L/kg for zinc, all referring to fresh weight. Lower values were found for accumulation in water of different origin and for non-growing (senescent) phytoplankton, so doses calculated using the recommended values above are likely to be conservative.

Irrespective of the state of cells (growing or senescent) or seawater origin, the CFs calculated from the present data are significantly lower than those currently implemented in PREDO and those recommended by the IAEA (IAEA 2014), so the dose calculations performed today for Swedish waters are most likely conservative.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

Klara Insulander Björk reports financial support was provided by Swedish Radiation Safety Authority. Klara Insulander Björk reports financial support was provided by Wilhelm and Martins Lundgrens foundation. Klara Insulander Björk reports financial support was provided by Herbert and Karin Jacobssons foundation. Klara Insulander Björk reports financial support was provided by The Royal Society of Arts and Sciences in Gothenburg.

Data availability

Data will be made available on request.

Acknowledgments

This work was funded by the Swedish Radiation Safety Authority under contract number SSM 2020-642-24. Additional funding for equipment was received from Wilhelm and Martina Lundgrens foundation, Herbert and Karin Jacobssons foundation and the Royal Academy of Arts and Sciences in Gothenburg. The authors are grateful for useful advice and help from Olga Kourchenko at the Gothenburg University algal bank GUMACC, Kristin Andreasson at SMHI, Maria Davidsson at the Ringhals NPP and Elin Lindehoff at the Linné University. Special thanks are directed to Nicholas Fisher at the State University of New York for invaluable advice on CF determination.

References

Ancellin, J., Guegueniat, P., Germaine, P., 1979. Radioecologie Marine. Eyrolles, Paris.
Beresford, N.A., Wood, M.D., Vives i Batlle, J., Yankovich, T.L., Bradshaw, C., Willey, N., 2016. J. Environ. Radioact. 151 (2), 373–386.
Bowen, H.J.M., 1979. Environmental Chemistry of the Elements. Academic Press, London.
Chouvelon, T., Strady, E., Harmelin-Vivien, M., Radakovitch, O., Brach-Papa, C., Crochet, S., Knoery, J., Rozuel, E., Thomas, B., Tronczynski, J., Chiffoleau, J.-F., 2019. Patterns of trace metal bioaccumulation and trophic transfer in a phytoplankton-zooplankton-small pelagic fish marine food web. Mar. Pollut. Bull. 146, 1013–1030.
Christian, G.D., O'Reilly, J.E., 1986. Instrumental Analysis". Allyn and Bacon Inc., MA, USA.
Copplestone, D., Beresford, N.A., Brown, J.E., Yankovich, T., 2013. An international database of radionuclide concentration ratios for wildlife: development and uses. J. Environ. Radioact. 126, 288–298.
Coughtrey, P.J., Jackson, D.J., Jones, C.H., Thorne, M.C., 1984. Radionuclide Distribution and Transport in Terrestrial and Aquatic Ecosystems - A Critical Review of Data, vols. 1–6. A. A. Balkema, Rotterdam.
Cresswell, L., 2010. Phytoplankton Culture for Aquaculture Feed". *SRAC Publications* No, p. 5004.
deWith, G., Maderich, V., Willemsen, S., Bezhenar, R., 2015. PREDO – Methodology for the Assessment of Radioactive Releases to the Aquatic Environment, QP.50000-63747891 V. 2.0. Vattenfall, Stockholm.
Eisler, R., 1981. Trace Metal Concentrations in Marine Organisms. Pergamon, New York.
Fábregas, J., Patiño, M., Morales, E.D., Cordero, B., Otero, A., 1996. Optimal renewal rate and nutrient concentration for the production of the marine microalga Phaeodactylum tricornutum in semicontinuous cultures. Appl. Environ. Microbiol. 62 (1), 266–268.
Fisher, N.S., Schwarzenbach, R.P., 1978. Fatty acid dynamics in thalassiosira pseudonana (bacillariophyceae): implications for physiological ecology. J. Phycol. 14 (2), 143–150.
Fisher, N.S., Bjerregaard, P., Fowler, S.W., 1983. Interactions of marine plankton with transuranic elements. I. Biokinetics of neptunium, plutonium, americium, and californium in phytoplankton. Limnol. Oceanogr. 28, 432–447.
Fisher, N.S., Guillard, R.R.L., Bankston, D.C., 1991. The accumulation of barium by marine phytoplankton grown in culture. J. Mar. Res. 49, 339–354.
Fisher, N.S., Bohe, M., Teyssie, J.-L., 1984. Accumulation and toxicity of Cd, Zn, Ag, and Hg in four marine phytoplankters. Mar. Ecol. Prog. Ser. 18, 201–213.
Fisher, N.S., 1985. Accumulation of metals by marine picoplankton. Mar. Biol. 87, 137–142.
Fisher, N.S., Reinfelder, J.R., 1991. Assimilation of selenium in the marine copepod Acartia tonsa studied with a radiotracer ratio method. Mar. Ecol. Prog. Ser. 70, 157–164.
Guillard, R.R.L., 1975. Culture of phytoplankton for feeding marine invertebrates. In: Smith, W.L., Chanley, M.H. (Eds.), Culture of Marine Invertebrate Animals. Springer, Boston, MA.
Heldal, H.E., Stupakoff, I., Fisher, N.S., 2001. Bioaccumulation of ^{137}Cs and ^{57}Co by five marine phytoplankton species. J. Environ. Radioact. 57, 231–236.
IAEA, 2004. Sediment Distribution Coefficients and Concentration Factors for Biota in the Marine Environment. IAEA, Vienna. Technical Report No. 422.
IAEA, 2014. Handbook of Parameter Values for the Prediction of Radionuclide Transfer to Wildlife. IAEA, Vienna. Technical Report No. 479.
ICRP, 2009. Environmental protection: transfer parameters for reference animals and plants. ICRP publication 114, ann. ICRP 39 (6).
Kremling, K., Petersen, H., 1978. The distribution of Mn, Fe, Zn, Cd and Cu in Baltic seawater; a study on the basis of one anchor station. Mar. Chem. 6 (2), 155–170.

Kremling, K., Streu, P., 2000. Further evidence for a drastic decline of potentially hazardous trace metals in Baltic Sea surface waters. Mar. Pollut. Bull. 40 (8), 674–679.

Kuenzler, E.J., 1967. Elimination of iodine, cobalt, iron, and zinc by marine zooplankton. In: Nelson, D.J., Evans, F.C. (Eds.), *Proc. 2nd National Symposium on Radioecology*. Ann Arbor, Michigan, May 15e17, 1967. USAEC Conf. No. 670503, 462e473.

Lee, B.-G., Fisher, N.S., 1993. Microbially mediated cobalt oxidation in seawater revealed by radiotracer experiments. Limnol. Oceanogr. 38, 1593–1602.

Masood Khan, A., Abu Bakar, N.K., Abu Bakar, A.F., Aqeel Ashraf, M., 2017. Environ. Sci. Pollut. Res. 24, 22764–22789.

Periáñez, R., Bezhenar, R., Brovchenko, I., Duffa, C., Iosjpe, M., Jung, K.T., Kim, K.O., Kobayashi, T., Liptak, L., Little, A., Maderich, V., McGinnity, P., Min, B.I., Nies, H., Osvath, I., Suh, K.S., de With, G., 2019. Environ. Model. Software 122, 104523.

Polikarpov, G.G., 1966. Radioecology of Aquatic Organisms: the Accumulation and Biological Effects of Radioactive Substances. Reinhold, New York.

SIME - Swedish Institute for the Marine Environment, 2019. Sveriges vattenmiljö, tillståndsrapport växtplankton. retrieved. https://www.sverigesvattenmiljo.se/sa-mar-vara-vatten/2019/variabelgrupper/85/12/62. (Accessed 17 March 2020).

Smythers, A.L., Perry, N.L., Kolling, D.R.J., 2019. Chlorella vulgaris bioaccumulates excess manganese up to 55× under photomixotrophic conditions. Algal Res. 43, 101641.

Truesdale, V.W., Nausch, G., Baker, A., 2001. The distribution of iodine in the Baltic Sea during summer. Mar. Chem. 74 (2–3), 87–98.

Truesdale, V.W., Danielssen, D.S., Waite, T.J., 2003. Summer and winter distributions of dissolved iodine in the Skagerrak. Estuar. Coast Shelf Sci. 57 (4), 701–713.

Vidmar, T., 2005. Efftran - a Monte Carlo efficiency transfer code for gamma-ray spectrometry. Nucl. Instrum. Methods 550, 603–608.

Wang, W.-X., Dei, R.C.H., 2001. Effects of major nutrient additions on metal uptake in Phytoplankton. Environ. Pollut. 111, 233–240.

Wilson, R.C., Vives i Batlle, J., Watts, S.J., McDonald, P., Parker, T.G., 2007. Uptake and depuration of 131I from labelled diatoms (Skeletonema costatum) to the edible periwinkle (Littorina littorea). J. Environ. Radioact. 96 (1–3), 75–84.