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Note

Rapid Tritium Analysis for Marine Products in the Coastal Area of Fukushima

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The analysis of tritium in aquatic biota is one of the most important research areas in Fukushima. The conventional method for measuring the concentration of tritium consists of complicated pretreatment procedures and requires skillful techniques as well as a significant amount of time. Consequently, there are only a few reports on tritium monitoring data in marine products from the coast of Fukushima. In this study, we measured the Tissue Free Water Tritium (TFWT) and Organically Bound Tritium (OBT) in flounders collected from the coast of Fukushima to examine the impacts of the nuclear accident on aquatic biota. The study was done for a period of 4 years; from 2015 to 2018. The conventional method of analysis was firstly used, after which the method was modified by improving the freeze-drying and combustion water recovery processes. Results from both methods show that the most of the concentrations of the TFWT and OBT in the flounder were below the detection or quantitative limit. The effect of the nuclear accident on humans, through internal exposure, was also examined and found to be negligible. Although some uncertainties exist due to the short cut of the processes, the modified version could be considered an effective and practical approximate method.

Key words: sample pretreatment, conventional method, oxygen combustion vessel, dose assessment, Japanese flounder

1. Introduction

Tritium, an isotope of hydrogen is a beta-emitting radionuclide with a half-life of 12.3 years. It emits a maximum energy of 18.6 keV. In living organisms, tritium exists in two basic forms; tissue free water tritium (TFWT) and organically bound tritium (OBT)[1]. OBT can be further divided into exchangeable OBT (E-OBT) and non-exchangeable OBT (NE-OBT)[2]. Specifically, NE-OBT exists in the form of organic matter in muscles and fats. It has a lower rate of metabolism and a longer biological half-life compared to the TFWT[3, 4]. It is, therefore, important to monitor its impacts on the biological system.

The Ministry of Education, Culture, Sports, Science and Technology (MEXT) manual (currently inherited by Nuclear Regulation Authority)[5] has been established as the standard measurement scheme of tritium (referred to,

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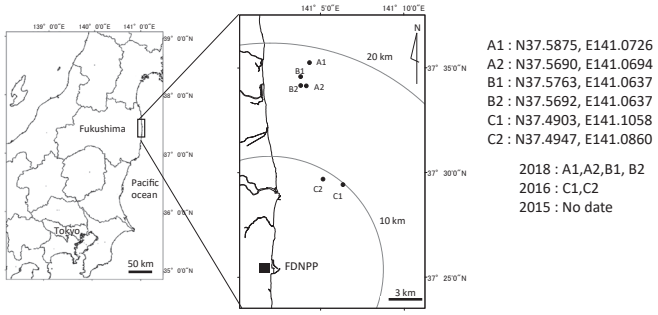

Fig. 1. Location of study area in Fukushima Japan, and collection points of Japanese flounder in the Fukushima coastal area (black point). The location of the Fukushima Daiichi Nuclear Power Plant (FDNPP) is indicated by black square.

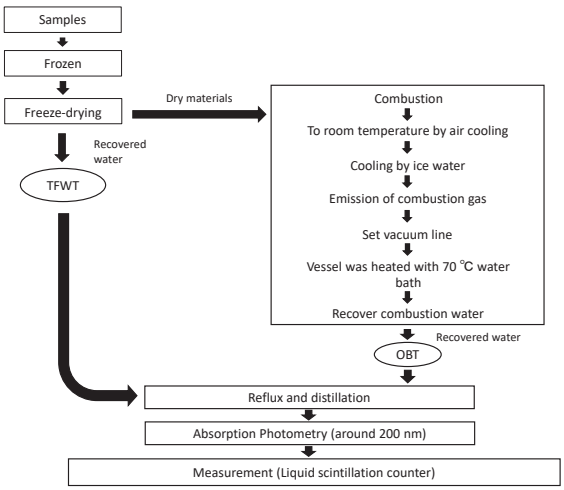

Fig. 2. Conventional tritium measurement method, CM.

in this paper, as the conventional method, CM) in Japan, This is comparable to the scheme used worldwide[6]. The scheme consists of freeze-drying, combustion, purification of combustion water, preparation of liquid sample, and measurement by the LSC. Such complicated pretreatments, however, require skillful techniques and a significant amount of time for analysis. For this reason, only a few institutions have the capacity to measure the tritium in biological samples, hence, only a limited number of data is available[2].

In comparison with other radionuclides, such as ^{137}Cs and ^{129}I, the concentration of tritium in the Fukushima coastal ocean has remained low since the Fukushima Dai-ichi Nuclear Power Plant (FDNPP) accident[7, 8]. One month after the accident, a water sample taken from the coastal sea surface near the Hirono-town, which is about 20 km south of FDNPP, had a tritium concentration of 5.0 Bq/L[9], whereas, the highest concentration recorded from 2013 to 2016 near the Fukushima coast was 6.2 Bq/L. At present, the concentration has gone below the detection limit[10]. However, there is still a need for evaluation of the effect of the FDNPP accident on aquatic biota. It was reported that TFWT was still in detectable range from 2014 to 2017, whereas, 0.5 Bq/L OBT was detected, in 2013, only in the Marbled sole flounder around the Tohoku Pacific Coast[11]. Therefore, there is a need to survey the trends of radioactive effects on fishes in the long term.

In this study, we measured the TFWT and OBT concentration in flounders collected from the Fukushima costal area, to investigate the effect of the FDNPP accident on marine products. In the case of such an accident, it is very important to provide data, and also, evaluate the environmental impacts as soon as possible. Deriving many approximate concentration values with practical methods is therefore, desirable. The oxygen combustion vessel (Model1121, Parr Instruments, USA)[12] was used, instead of the tube furnace combustion system[4], due to its higher rate of combustion. Here, we propose a improved method which could further reduce the pretreatment time. This proposed method is named Rapid Method (RM). We analyzed generally, the OBT without specifying the E-OBT and NE-OBT.

In this note, we report our method of analysis and the obtained results; only new results are presented. Lastly, we evaluated the resulting internal exposure to humans based on the results of the TFWT and OBT concentrations in the marine products in Fukushima.

2. Method

2.1. Sample preparation

This study was carried out for a period of 4 years; from 2015 to 2018. Flounders from the sea of the Fukushima Daiichi Nuclear Power Station were used as the experimental samples. The samples were collected from the sea, within a sea area of 20 km. Figure 1 shows the outline of this study area and the location of sampling points. Twenty kg (about 20 fish, and approximately 50 cm) of flounder were collected within a bottom net. The collected fishes were put into a 3-layer walled plastic bags to keep them out of direct contact with ice and air. The bones were then, carefully removed from each of the fish samples, leaving out only the fleshy parts. These were cut into pieces; each measuring about 1 cm^3. The pieces were properly mixed such that the batch of the sample becomes evenly. These were sealed with aluminum packs and stored in a freezer. The samples were allowed to freeze to a temperature of -40 ℃ or less.

2.2. Procedure

2.2.1. Analysis of Tritium concentration by CM

We first reviewed the analytical scheme of measuring

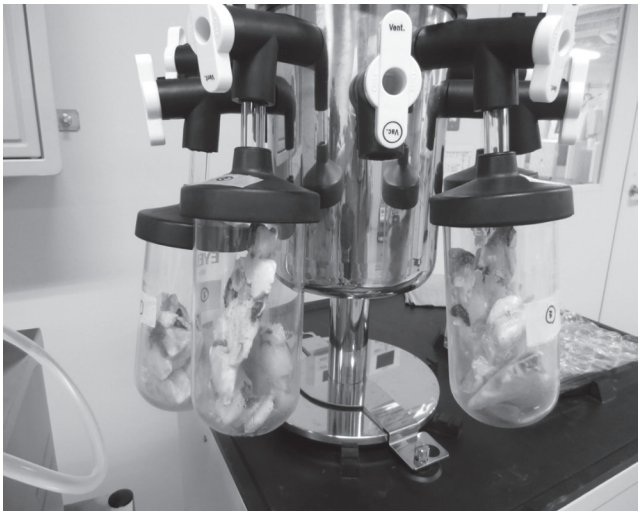

Fig. 3. Multiple drying containers in our tritium measurement pretreatment system.

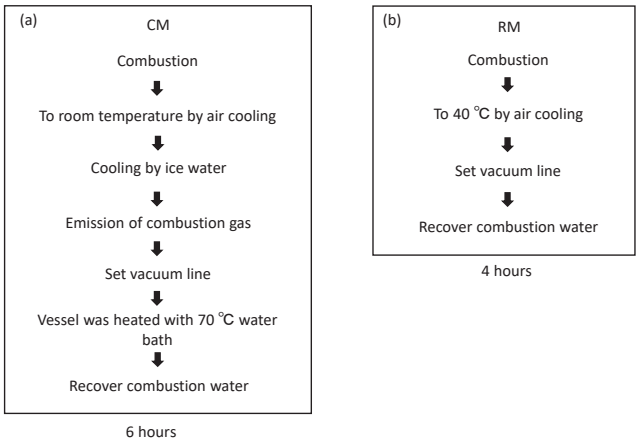

Fig. 4. Flowchart of gas recovery after combustion. CM requires 6 hours to recover the combustion gas (a), and RM can recover combustion gas in 4 hours (b).

tritium. Figure 2 shows a schematic representation of the workflow. The overall procedures can be summarized as follows:

The TFWT was extracted from the frozen samples by the freeze-drying system, leaving behind, the freeze-dried fish tissue for the OBT analysis. The extracted TFWT was further refined by reflux and distillation. The freeze-dried tissue, on the other hand, was combusted using oxygen combustion vessel and the combustion water collected. The OBT was prepared by refining the collected water also by reflux and distillation. The prepared TFWT and OBT were analyzed using absorption photometry (UV-1800, SHIMADZU, Japan) to confirm the decomposition of the organic matter. The TFWT and OBT were further analyzed using the low-background LSC (LSC-LB7, HITACHI, Japan) using 100 ml and 20 ml vials. The External Standard Channel Ratio method was used to correct the quenching effects on the sample. Ultima Gold uLLT was used as a cocktail. The mixing ratio of the sample and the cocktail was adjusted to 1:1. When the 100 ml vial is used, the mixing volume was 50 ml : 50 ml. When 20 ml vial is used, that volume was 10 ml :10 ml. The uncertainties of the pipette are 3.6% for a 100 ml vial and 1.6% for a 20 ml vial.

Each measurement was performed for 600 minutes. The results of the detection limit (DL) value were measured by the LSC and calculated using the MEXT formula. The calculated values were further converted to Bq/kg-fresh using the moisture content of the sample for TFWT and the hydrogen content of the sample measured by Elemental Analyzer (MT-6, Yanako, Japan) for OBT.

The above-described method has been used so far for the analysis of the tritium concentration. However, it takes about one month and two months to analyze TFWT and OBT, respectively. This is evidenced in this study; the extraction of the TFWT from the frozen samples and its separation from the freeze-dry took about 6 days for 500 g of samples whereas, the combustion of the freeze-dried tissue and the collection of the combustion water took about 6 hours for a 10 g of sample. Therefore, to run these processes several times, several working days would be required to terminate.

In emergency cases, an order of magnitude evaluation is firstly required (accurate measurement may be done subsequently) and the time for analysis need to be shortened. As previously stated, in such cases, evaluation of the total OBT is an important and accurate value of individual NE-OBT and E-OBT, which is difficult to evaluate with such shorten processes, may be neglected. The total OBT is, therefore, analyzed by taking into consideration, the potential uncertainties due to the shortening. By modifying the freeze-drying process, and the combustion water recovering process in CM, we may perform the pretreatment more efficiently and practically.

2.2.2. Analysis of tritium by RM

In the drying process, the adequate size of the sample to be dried was unknown, thus we varied the size of sample and measured the drying time. Since our system has multiple drying containers, as shown in Figure 3, we could divide the sample into smaller batches and treat all the samples at the same time. The samples were also cut into 1 cm^3 dice form to enhance drying.

In the combustion water recovery process by CM, the oxygen combustion vessel was cooled in air after combustion, after which it was further cooled in ice. The carbon dioxide and generated combustion gases were discharged until the pressure inside the vessel

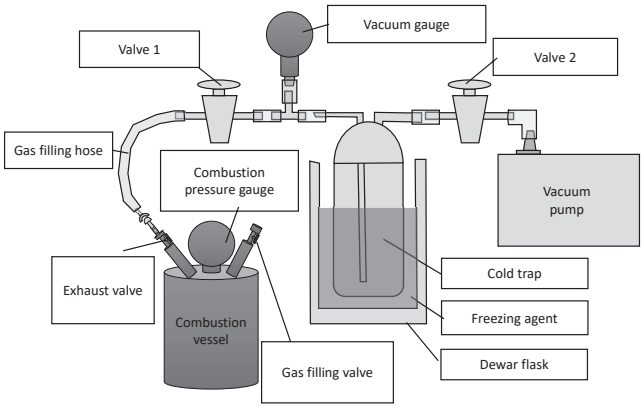

Fig. 5. System configuration of gas recovery process after combustion.

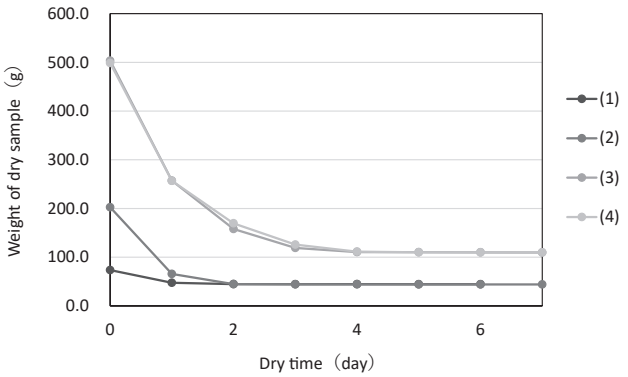

Fig. 6. Changes in dry weight by freeze-drying.

Table 1. Results of TFWT and OBT measurements of flounder using CM. TFWT and the most of OBT were below the detection and quantitative limits. DL were basically calculated as 3σ

	Collected date	Vial size	TFWT(Bq/kg-fresh)		OBT(Bq/kg-fresh)	
			concentration	DL	concentration	DL
CM-1	2015.11.25	100ml	ND	0.84[a)]	ND	0.15[a)]
CM-2	2016.12.22	100ml	ND	0.19	0.04 ± 0.02	0.04
CM-3	2016.12.22	20ml	ND	1.02	ND	0.18

Note : a. CM-1:These results were calculated as quantitative limit(10σ).

approached the atmospheric pressure. Furthermore, the combustion water was recovered as water vapor while the water is being heated in a water bath to a temperature of about 70℃. It took about 6 hours to combust and recover a dry sample of about 10 g. NH_4 ion was detected in the combustion gas, hence, tritium could be lost in this method. For this reason, we tried to eliminate the gas release by connecting the recovery line while the equipment was warm after combustion in RM. In this way, we can simultaneously recover the combustion gases and water, as shown in Figures 4 and 5.

The followings are the newly obtained procedures (modified procedures are *italicized*):

Freeze-drying

A) *After pre-freezing the sample at -40℃ or lower, divide the sample to be freeze-dried at about 200 g per bottle and prepare multiple lines.*
B) Freeze-drying (FDU-2110, EYELA, Japan) until the sample reaches to a constant weight.

Combustion

A) Oxygen combustion vessel is assembled and checked for leakage using nitrogen gas.
B) Measure approximately 10 g of the sample and set it in the sample container of the oxygen combustion vessel.
C) Measure the electrical resistance and confirm that the Nichrome wire is energized.
D) Inject about 2.1 Mpa of oxygen gas, ignite with electricity, and combust.
E) Check the pressure gauge and cool it for about 30 minutes after combustion.

Recovery of combustion water

A) *Put calcium chloride solution into the Dewar flask and make a cold trap with about 1 kg of finely crushed dry ice.*
B) *Attach a cold trap and vacuum pump to the combustion water recovery line to make the vacuum state.*
C) *Connect oxygen combustion vessel to the recovery line and recover the entire amount of combustion gas over 2 hours.*
D) *Heat the combustion water and collect it in the cold trap.*

3. Results and Discussions

3.1. TFWT and OBT concentrations by CM

Table 1 shows the results of TFWT and OBT concentrations analyzed using CM. From the results, the concentrations of tritium in all the TFWT and the most of OBT were below the detection or quantitative limit. The data for the samples collected and analyzed in 2015 were evaluated

Table 2. Results of TFWT and OBT measurements of flounder using RM. Both TFWT and OBT were below the detection limit. DL were calculated as 3 σ

	Collected date	Vial size	TFWT(Bq/kg-fresh)		OBT(Bq/kg-fresh)	
			concentration	DL	concentration	DL
RM-1	2016.12.22	20ml	ND	0.96	ND	0.16
RM-2	2016.12.22	20ml	(ND)[a)]	(0.96)	ND[b)]	0.73
RM-3	2018.12.18	20ml	ND	0.90	ND[b)]	0.84

Notes : a. The sample of 2016.12.22 is indicated by ( ) because these are same analysis sample.
b. Results in RM-2, RM-3 are obtained by diluting the combustion water to 25 ml (collected after one combustion).

Table 3. Time (days) needed to dry sample by freeze drying. When the total amount is 200 g, the drying time was shortened to 4 days

	Total amount (g)	Freeze-drying		
		Drying time (day)	Weight (g)	Ratio of TFWT (%)
(1)	203.7	4	44.5	78.2
(2)	202.5	4	44.2	78.3
(3)	503.3	5	109.9	78.2
(4)	498.5	6	109.8	78.0

with a quantitative limit because the tritium analysis of marine products in this year had already been initiated, and the uncertainties of pretreatments were considered to be large.

3.2. TFWT and OBT Concentrations in by RM

Table 2 shows the results of TFWT and OBT concentrations analyzed using the RM. In these results, the TFWT and OBT in flounder are observed to be below the detection limit. The measurement conditions of the LSC are the same as that of the CM.

3.3. Reduction of time in freeze-drying and combustion water recovering procedures

As earlier stated, to increase the drying efficiency, we adjusted the suited amount of sample to be filled into the container by changing the amount of sample per bottle which results in a change of the surface area of the sample. When CM was used, an average of 6 days was spent in drying about 500 g per bottle. However, samples filled in 200 g per bottle were dried to a constant weight in 4 days (Table 3, Figure 6). From the above results, it is inferred that reducing the number of samples filled per sample bottle and reduces the freeze-drying time, hence it is an effective way for shortening the drying time.

For the combustion water recovering process using RM, the combustion water recovery line was connected, about 20 minutes after the combustion while the vessel' s surface temperature was still about 40 °C. With this method, the combustion water was able to be recovered in 2 to 3 hours; about half the time required for the CM.

3.4. Indirect Evaluation of RM

Due to the low tritium concentration in samples, the validity of the RM analysis cannot be directly ascertained. Thus, to validate this new procedure, the recovery rate and the amount of chemical compositions were compared with that of the CM,

Table 4 shows the recovery rates of both methods. As can be observed from the table, there is no significant difference in the rates of combustion water recovery for the CM and RM.

Furthermore, 12 kinds of ions were measured by ion chromatography dual system (Prominence, SHIMADZU, Japan) to discern the difference in the chemical compositions of the combustion water treated by CM and that treated by RM. In both methods, the combustion water was reflux and distilled, and the complete removal of the organic materials from the samples was confirmed. As shown in Table 5, CM and RM recorded almost the same value for each of the ions. Although some uncertainties such as incomplete CO_2 exclusion may exist, RM may be considered an effective and practical approximate method.

3.5. Dose Assessment

We applied the following widely used equation for the dose assessment:

$$H = m \times d \times p \times a$$

where H is the committed effective dose (mSv), m the ingestion amount (kg-fresh/day), d the ingestion day (day), p the effective dose coefficient (TFWT: 1.8×10^{-8} mSv/Bq, OBT: 4.2×10^{-8} mSv/Bq), and a the radioactivity

Table 4. Recovery rate of combustion water. The results of four measurements by the CM is shown in (a), and the results of four measurements by the RM is shown in (b)

(a)	CM	(1)	(2)	(3)	(4)
	Dry material (g)	10.2	10.1	10.5	10.2
	Recovery water (g)	5.5	4.6	4.3	4.5

(b)	RM	(1)	(2)	(3)	(4)
	Dry material (g)	10.1	10.2	10.3	10.2
	Recovery water (g)	5.2	5.5	4.6	4.7

Table 5. Result of ion chromatography data. Twelve ions were measured for combustion water treated by CM and RM. Both combustion waters were refluxed and distillated

Sample (flounder)	F^-	Cl^-	NO_2^-	Br^-	NO_3^-	PO_4^{3-}	SO_4^{2-}	Na^+	NH^{4+}	K^+	Mg^{2+}	Ca^{2+}
CM	< 0.1	0.13	< 0.5	< 0.5	< 0.5	< 1	< 0.5	0.28	< 0.05	< 0.1	< 0.2	< 0.2
RM	0.11	0.15	< 0.5	< 0.5	< 0.5	< 1	< 0.5	0.35	< 0.05	< 0.1	< 0.2	< 0.2

Table 6. Result of committed effective dose assuming 100 g of flounder were eaten a year using each detection or quantitative limit

	Collected date	Vial size	TFWT(mSv)	OBT(mSv)
CM-1	2015.11.25	100 ml	5.5E-07	2.3E-07
CM-2	2016.12.22	100 ml	1.2E-07	6.1E-08
CM-3	2016.12.22	20 ml	6.7E-07	2.8E-07
RM-1	2016.12.22	20 ml	6.3E-07	2.5E-07
RM-2	2016.12.22	20 ml	6.3E-07	1.1E-06
RM-3	2018.12.18	20 ml	5.9E-07	1.3E-06

concentration (Bq/kg-fresh)[3].

When the measured concentration was lower than the detection or quantitative limit, the committed effective dose was calculated using the limits. The result of the calculations of the commissioned execution dose, with the assumption that 100 g[12] of flounder are eaten a year, are shown in Table 6. As shown in the table, there is no significant difference in the value of the committed effective dose calculated for the CM and RM. Since both are in nSv level, it is considered that the effects on humans due to internal exposure are very low, hence, negligible.

4. Conclusion and Remarks

In this study, we focused on determining the tritium concentration in marine products in the Fukushima costal area.

Result obtained show low tritium concentration in flounder from off the coast of Fukushima Prefecuture; most of the TFWT and OBT were below the detection or quantitative limit. The tritium ingestion dose to humans due to internal exposure was also evaluated using the detection limit. Very low dose (nSv levels) were obtained, hence, the contribution of internal exposure is negligible. A reduction in the freeze-drying time was achieved by reducing the size of the samples, thereby, increasing the surface area. In an attempt to improve the freeze-drying process, and the recovery of the combustion water, we introduced the RM technique. This method was able to reduce the recovery time by two. Combustion water recovered by RM was, however, characterized; the properties are in good agreement with that recovered by the conventional CM. We, therefore, infer that the proposed method, RM, is an effective and improved method of combustion water recovery.

In this note, the most of the results were under detection limit, but we are also measuring other materials that possess more tritium. We have contributed the inter-laboratory OBT comparison exercises this year with RM, and although we cannot indicate the results here since the official reports are yet unpublished (to be published in 2020), we internally confirmed in the OBT workshop[6] that our RM (without the drying processes) satisfactorily produced suitable values for fruit samples (Quinces-Cydonia oblonga). In the next step, by using such results and others (plants, marine products, fruits), we will accumulate convincing evidences to prove the effectiveness of the RM.

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Conflict of Interest

The authors declare that they have no conflicts of interest.

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