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Human exposure risk via algae-induced transfer of tritiated water in the marine food chain

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Understanding the health impact of human radiation exposure from tritiated water release is crucial for the management and sustainability of nuclear energy. However, it remains not fully explored owing to the neglect of bioconversion products (organically bound tritium, OBT) along the food chain. Here by evaluating the uptake and chemical transfer of tritium in biota, we show the critical role of algae in rapidly incorporating and transferring tritiated water into OBT, which serves as nutrients for trophic transfer to fish. Notably, the specific retention of OBT in the fish brain, by integrating into biomolecules, potentially disrupts key metabolic reactions. The derived concentration factors and biomagnification factors are instrumental in estimating the internal exposure dose to human individuals, thereby enabling more accurate risk assessments for both planned tritium releases and accidental leakages. This work highlights the importance of comprehensive evaluation and mitigation of tritium exposure risks.

As one of the most abundant nuclides in liquid radioactive effluents from nuclear power plants, tritium (^{3}H) cannot be effectively removed by separating tritiated water (HTO) from H_2O using current technologies, owing to its chemical similarity to hydrogen (^{1}H)[1–4]. The annual global anthropogenic release of ^{3}H is approximately 200 g (7.2×10^{16} Bq), resulting in ^{3}H concentrations in nearby waters that far exceed background levels[5]. Public concern over radioactive exposure has recently intensified following the announcement of Fukushima's wastewater treatment plan, which involves discharge of over 1 million tons of ^{3}H-containing wastewater into the ocean beginning in August 2023[6–12]. In the early stages of the Fukushima nuclear accident, a considerable amount of ^{3}H ($3–7 \times 10^{14}$ Bq) was leaked into the western Pacific Ocean, causing an abnormal increase in seawater ^{3}H concentrations[13,14]. It is noted that a direct release of wastewater with a high concentration of 1×10^6 Bq l^{-1} could pose a considerable threat to marine organisms[6], which, if assuming a concentration factor (CF) of 1.0 according to the International Atomic Energy Agency (IAEA), would lead to as high as 10^6 Bq kg^{-1} incorporation into marine organisms[15]. Although the release of ^{3}H from nuclear facilities has not caused substantially increased coastal water concentration owing to oceanic dilution, the total released activity remains notable. Furthermore, if there were a change in policy or an accident resulting in the release of untreated wastewater with levels significantly exceeding the recommended regulatory concentration limits, it would cause further serious concerns. Therefore, it becomes essential to understand the environmental consequences of abnormal ^{3}H output.

Most external radiation from this β^--emitting nuclide can be effectively shielded by the animal epidermis, as its radiation has a maximum

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range of approximately 5 mm in air and 0.6 μm in tissue[16]. In aquatic environments, however, ^{3}H predominantly exists as HTO, which exhibits high mobility and undergoes cyclic behaviour within the biosphere. Consequently, ^{3}H is inevitably taken up by organisms via respiration and the ingestion of water, leading to internal irradiation[16–18]. Current health risk assessments primarily focus on the accumulation of ^{3}H within individual organisms, largely neglecting the migration and associated risks of organically bound tritium (OBT) within food chains. This approach overlooks the important exposure pathways in which inorganic tritium is internalized and irreversibly sequestered in organic matter[19–27]. The potential human health risk posed by radionuclides via internal exposure can be assessed on the basis of seafood consumption and the radionuclide concentration in each food item. Therefore, an individual human exposure dose can be determined if three critical parameters can be identified.

First, the average annual seafood consumption per individual (kg yr^{-1}) has risen notably, with global per capita consumption of aquatic foods, excluding algae, exceeding 20 kg in the 2020s. Marine-captured fish account for the majority (~62.9%) of this consumption. Additionally, a portion of edible algae is artificially cultivated for use as food and industrial raw materials, representing 20.2% of the seafood incorporated into the diet of residents[28]. Therefore, both fish and algae should be considered when estimating the internal exposure dose for humans.

The second parameter is the radionuclide concentration in typical seafood (Bq kg^{-1}). The IAEA has suggested estimating radionuclide concentrations in seafood using a CF approach. This method transfers radionuclide concentrations from seawater and sediments to biota and has been widely applied[15]. The use of CF provides a pragmatic simplification for estimating the transport or inventory of radionuclides in biota, particularly given the difficulty in obtaining actual activity concentrations in biota tissues. However, employing a CF value of 1.0 for estimating ^{3}H biota concentration, assuming HTO uptake by only diffusion and permeation, is probably oversimplified and can be misleading[15]. The formation of OBT, which is more persistent than HTO in biota, would make the use of a 1.0 CF value for tritium assessments inappropriate. The incorporation of OBT-specific parameter values into the calculation of transport processes and dosimetry impacts has been proposed and implemented in various models[29–31]. The accumulation of tissue free water tritium (TFWT) generally exhibits a short biological half-life and is rapidly eliminated by aquatic organisms[6]. However, HTO can be assimilated by primary producers, such as phytoplankton, during photosynthesis, converting HTO into ^{3}H-carbohydrates. These ^{3}H-labelled biomolecules can subsequently be metabolized to form other biomolecules, including ^{3}H-protein and ^{3}H-lipid[32,33]. The OBT retained within producers becomes integrated into their biomass, and can be transferred to predators at higher trophic levels through the food chain[34]. Once ingested, OBT serves as a nutrient source, maintaining its persistence within the food web and increasing the risk of internal exposure. Therefore, determining tritium concentrations in seafood by accounting for its transfer through food chains is a fundamental aspect of assessing human exposure risks to ^{3}H.

The final parameter is the dose coefficient (DC) for individual radionuclides (Sv Bq^{-1}). The DC serves to convert the ingested radionuclide dose into an 'effective dose', a key metric within the framework established by the International Commission on Radiological Protection (ICRP)[35]. However, current ICRP recommended DCs for HTO or OBT are calculated using a standardized model, in which certain biokinetic parameters are not derived from human metabolic data[36]. Consequently, these extrapolated DC values may be systematically smaller than those calculated using parameters derived from human exposure. This discrepancy is particularly critical for OBT, which may exhibit prolonged biological retention in specific human organs[37]. Thus, a model is needed to modify DC values for a more accurate evaluation of human exposure dose.

Here, we integrate biogeochemical cycling and radiation exposure data from an established marine food chain model to assess how HTO impacts human exposure risks through its transformation into OBT along food chain. By systematically assessing tritium uptake and chemical transfer in biota, we highlight the key role of algae, which rapidly convert HTO into OBT, facilitating efficient trophic transfer to fish. Focusing on the critical health effects associated with OBT retention in fish tissues, we identify the incorporation of tritium into metabolic reactions within fish brain following assimilation into metabolites. Using CFs and biomagnification factors (BMFs) derived from quantitative studies, we have estimated the internal exposure dose to individuals (IEDI). These analyses were performed across various scenarios, including routine releases or accidental leaks, with different environmental conditions of marine tritium exposure. Our integration of radiobiology, chemical analysis and ecological risk assessment emphasizes the importance of adopting comprehensive strategies to assess and mitigate tritium exposure risks, thereby supporting the development of sustainable nuclear energy.

Results

HTO organification by algae

To explore the organification efficiency of HTO by producers at various growth stages, *Dunaliella salina*, in either the saturated or logarithmic growth phase, was initially exposed to artificial seawater containing 10,000 Bq l^{-1} of HTO for 14 days (Supplementary Fig. 1). The average specific radioactivity of OBT in the growing phase algae was measured at 4.9 Bq mg^{-1} (Fig. 1a), approximately 4.5 times higher than that observed in stationary phase algae (1.1 Bq mg^{-1}), consistent with previous findings[32]. The marked increase in OBT activity during the growth phase could be attributed to enhanced photosynthetic activity. During this phase, algae convert water and carbon dioxide into organic matter more rapidly[32], resulting in greater incorporation of tritium into organic compounds.

We subsequently monitored the kinetics of ^{3}H incorporation in *D. salina* during HTO exposure in the growth phase. Given the rapid rate of water exchange in the organism, the specific activity of HTO was equilibrated to the surrounding water environment (10 Bq ml^{-1}) within 1 day and remained essentially stable (~10 Bq g^{-1}; Supplementary Fig. 2). The OBT was observed accumulating at a steady rate (~6.21 Bq g^{-1} d^{-1}) over the first 10 days, followed by a rapid increase for 2 days (~24.85 Bq g^{-1} d^{-1}), and then slowed during the remaining exposure period (~8.18 Bq g^{-1} d^{-1}). The initial slow accumulation during the initial phase was because of the limited number of algae during early incubation. The subsequent fourfold increase in OBT accumulation corresponded to the logarithmic growth phase, characterized by both a higher number of algal cells and an enhanced photosynthetic activity per cell. As the algal population reached high concentrations, spatial constraints led to a plateau in algal proliferation, and the light-shielding effect of the dense algae diminished the photosynthetic rates, resulting in a deceleration of OBT accumulation.

Furthermore, the OBT/HTO ratios calculated for algae over the tested period of 14–28 days ranged from 2.84 to 3.19 (Supplementary Fig. 3), aligning well with previous studies[8,25,38]. For instance, Kim et al. reported an OBT activity concentration in algae of 99.6 ± 3.2 Bq l^{-1}, while the TFWT activity concentration was measured as 33.6 ± 1.1 Bq l^{-1}, corresponding to an OBT/TFWT ratio in their study of approximately 2.96 (ref. 25). In general, OBT activity concentrations in algae exceed those in the TFWT and the surrounding water. This phenomenon can be attributed to the fact that OBT formed via biosynthetic processes (for example, photosynthesis) in algae requires more time to be depurated[38]. We therefore investigated the depuration of OBT from *D. salina* by transferring them into a tritium-free medium. OBT was scarcely excreted over a 7-day depuration period (Fig. 1c), as confirmed by measuring the radioactivity in the medium (Supplementary Fig. 4). Our results were consistent with a previous report that accumulated

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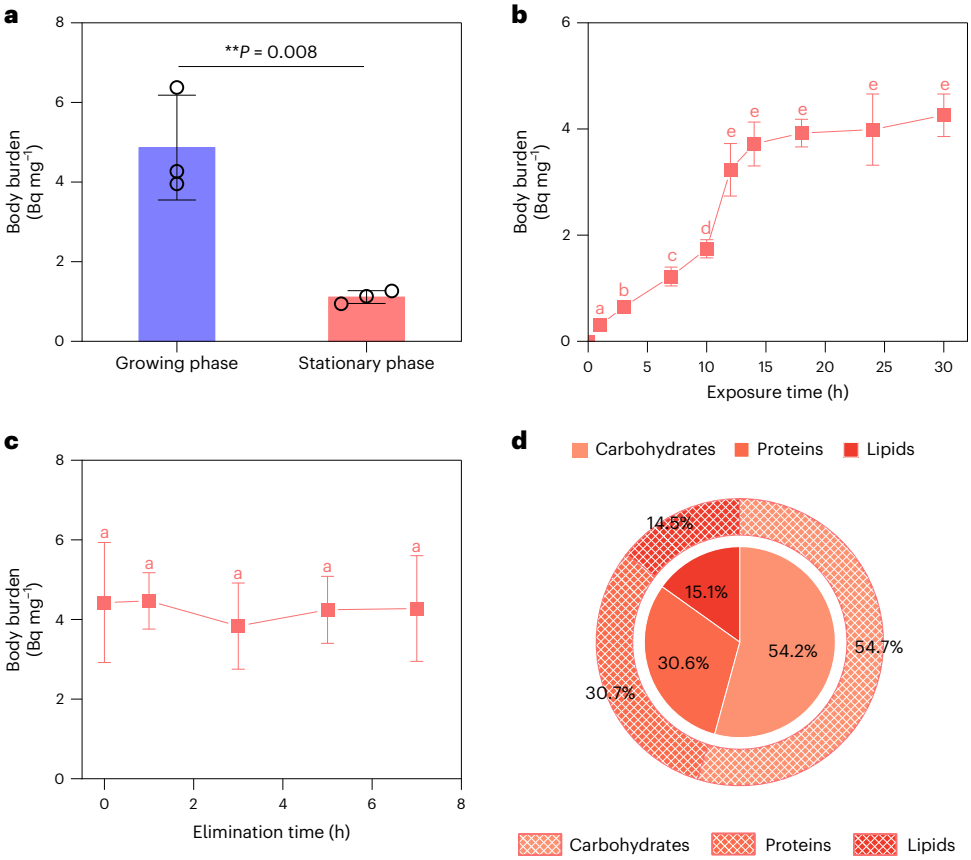

Fig. 1 | HTO organification by algae. a, The radioactive concentrations (body burden) of OBT in *D. salina* dry mass at various growth stages after exposure to 10,000 Bq l^{-1} HTO. The data are shown as mean values ± s.d. ($n = 3$). Student's *t*-test (two-sided) was used for the analysis of comparisons between two data points, with $P < 0.05$ for significant differences. **b**, The radioactive concentrations (body burden) of OBT in *D. salina* dry mass in the growing phase during a period of 30-day exposure to 10,000 Bq l^{-1} HTO. The data are shown as mean values ± s.d. ($n = 3$). **c**, The OBT body burden of algae during a period of 7-day elimination in tritium-free medium. The data are shown as mean values ± s.d. ($n = 3$). **d**, The intrinsic percentage of carbohydrates, proteins and lipids in *D. salina* biomass with HTO exposure (inner circle) and the percentage of OBT in carbohydrates, proteins and lipids in *D. salina* via HTO exposure (outer circle). The data are shown as mean values ($n = 3$). Accumulation and elimination results data were analysed by one-way analysis of variance (ANOVA). Data points with the same letter suggest no significant difference ($P > 0.05$).

tritiated glycine was hardly depurated by marine mussels over its test period[7]. Therefore, it was speculated that OBT, involving stronger carbon–tritium bonds, exhibited a longer biological half-life than TFWT. Owing to the extremely low elimination rate of OBT by algae, a significantly higher CF of 377.90 was found in the primary producer, compared with the value of 1.0 suggested by the IAEA[15].

To elucidate the components contributing to OBT in algae, we analysed the relationship between the forms of OBT and the organic composition of *D. salina* (Fig. 1d). The distribution of OBT among carbohydrates (55.8%), proteins (30.7%) and lipids (14.5%) closely matched the intrinsic proportions of carbohydrates (54.3%), proteins (30.6%) and lipids (15.1%) within *D. salina*. This suggests that the distribution of OBT in different chemical forms derived from HTO is dependent on the intrinsic organic composition of the producer organism. Edible algae serve as a direct food source for humans, bypassing other consumers, as seaweeds constitute approximately 20.2% of seafood consumed in the daily diet in certain countries[30]. Given this substantial dietary contribution, OBT incorporated into algae can contribute significantly to the internal human exposure dose. Therefore, it is essential to consider this uptake pathway in the estimation of human exposure doses.

Trophic transfer of ^{3}H in the food chain

To evaluate the contribution of trophic transfer of tritium within the marine ecosystem, brine shrimp (*Artemia salina*) and Japanese medaka (*Oryzias latipes*) were selected as representative organisms occupying higher trophic levels to establish a simplified marine food chain. The uptake of tritium by brine shrimp via direct uptake of HTO was compared with the trophic transfer of OBT. The tritium body burden observed in brine shrimp (Fig. 2a) indicated that ingestion of OBT constituted a substantially more efficient pathway for ^{3}H accumulation than direct exposure to HTO. Specifically, at 24, 48, 72 and 96 h, OBT trophic transfer resulted in body burdens 1.6, 3.8, 9.8 and 12.7 times higher, respectively, than those from direct HTO exposure. These findings indicated that the accumulation of OBT was positively correlated with the intake of tritiated food, whereas equilibrium was not achieved even after 4 days of continuous exposure. Additionally, the OBT transfer efficiency gradually increased with higher dietary concentrations (Supplementary Fig. 5). In contrast, brine shrimp directly exposed to HTO with equivalent activities did not exhibit an appreciable correlation between exposure level and tritium body burden (Supplementary Fig. 6). Furthermore, we explored the depuration process of OBT by brine shrimp. As shown in Fig. 2b, the radioactivity of OBT gradually decreased over the testing period, presenting a pattern distinct from that observed in *D. salina*. This difference could be attributed to the excretion of undigested tritiated food from the intestinal tract of brine shrimp. Notably, the calculated BMF (defined as the ratio of ^{3}H concentration in predator to prey)[39] was much lower than 1.0 for the tested food chain, indicating an absence of biomagnification during trophic transfer, consistent with previous studies on the fate of tritium in aquatic biota[1,16,32,34,40]. Nevertheless, our results indicated

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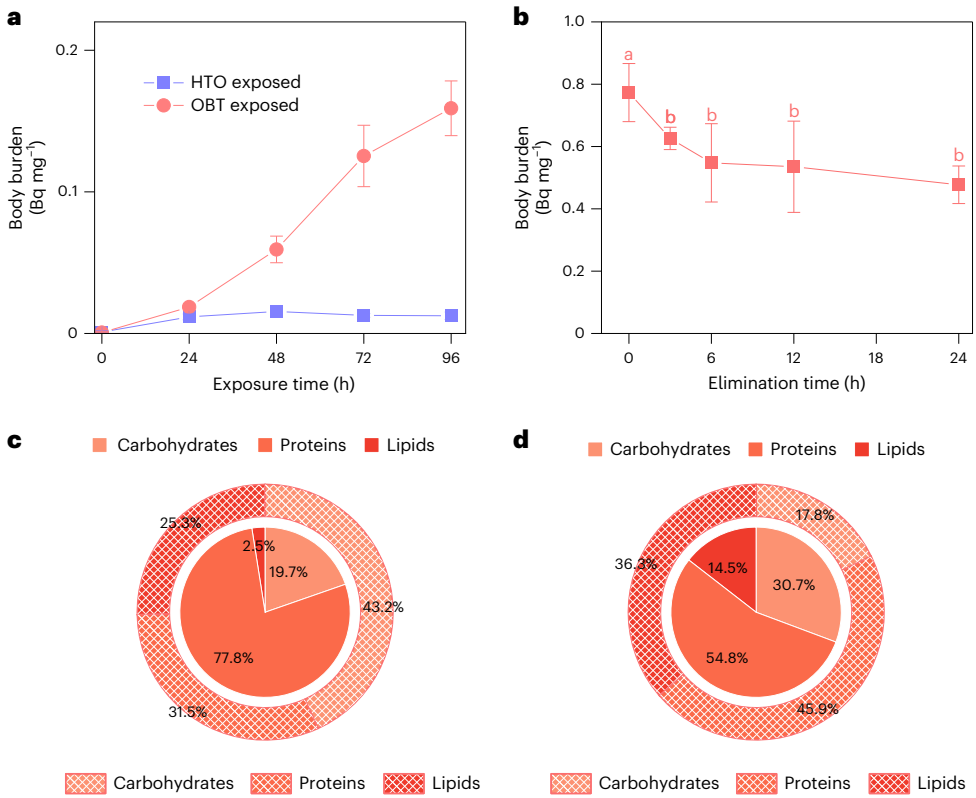

Fig. 2 | The enrichment and transfer of ^{3}H through exposure of HTO or ^{3}H-algae (tritiated *D. salina*) to brine shrimp. a, The activity concentrations (body burden) of OBT in brine shrimp dry mass during direct exposure to HTO and trophic transfer from 50 mg of ^{3}H-algae with specific activity of 4,000 Bq g^{-1}. The data are shown as mean values ± s.d. ($n = 3$). **b**, The OBT body burden of brine shrimp during 24 h of elimination in tritium-free artificial seawater. The data are shown as mean values ± s.d. ($n = 3$). **c**, The intrinsic percentage of carbohydrates, proteins and lipids in brine shrimp biomass (inner circle)[38] and the percentage of OBT in carbohydrates, proteins and lipids of brine shrimp via HTO exposure (outer circle). The data are shown as mean values ($n = 3$). **d**, The percentage of OBT in carbohydrates, proteins and lipids of *D. salina* (inner circle) or brine shrimp (outer circle) fed on it. The data are shown as mean values ($n = 3$). Accumulation and elimination results data were analysed by one-way ANOVA. Data points with the same letter suggest no significant difference ($P > 0.05$).

that organisms feeding on algae containing significant levels of tritium would accumulate substantial OBT body burdens, highlighting the important role of algae in introducing tritium into marine ecosystems.

We further analysed the relationship between the forms of OBT and organic composition in brine shrimp, either exposed directly to HTO or fed on algae containing OBT (Fig. 2c). Unlike the transformation dynamics observed in algae, the distribution of OBT in brine shrimp was markedly different from their intrinsic organic composition after a 4-day exposure to HTO. However, as high-protein and low-fat zooplankton, brine shrimp exposed to HTO appeared to develop OBT in relatively balanced forms compared with their counterparts fed with OBT-incorporated algae. Specifically, brine shrimp exposed directly to HTO contained carbohydrates (19.7%), proteins (77.8%) and lipids (2.5%), while the corresponding radioactivity ratios were 43.2%, 31.5% and 25.3%, respectively[41]. In contrast, after consuming OBT-containing algae, the distribution of radioactivity in carbohydrates (17.8%), proteins (45.9%) and lipids (36.3%) diverged substantially from the composition in algae, measured in carbohydrates, proteins and lipids at 54.8%, 30.7% and 14.5%, respectively. These findings suggest that once HTO is converted to OBT by the primary producer, its subsequent metabolic processing within the consumer (brine shrimp) can reorganize them following different pathways, determined by the organism's specific biochemical processes.

For medaka directly exposed to HTO, ^{3}H uptake reached 0.06 Bq mg^{-1} on the first day, followed by minor fluctuations (Fig. 3a). Despite this initial uptake, no notable accumulation of ^{3}H was observed in the organs (Fig. 3b), indicating minimal retention of tritium within the fish. However, when fed with ^{3}H-containing brine shrimp, the OBT content in medaka increased at a rate of 0.16 Bq mg^{-1} d^{-1}, reaching 12.6 times specific activity in comparison with that of the HTO exposure group at 96 h. This clearly demonstrates the efficient transfer of ^{3}H along the food chain that led to notable OBT accumulation within the fish. A previous study similarly showed that OBT concentrations in fish ingesting OBT-containing food increased more rapidly than those resulting from an equivalent intake of tritium in the form of HTO[7]. Notably, as observed in the food chain of algae to shrimp, biomagnification was not evident in the current trophic transfer, with the calculated BMF remaining below 1.0 (Supplementary Table 1). Given the low uptake efficiency via direct HTO exposure (Fig. 3a) and the absence of biomagnification via trophic transfer, bioaccumulation of tritium in medaka is unlikely, consistent with findings from previous studies[16].

To illustrate the retention of ^{3}H in the tested fish, we monitored the elimination of ^{3}H in medaka that had accumulated ^{3}H via food chain exposure. The body burden curve of ^{3}H (Fig. 3c) showed an initial rapid decrease within the first 2 days, followed by a plateau phase. These results were consistent with a previous study, which found that fishes fed with a tritiated diet appeared to have minor decreases in activity concentration after 1 day of depuration[7]. Compared with the initial stage, 61.5% of ^{3}H remained at 96 h, with no observable trend towards further elimination. In addition, medaka were dissected before and after the 4-day depuration period to examine the radioactivity change across various tissues (Fig. 3d). Following 4 days of dietary exposure, total activities were 6.1, 11.5, 14.0, 0.42 and 0.39 Bq in brain, body, gut, liver and gill, respectively. After an additional 4 days of depuration, the ^{3}H content in the roe (8.5 Bq) and gut (4.5 Bq) decreased by 26.6% and 68.0%, respectively, with no detectable radioactivity in the gills.

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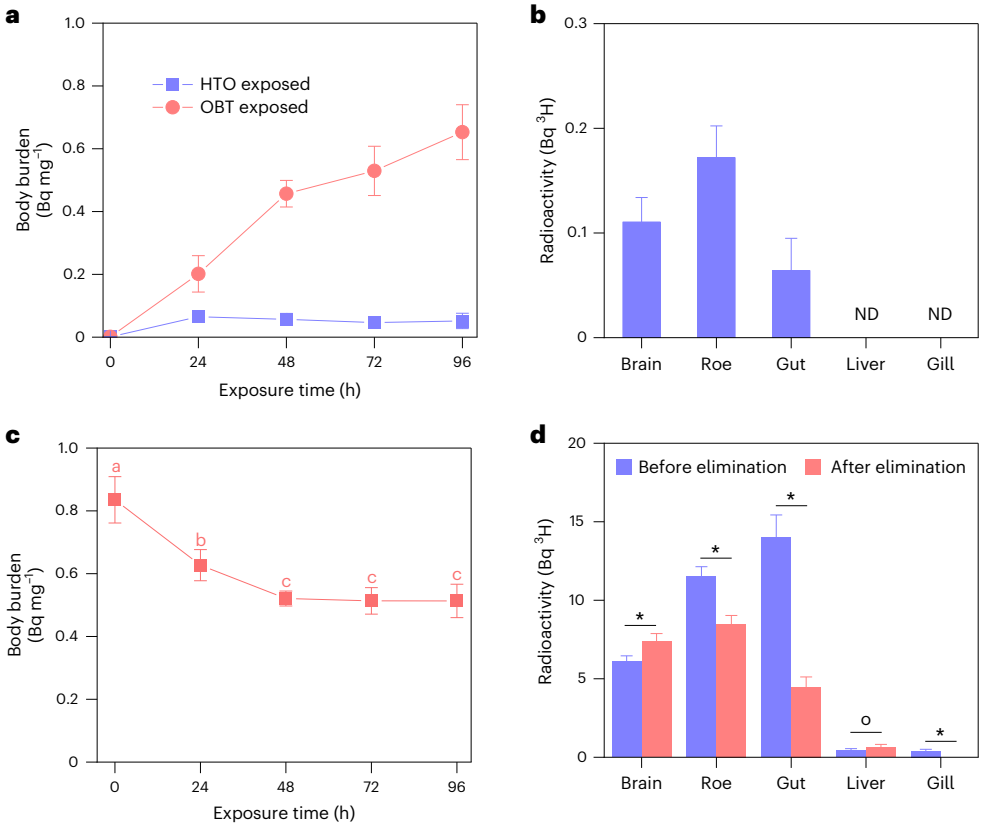

Fig. 3 | The enrichment and transformation of ^{3}H through exposure of HTO or ^{3}H-brine shrimp (tritiated brine shrimp) to medaka. a, The activity concentrations of OBT in medaka during direct exposure to HTO and trophic transfer from ^{3}H-brine shrimp. The data are shown as mean values ± s.d. ($n = 3$). **b**, The biodistribution of OBT in medaka after direct exposure to HTO for 96 h. The data are shown as mean values ± s.d. ($n = 3$). **c**, The elimination of ^{3}H by medaka. The data are shown as mean values ± s.d. ($n = 3$). Data points with the same letter suggest no significant difference ($P > 0.05$). **d**, The biodistribution of OBT in medaka after trophic transfer from ^{3}H-brine shrimp for 96 h and subsequent elimination for 96 h. The data are shown as mean values ± s.d. ($n = 3$). Statistically significant differences were performed using Student's *t*-tests (two-sided), and asterisks (*) indicate statistically significant differences ($P < 0.05$) or circles (o) indicate not statistically significant differences ($P > 0.05$) . Accumulation and elimination results data were analysed by one-way ANOVA. Data points with the same letter suggest no significant difference ($P > 0.05$). ND, not detectable.

However, there was no statistically significant difference in ^{3}H content in the liver before and after the excretion, remaining at approximately 0.6 Bq. It is noteworthy that the radioactivity in the brain (7.4 Bq) did not decrease but increased by 21.0%. The sustained accumulation of ^{3}H in the brain may be attributed to the continuous transfer of OBT through metabolic pathways for the construction of newborn neurons. In this form, tritium could persist over extended periods, similar to the integration of ^{14}C in synthesized genomic DNA[42,43].

Blending of ^{3}H in metabolism

The integration of radioactive ^{3}H into the DNA of fish has been shown to cause genetic damage[24]. To investigate the potential physiological effects, we monitored behavioural changes in medaka by feeding them ^{3}H-accumulated brine shrimp (trophic transfer) or directly culturing them in HTO (direct exposure) for 4 days. Our observations showed a significant reduction in mobility and cognitive functions amongst fish exposed to OBT (Supplementary Text and Supplementary Figs. 7 and 8), suggesting that OBT incorporation into the brain may induce behavioural abnormalities and potentially lead to neurodegeneration in medaka. Given that such behavioural abnormalities may arise from imbalances among critical biomolecules involved in metabolism[44], we examined the incorporation of ^{3}H into amino acids, which play a vital role in metabolism, using ultra-high performance liquid chromatography with quadrupole time-of-flight mass spectrometry. After extracting and filtering MS1 and MS2 peaks from the raw mass data, amino acid isotopologues in ^{3}H-labelled brain samples were identified by comparison with standard amino acid samples, according to the following criteria: (1) mass error <5 ppm, (2) differences in isotope abundance ratios <10% and (3) at least two matched daughter ion fragments in the MS/MS map. The extent and pattern of labelling for each amino acid were determined by searching all potential isotopologues using the accurate mass maps available in the MassBank database. In total, eight ^{3}H-labelled amino acids with 14 fractions of isotopologues were confirmed, as listed in Fig. 4. Our results confirmed the bioavailability of OBT and its incorporation into organisms through the food chain, highlighting the participation of ^{3}H in metabolism and lifecycle processes through hydrogen substitution in amino acids.

The involvement of ^{3}H in metabolism warrants specific concern, as the replacement of hydrogen (^{1}H) by a heavier tritium (^{3}H) isotope can disrupt the binding affinities and reaction rates of critical biomolecules. Isotopically substituted molecules theoretically exhibit altered reaction rates owing to changes in bond energies and vibrational modes, known as isotope effects[45]. In the present study, ^{3}H-proline was identified as being retained in medaka brain samples (Fig. 4). The catalytic transformation of proline determines the production of pyrroline-5-carboxylate, a central component of the tricarboxylic acid cycle, the urea cycle and the proline cycle[46]. We thus conducted the frequency calculations after geometry optimizations of ^{3}H-proline (^{3}H substitution at position 1 and 5, where dehydrogenation occurs) or ^{1}H-proline to calculate theoretical kinetic isotope effects (^{1}H/^{3}H; KIEs) using the ISOEFF package[47]. The calculated KIEs at the atomic positions of interest were 2.065 (1-^{3}H) or 3.851 (5-^{3}H)

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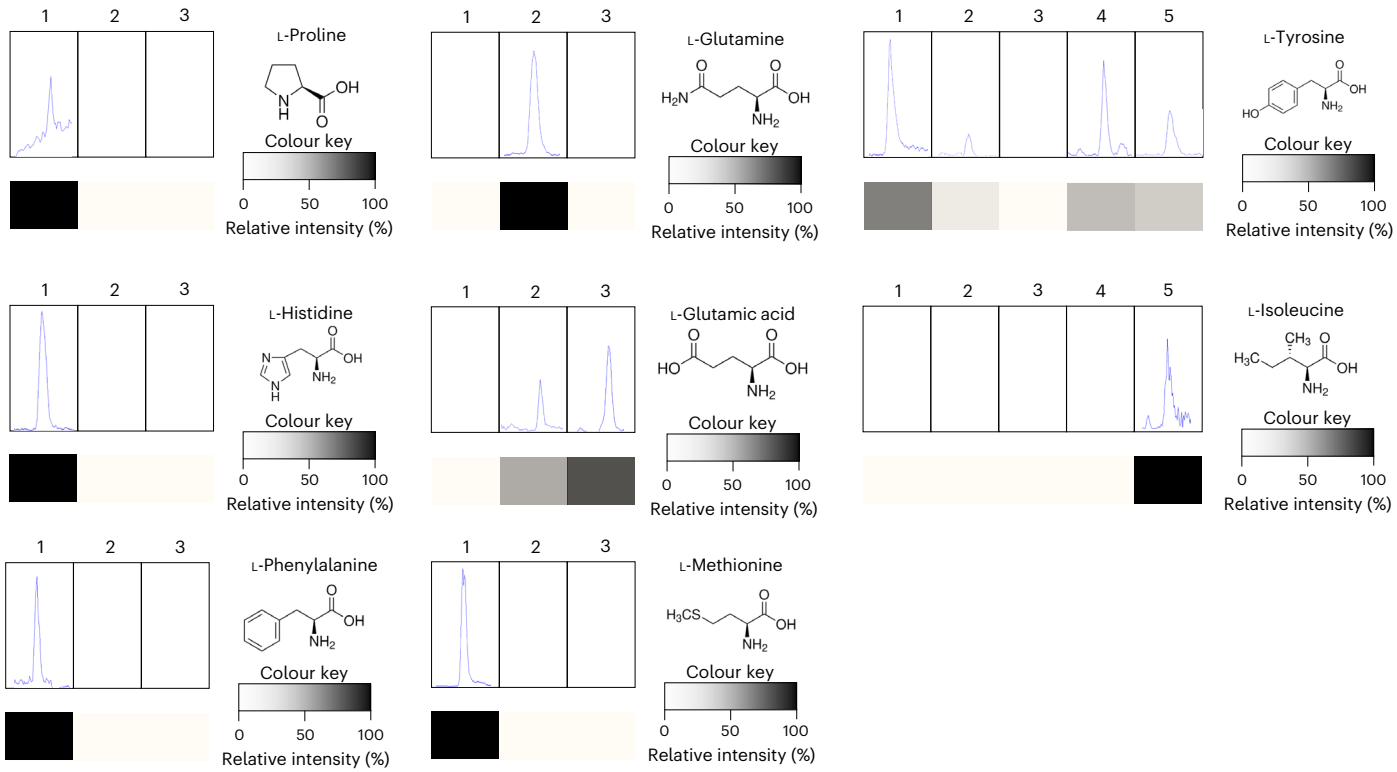

Fig. 4 | Blending of ^{3}H in amino acids. Eight ^{3}H-labelled amino acids with 14 fractions of isotopologues in the brain of medaka that were exposed via feeding of ^{3}H-containing brine shrimp. Isotopologues were identified by ultra-high performance liquid chromatography with quadrupole time-of-flight mass spectrometry by comparing with standard amino acid samples.

(Supplementary Fig. 9), indicating that ^{3}H substitution at a critical site reduced the reaction rate by 51.6% and 74.0%, respectively. Substantial KIEs have also been observed in reactions involving deuterium substituted molecular. For instance, a KIE of 15.3 was reported in the dehydration of 1,2-propanediol for the deuterium substitution at the critical reaction site[48]. We further evaluated the disturbance of H isotope substitution on proline transformation in vitro by measuring the reaction kinetic mediated by proline dehydrogenase (PRODH) and flavin adenine dinucleotide (FAD). Under the same concentration, the reaction kinetics of ^{2}H-proline were slightly slower compared with those of ^{1}H-proline (Supplementary Fig. 10). In the present study, isotope substitution was observed in numerous amino acids involved in critical metabolic pathways, raising concerns on the potential isotope effects that could impair the function of critical proteins following the incorporation of tritium via trophic transfer.

Human exposure evaluation

Given the critical role of internal ^{3}H exposure via trophic transfer for assessing human exposure doses, we adopted a simplified approach to estimate the concentrations of radionuclides in seafood, using the CF and BMF. The CF is defined as the ratio of radionuclide concentrations in biota to those in seawater, whereas the BMF describes the transfer efficiency of radionuclide from prey to predator, expressed as the ratio of specific activity between them. As a typical phytoplankton, the uptake of tritium by algae from sediment is supposed to be minimal. Accordingly, the CF for ^{3}H was derived from its accumulation kinetics in aquatic environments (Fig. 1b), yielding 377.90 kg l^{-1} for algae and 5.17 kg l^{-1} for medaka (Fig. 3a), both substantially higher than the reference value of 1.0 suggested by the IAEA. Using the calculated CF values, ^{3}H concentration and the proportions of HTO and OBT in aquatic organisms were determined. For fish, BMF values were derived from the trophic transfer results from this study. The tested scenarios were classified into five categories: (S1) ^{3}H background in coastal zones and oceans (~0.1 Bq l^{-1})[49], (S2) regular discharges from nuclear reprocessing plants including La Hague in France (~5 Bq l^{-1})[50] and Sellafield in the UK (~5 Bq l^{-1})[51], (S3) a river receiving contaminated effluent from nuclear weapons material production facilities at Fourmile Branch in the USA, with an average concentration of 1,790 Bq l^{-1} (ref. 49) and (S4) discharge of unexpected tritium contaminated water from Fukushima, based on monitored concentrations reported at the Treated Water Portal Site[52]. Two typical concentrations were evaluated: the planned discharge concentration of 1,500 Bq l^{-1} and the maximum coastal seawater concentration of ~29 Bq l^{-1} within 3 km from the outlet. According to a simulation result of tritium-contaminated water, the dispersion of HTO was relatively limited in a short period[53]. Given the rapid uptake of HTO by primary producers, algae growing near effluent outfalls are at risk of exposure to high concentrations close to that of the effluent. Given the presence of marine producers such as algae ubiquitously growing in the sea, the dispersal of phytoplankton blooms formed in the vicinity of tritium release sites can lead to contamination of filter-feeding organisms even in areas where the water is not directly contaminated. Thus, as a conservative approach, dilution and dispersion of HTO were not considered for dose calculations; instead, the discharge concentrations used were seawater concentrations for the prediction of the IEDI. This conservative method provides an upper-bound estimate of potential radiological risks, even in scenarios where coastal areas are closed to fishing.

The IEDI from ingestion of seafoods (algae and fish), on the basis of modelled seawater concentrations under the tested scenarios, is presented in Table 1. Under the Fukushima discharge scenario (S4), the currently monitored concentration of 29 Bq l^{-1} within a 3 km coastal area from the outflow was used as the input for the model. On the basis of this concentration, the IEDI was calculated as 1.02 μSv yr^{-1}, indicating that this unexpected additional tritium input contributes more to the annual human radiation dose than regular discharges from nuclear reprocessing facilities at La Hague (France) and Sellafield

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Table 1 | The IEDI from annual seafood consumption

Radionuclide		^{3}H	
		IAEA method	Current work
CF from water (l kg^{-1})		1.00[a]	377.90, algae; 5.17, medaka
BMF from algae to brine shrimp		NA	0.041
BMF from brine shrimp to medaka		NA	0.091
CF from sediment (m^2 kg^{-1})		0	
Decay half-life (years)		12.3	
K_d (l kg^{-1})		1[b]	
Annual fish consumption per capita (kg)		20.87[c]	
Annual algae consumption per capita (kg)		4.174	
Ingestion DC (nSv Bq^{-1})		0.042[d]	0.35[e]
Scenario	Discharge sites	Exposure dose (μSv yr^{-1})[f]	
S1	Coastal background	0.000206	0.0035
S2	La Hague, France	0.0103	0.176
	Sellafield, UK	0.0103	0.176
S3	Fourmile Branch, USA	3.7	63
S4	Fukushima, planned	3.1	53
	Fukushima, monitored seawater	0.060	1.02

[a]CF from ref. 15 [b]Partition coefficient from ref. 15 [c]Seafood consumption from ref. 28 [d]DC from ref. 35 [e]DC from ref. 37 [f]Scenarios applied to prediction: (S1) ^{3}H background concentration in the ocean (~0.1 Bq l^{-1}); (S2) areas near nuclear power plants that regularly discharge ^{3}H, including La Hague in France (~5 Bq l^{-1}) and Sellafield in the UK (~5 Bq l^{-1}); (S3) an area at Fourmile Branch in the USA that received contaminated effluent from nuclear weapon material production facilities, an average concentration of 1,790 Bq l^{-1}; and (S4) discharge of unexpected extra tritium-containing Fukushima contaminated water: the planned discharge concentration of 1,500 Bq l^{-1} and maximum coastal seawater concentration of ~29 Bq l^{-1} within 3 km of the outlet.

(UK), which were calculated as 0.176 μSv yr^{-1}. Furthermore, this IEDI exceeds the estimated dose of 0.072 μSv yr^{-1} from seafood consumption in coastal waters of Fukushima Prefecture before the 2011 nuclear incident, which did not account for artificial radionuclides introduced into the ocean[54]. Compared with previously reported models estimating the tritium-induced extra human exposure dose (0.0014–0.037 μSv yr^{-1}) (refs. 55–57), the value from our estimation is approximately two orders of magnitude higher. This significant increase arises from the incorporation of rigorous experimental data on bioaccumulation and the transfer of radioactive substances through the food chain. The inclusion of newly determined CF and BMF values (Table 1) provides a more accurate representation of potential health risks associated with the discharge of radioactive wastewater into the ocean. Under the planned discharge concentration of 1500 Bq l^{-1}, the modelled IEDI was calculated as 0.053 mSv yr^{-1}, exceeding the dose constraint of 0.05 mSv yr^{-1} established by Japan's Nuclear Regulation Authority for radiological environmental impact assessments[58]. Current IEDI estimation thus provides a more rigorous risk assessment of potential exposure to the planned discharge or accidental tritium release.

Discussion

This study demonstrates that the organification of HTO to OBT by algae is a key factor in increasing the ^{3}H concentration in fish through trophic transfer. Our results show that algae rapidly incorporate HTO and convert it into OBT, integrating it into biomolecules corresponding to their intrinsic organic components (carbohydrates, proteins and lipids). Comparisons of ^{3}H accumulation in fish via direct exposure to HTO or trophic transfer from OBT indicate that OBT is more efficiently transferred through the food chain and persists longer in predators. Specifically, the retention of OBT in the fish brain can integrate with biomolecules such as ^{3}H-bound amino acids, causing potential disruption to reactions in critical metabolic pathways.

When it comes to the health risk assessment of ^{3}H to humans, marine producers play a pivotal role since they contribute over 70% to the IEDI and facilitate the conversion of HTO into bioavailable OBT, which is retained in seafood as the major component of per capita consumption. Using parameters derived from the current study, the estimated IEDI from seafood ingestion was 1.02 μSv yr^{-1} under monitored seawater concentrations following Fukushima wastewater release scenarios. Although this dose is far below regulatory limits, it represents an additional source of exposure for both marine organisms and humans. While dilution inevitably occurs in the ocean, it does not justify unlimited or unchecked releases of contaminated water, as each discharge contributes to the long-term accumulation of radiation in the marine environment, posing potential ecological and human health risks through bioaccumulation in the food chain. On the basis of the results in this study, the potential ecosystem and human health risks of radionuclide discharge may be underestimated in relation to bioaccumulation and trophic transfer, particularly for scenarios where incidental high discharge concentrations may happen. It highlights the necessity to develop effective technologies to remove tritium from wastewater before its release, thereby mitigating the risk of bioaccumulation and protecting both ecosystem integrity and public health. Our findings underscore the importance of monitoring and further evaluation of radioactive wastewater discharges to ensure sustainable nuclear energy management.

Methods

Test organisms

HTO was purchased from China Isotope & Radiation Corporation. *D. salina* was obtained from Shanghai Guangyu Biological Technology Co., Ltd. The cysts of brine shrimp were purchased from the Institute of Marine Sciences, Shandong, and stored at 4 °C before experimentation. Medakas (*Oryzias latipes*) were obtained from Shanghai Feixi Biotechnology Co., Ltd. All animal experiments were conducted in strict accordance with the guidelines of the Chinese Council for Animal Care, approved by the Animal Care Committee of the Laboratory Animals in Nanjing University.

Tritium exposure of *D. salina*

Marine phytoplankton *D. salina* was cultured in artificial seawater with a salinity of 35‰ in an incubator, maintained at 22 ± 1 °C under a 16:8 h light:dark photoperiod. Algal cultures were manually shaken three times daily, and their positions in the incubator were changed randomly daily. The algal cells were harvested during the exponential growth stage and centrifuged at 100 × *g* for 5 min at 4 °C, after which the pellet was resuspended in fresh artificial seawater. The optical density of the stock suspension was measured using an ultraviolet–visible spectrophotometer (Cary 50, Varian) at the maximum absorption wavelength of 678 nm. Cell density was determined using a Neubauer-improved counting chamber (Marienfeld, Germany), with three replicate samples counted at least twice per replicate.

Incorporation of ^{3}H into *D. salina* was carried out in 250-ml sterile culture containers. Given that the algal growth phase may affect metabolism, three treatments were established: control, stationary-culture exposure and exponential-culture exposure. The stationary-culture treatment consisted of 100 ml of *D. salina* stock culture that had reached growth plateau for 2 weeks, spiked with HTO to an activity concentration of 10,000 Bq l^{-1}. The exponential-culture treatment was prepared by inoculating 10 ml of stock culture into 90 ml of fresh medium, followed by the addition of HTO to 10,000 Bq l^{-1}. The selected activity concentration, although higher than typical environmental levels (except near release points), was necessary to ensure measurable activities of OBT in the phytoplankton. Control treatments were identical to the exponential-culture exposure except for HTO addition.

Three replicates were conducted for each condition. After 14 days of exposure, exponential-culture treatments reached a stationary phase. For HTO kinetics, *D. salina* from exponential-culture treatments were sampled multiple times over a 2-week period.

At sampling times (1, 3, 7, 10, 12 and 14 days), approximately 30 ml of algae suspension from each replicate was collected and filtered onto pre-weighed polycarbonate membranes with a pore size of 3 µm (Nuclepore Track-Etched Membranes, Whatman). The algae cells were rinsed with 10 ml of distilled water. Each filter was then freeze-dried, weighed and placed into 20 ml of alkaline scintillation liquid (Oxysolve C-400, Zinsser Analytic). Samples were left until the filter papers completely dissolved and were dark acclimatized for at least 2 h. Our preliminary data confirmed that these filter papers dissolve effectively in the alkaline scintillation cocktail (Supplementary Fig. 11). The radioactivity of ^{3}H in algae cells was quantified using a liquid scintillation counter (LSC; Tri-Carb 5110 TR; PerkinElmer, USA)[59]. Notably, our preliminary tests indicated that the multiple sample-handling steps, including filtration, centrifugation and lyophilization did not significantly affect the accurate determination of HTO and OBT (Supplementary Fig. 12).

Extraction of lipid, protein and carbohydrate from algae cells

After 14 days of exposure, algae cells were collected and washed at least three times with fresh medium. Approximately 0.4 g of algae cells was placed in a 10 ml tube, and 6 ml of methanol was added[60]. The mixture was heated at 70 °C for 1 h and then sonicated for an additional 30 min. Subsequently, the mixture was centrifuged at 280 × *g* for 2 min, and the supernatant and sediment were collected, separately. The sediment was washed twice with methanol. All supernatants were combined, and the methanol was evaporated to isolate lipids, which were then quantified using a LSC. The sediment was dissolved in 5 ml of 3% KOH solution and heated at 80 °C for 1 h, followed by 30 min of sonication. To extract proteins, 2 ml of a chloroform: *n*-butyl alcohol solution (*v*:*v* of 4:1) was added, shaken for 10 min and centrifuged. This extraction process was repeated at least five times to ensure complete protein isolation, and the obtained proteins were quantified with a LSC. For carbohydrate separation, 10 ml of methanol was added to the supernatant, the sediment was collected and the carbohydrates were determined by a LSC.

Tritium exposure to brine shrimp

Before hatching, the cysts of brine shrimp were disinfected and pretreated by soaking in 300 mg l^{-1} $KMnO_4$ solution for approximately 5 min, followed by thorough rinsing with artificial seawater until the effluent was colourless. Subsequently, 0.2 g of cysts were placed in a 20 g l^{-1} NaCl solution and incubated at 26–28 °C for 24 h under continuous light illumination, with continuous oxygenation to enhance hatching rates. The growth and behaviour of brine shrimp were monitored microscopically, and no abnormal behaviour or mortality was observed during culture.

Brine shrimps were exposed to tritium through two pathways: direct HTO exposure or feeding with tritium-fed *D. salina* (^{3}H-algae). The ^{3}H-algae, with a radioactivity of 4,000 Bq g^{-1} were washed as described above and diluted in fresh artificial seawater to final concentrations of 20, 100 and 200 mg l^{-1}. In total, 10,000 freshly hatched brine shrimp were introduced into each culture dish containing 500 ml of ^{3}H-algae at the respective concentrations. For direct HTO exposure, 10,000 freshly hatched brine shrimp were added to 500 ml of artificial seawater containing HTO at an equivalent activity and were fed equal amounts of ^{3}H-free *D. salina*. A control setup containing only freshly hatched brine shrimp was maintained to account for background measurements.

At sampling times (24, 48, 72 and 96 h), all brine shrimp were collected and pipetted vigorously, with this step repeated three times to ensure thorough handling. The brine shrimp from each container were then transferred to scintillation vials containing 3 ml of Gold Star cocktail, ultrasonicated for 30 min, allowed to sit for at least 24 h and subsequently analysed using a LSC. Three replicate experiments were performed for each treatment condition. Additionally, carbohydrates, lipids and proteins from brine shrimp were extracted and quantified using a LSC following the procedures described above.

^{3}H exposure of medaka

Adult marine medakas with body lengths of 31.86 ± 1.4 mm were cultured in filtered artificial seawater with a salinity of ~30 ‰ under a 14:10 h light:dark cycle at 27 ± 1 °C and fed newly hatched brine shrimp at approximately 2% of their body weight once daily. Before uptake experiments, medakas were acclimatized in the laboratory for at least 1 week, with no abnormal behaviour or mortality observed during the uptake and depuration experiments.

Each medaka was placed in a beaker containing 200 ml of test medium. Tritiated brine shrimp, obtained by feeding on ^{3}H-algae, were used for trophic transfer exposure. Medakas were placed in tritium-free medium and fed 25 mg of 20,000 Bq g^{-1} ^{3}H-brine shrimp once daily. For direct HTO exposure, medakas were exposed to medium containing 10,000 Bq l^{-1} HTO and fed 25 mg of ^{3}H-free brine shrimp daily. All brine shrimp were fully consumed every day. To prevent HTO evaporation, aeration was not applied during ^{3}H exposure, and preliminary data confirmed that medaka growth was not significantly affected by aeration. At predetermined time points (24, 48, 72 and 96 h), medakas were euthanized using 3-aminobenzoic acid ethyl ester methanesulfonate (MS-222, 0.015%). Body length was measured, epidermis was peeled and fins were removed to eliminate the tritium adhering to external surfaces. The medakas were then freeze-dried, weighed and digested for 40 min using a mixture of 60% HNO_3 and 30% H_2O_2 (*v*:*v* of 4:1) under an ice–water bath. The digest was neutralized using 5 mol l^{-1} NaOH, and 10 ml of alkaline scintillation cocktail (Zinsser Analytic, Germany) was added. Radioactivity was subsequently determined using a LSC, with background activity from control medakas subtracted from uptake measurements.

Elimination experiments were conducted following procedures similar to the uptake experiments. Medakas were placed in ^{3}H-free medium and fed 25 mg of 20,000 Bq g^{-1} ^{3}H-brine shrimp once daily. Notably, the ^{3}H-brine shrimp were not freeze-dried, as preliminary data showed that, when placed in ^{3}H-free medium, the TFWT in ^{3}H-brine shrimp rapidly exchanged with H_2O, reducing TFWT concentrations to near background levels, much lower than the concentration of OBT. After 4 days, ^{3}H-medakas were transferred to containers with 200 ml of tritium-free medium and 25 mg of ^{3}H-free brine shrimp once daily. At predetermined intervals (0, 24, 48, 72 and 96 h), medakas were collected, washed, freeze-dried, weighed, digested and analysed for radioactivity using LSC as described above.

To quantify the ^{3}H distribution in various organs, ^{3}H-medakas before and after 96 h of elimination were sacrificed and preprocessed as described. Brain, liver, gill, gut and carcass tissues were harvested, freeze-dried, weighed, digested and their radioactivity determined using LSC following the same procedures.

Identification of ^{3}H-labelled amino acids in medaka brain

After 4 days of daily feeding with 25 mg of ^{3}H-brine shrimp at a body burden of 20,000 Bq g^{-1}, the exposed medakas via trophic transfer were transferred to a 3-day elimination scenario. The ^{3}H-accumulated medakas were then sacrificed, and brain tissues were carefully dissected. Brain samples were rapidly frozen in liquid nitrogen, and stored at −80 °C. To ensure sufficient detection limits, ten brain samples were pooled as one replicate. For amino acid extraction, brain samples were homogenized with 200 µl of H_2O and 20 ceramic beads (0.1 mm) using a homogenizer, followed by extraction with 800 µl of MeOH and 10 min of sonication at 4 °C. Samples were then mixed with 1 ml of 6 M HCl and incubated for 2 h, followed by centrifugation at 16,200 × *g* for 15 min to remove insoluble material. Supernatants were transferred to high-performance liquid chromatography glass vials for liquid chromatography–mass spectrometry analysis.

Chromatographic separation was performed using an Exion UHPLC system (Shimadzu, Japan) coupled to a quadrupole time-of-flight mass spectrometer (Triple TOF 7600, SCIEX) equipped with a C18 column (Waters ACQUITY HSS T3, 2.1 × 100 mm, 1.8 µm). The column temperature was maintained at 40 °C, with a flow rate of 0.3 ml min^{-1}. The injection volume was 5 µl. Mobile phase A was water, and mobile phase B was methanol; both containing 0.1% (*v*:*v*) formic acid and 2 mM ammonium formate. The gradient programme was as follows: 0–1.0 min, hold 3% B; 1.0–1.5 min, linear gradient to 15% B; 1.5–2.5 min, linear gradient to 50% B; 2.5–25 min, linear gradient to 67.4% B; 25–26.1 min, linear gradient to 98% B; 26.1–28 min, hold 98% B; 28–28.1 min, linear gradient to 3% B; and 28.1–30 min and hold 98% B. Mass spectrometry acquisition was performed in electrospray ionization+ mode.

The parameters were as follows: ion spray voltage, 5,500 V for the positive mode and −4,500 V for the negative mode; temperature, 350 °C; nebulizing gas, 55 psi; heating gas, 55 psi; and curtain gas, 35 psi. Time-of-flight mass spectrometry information-dependent acquisition mass spectrometry was performed using the following criteria: ions that exceed 100 cps; ion tolerance 50 mDa; collision energy was ramped over an interval by entering a collision energy spread value; and the collision energy and collision energy spread were set at 35 and 15 eV, respectively. Data were acquired from *m*/*z* 50–1000 in full-scan time-of-flight mass spectrometry and information-dependent acquisition mass spectrometry mode.

Radionuclide risk assessment

The primary pathway for human internal exposure to these radionuclides is through the consumption of seafood, including fish, shrimp and seaweeds. This internal exposure is calculated on the basis of both seafood consumption and the concentrations of radionuclides in the food. The radionuclide concentrations in seafood are typically determined using a CF approach, which transfers radionuclide concentrations from seawater to biota (Table 1)[15]. The associated human health risk is proportional to the radiation dose received, measured as the energy absorbed by the body from ionizing radiation with a unit of sievert (Sv). The rate of radioactive decay (for example, in a unit of Bq l^{-1}) is transferred by the DC, which depends on the type of radiation decays (for example, α or β type) and the half-life of the radionuclides[35]. The total internal dose is then calculated as the sum of radionuclide concentrations in fish and algae, each weighted by the corresponding DC.

We adopted a simplified approach to estimate the concentrations of radionuclides in seafood using the following parameters: (1) the CF, which represents the enrichment of radionuclides in biota relative to ambient seawater and is defined as the concentration ratio between biota and seawater, and (2) the BMF, which describes the radionuclide transfer efficiency from prey to predator and is defined as the ratio between specific activity in the predator to that in the prey. On the basis of these parameters, the concentrations in edible algae (C_{algae}) and fish (C_{fish}) were calculated from the modelled seawater concentration (C_{sw})

$$C_{algae} = C_{sw} \times CF_{algae} \quad (1)$$

$$C_{zooplankton} = C_{sw} \times CF_{zooplankton} + C_{algae} \times BMF_{algae \to zooplankton} \quad (2)$$

$$C_{fish} = C_{sw} \times CF_{fish} + C_{zooplankton} \times BMF_{zooplankton \to fish} \quad (3)$$

where CF_{algae}, $CF_{zooplankton}$ and CF_{fish} represent the CFs calculated on the basis of the specific activities of tritiated seawater and exposed *D. salina* (14 day), brine shrimp (4 day) and medaka (4 day), respectively. $BMF_{algae \to zooplankton}$ was calculated using the specific activity of brine shrimp relative to the added *D. salina*, while $BMF_{zooplankton \to fish}$ was calculated using the specific activity of fish relative to the added zooplankton. The IEDI was then calculated using the C_{food} and the corresponding seafood intake (*I*)

$$IEDI = I \times \sum_{i=1}^{n} C_{food}^{i} \times DC^{i} \quad (4)$$

$$IEDI = I_{algae} \times CF_{algae} \times DC + I_{algae} \times CF_{fish} \times DC \quad (5)$$

The seafood intake data were obtained from the Food and Agriculture Organization of the United Nations[28]. The radiation dose to humans depends on both the age of the individual and the specific characteristics of the radionuclides[61]. DCs for individual radionuclides, expressed in nanosieverts (nSv) per becquerel (Bq), incorporate human physiology, radiation physics and the spatiotemporal deposition of energy associated with the consumption of radionuclide-containing food. These DCs are used to calculate the IEDI, and we originally employed the DC data provided by the ICRP[35].

Reporting summary

Further information on research design is available in the Nature Portfolio Reporting Summary linked to this article.

Data availability

All data needed to evaluate the conclusions in the paper are present in the paper and/or the Supplementary Information files. Source data are provided with this paper.

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Author contributions

L.M. conceptualized and planned the project. S.D., K.L., Y.M., P.X. and L.Z. carried out figure preparation, the main experiments and data interpretation. S.D. and Y.Z. performed the model establishment. S.D. and Y.M. wrote the manuscript, with input from all authors. F.C., S.W., J.D. and Z.C. reviewed and supervised the manuscript. All authors analysed results and approved the final manuscript.

Competing interests

The authors declare no competing interests.

Additional information

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Reporting Summary

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Statistics

For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.

n/a	Confirmed
☐	☒ The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement
☐	☒ A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒ The statistical test(s) used AND whether they are one- or two-sided *Only common tests should be described solely by name; describe more complex techniques in the Methods section.*
☒	☐ A description of all covariates tested
☒	☐ A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
☐	☒ A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
☐	☒ For null hypothesis testing, the test statistic (e.g. *F*, *t*, *r*) with confidence intervals, effect sizes, degrees of freedom and *P* value noted *Give P values as exact values whenever suitable.*
☒	☐ For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☒	☐ For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☒	☐ Estimates of effect sizes (e.g. Cohen's *d*, Pearson's *r*), indicating how they were calculated

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection	1. The D.salina, brine shrimp and adult marine medakas were cultured in an artificial climate incubator (PRX-2000D, Ningbo Saifu Experimental Instrument Co., Ltd, China) 2. The activity of HTO or OBT was counted using a liquid scintillation counter (LSC; Tri-Carb 5110 TR; PerkinElmer, United States). 3. The absorbance of D.salina was quantified by UV-vis spectrophotometer (Carry 50, Germany). 4. The cell density of the stock suspension was determined using a Neubauer-improved counting chamber (Marienfeld, Germany). 5. The D.salina cells were observed using an inverted light microscope (Eclipse Ti, Nikon). 6. The samples were freeze-dried using a lyophilizer (Labconco, America). 7. The medakas activity were recorded for 10 min and videos were processed with Noldus Ethovision® XT16 (Noldus Information Technologies, Inc.) tracking software.
Data analysis	1. Numerical data were statistically analyzed by SPSS 22.0 (PASW Statistics, IBM Company). 2. The graphs were generated with the help of GraphPad Prism software (GraphPad Software 8.0), OriginLab (Origin 2023b) and Microsoft excel software. 3. The heatmap of trajectories was with Noldus Ethovision® XT16 (Noldus Information Technologies, Inc.) tracking software.

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Portfolio guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our policy

All data required to to interpret, verify and extend the research in the article are provided through a link in the manuscript and available from the corresponding author upon reasonable request.

Research involving human participants, their data, or biological material

Policy information about studies with human participants or human data. See also policy information about sex, gender (identity/presentation), and sexual orientation and race, ethnicity and racism.

Reporting on sex and gender	This study did not involve human
Reporting on race, ethnicity, or other socially relevant groupings	N/A
Population characteristics	N/A
Recruitment	N/A
Ethics oversight	N/A

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Field-specific reporting

Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☒ Life sciences ☐ Behavioural & social sciences ☐ Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	No statistical method was used to predetermine the sample size for each study. For property measurement experiments, samples were prepared and tested at least three times. Notably, (1) the number of algae cells was determined on the preliminary test results as well as the minimal requirements in Student T-test; (2) the number of brine shrimp was calculated based on the recommended intake of adult medakas.
Data exclusions	No data was excluded from the analyses.
Replication	1. HTO organification by algae: three replicates (Fig. 1). 2. Enrichment and transformation of 3H through exposing HTO or 3 H-algae (tritiated D. salina) to brine shrimp: three replicates (Fig. 2). 3. Enrichment and transformation of 3 H through exposing HTO or 3 H-brine shrimp (tritiated brine shrimp) to medaka: three replicates (Fig. 3).
Randomization	All experiments were randomly allocated into experimental groups by throwing dice.
Blinding	The investigator was not blind to the treatment and sampling collection, which can not be conducted in a blind way. However, several critical experiments, including the HTO exposure of D. salina, the HTO exposure of brine shrimp, the HTO exposure of medaka, and the behavioral effects of HTO on medaka, were performed random. Blinding was not available in some experiments and studies because the operator could easily identify the structure by characteristic signals (Microscopy images of algae, Absorption spectra of algae).

Behavioural & social sciences study design

All studies must disclose on these points even when the disclosure is negative.

Study description	*Briefly describe the study type including whether data are quantitative, qualitative, or mixed-methods (e.g. qualitative cross-sectional,*

Study description	*quantitative experimental, mixed-methods case study).*
Research sample	*State the research sample (e.g. Harvard university undergraduates, villagers in rural India) and provide relevant demographic information (e.g. age, sex) and indicate whether the sample is representative. Provide a rationale for the study sample chosen. For studies involving existing datasets, please describe the dataset and source.*
Sampling strategy	*Describe the sampling procedure (e.g. random, snowball, stratified, convenience). Describe the statistical methods that were used to predetermine sample size OR if no sample-size calculation was performed, describe how sample sizes were chosen and provide a rationale for why these sample sizes are sufficient. For qualitative data, please indicate whether data saturation was considered, and what criteria were used to decide that no further sampling was needed.*
Data collection	*Provide details about the data collection procedure, including the instruments or devices used to record the data (e.g. pen and paper, computer, eye tracker, video or audio equipment) whether anyone was present besides the participant(s) and the researcher, and whether the researcher was blind to experimental condition and/or the study hypothesis during data collection.*
Timing	*Indicate the start and stop dates of data collection. If there is a gap between collection periods, state the dates for each sample cohort.*
Data exclusions	*If no data were excluded from the analyses, state so OR if data were excluded, provide the exact number of exclusions and the rationale behind them, indicating whether exclusion criteria were pre-established.*
Non-participation	*State how many participants dropped out/declined participation and the reason(s) given OR provide response rate OR state that no participants dropped out/declined participation.*
Randomization	*If participants were not allocated into experimental groups, state so OR describe how participants were allocated to groups, and if allocation was not random, describe how covariates were controlled.*

Ecological, evolutionary & environmental sciences study design

All studies must disclose on these points even when the disclosure is negative.

Study description	*Briefly describe the study. For quantitative data include treatment factors and interactions, design structure (e.g. factorial, nested, hierarchical), nature and number of experimental units and replicates.*
Research sample	*Describe the research sample (e.g. a group of tagged Passer domesticus, all Stenocereus thurberi within Organ Pipe Cactus National Monument), and provide a rationale for the sample choice. When relevant, describe the organism taxa, source, sex, age range and any manipulations. State what population the sample is meant to represent when applicable. For studies involving existing datasets, describe the data and its source.*
Sampling strategy	*Note the sampling procedure. Describe the statistical methods that were used to predetermine sample size OR if no sample-size calculation was performed, describe how sample sizes were chosen and provide a rationale for why these sample sizes are sufficient.*
Data collection	*Describe the data collection procedure, including who recorded the data and how.*
Timing and spatial scale	*Indicate the start and stop dates of data collection, noting the frequency and periodicity of sampling and providing a rationale for these choices. If there is a gap between collection periods, state the dates for each sample cohort. Specify the spatial scale from which the data are taken*
Data exclusions	*If no data were excluded from the analyses, state so OR if data were excluded, describe the exclusions and the rationale behind them, indicating whether exclusion criteria were pre-established.*
Reproducibility	*Describe the measures taken to verify the reproducibility of experimental findings. For each experiment, note whether any attempts to repeat the experiment failed OR state that all attempts to repeat the experiment were successful.*
Randomization	*Describe how samples/organisms/participants were allocated into groups. If allocation was not random, describe how covariates were controlled. If this is not relevant to your study, explain why.*
Blinding	*Describe the extent of blinding used during data acquisition and analysis. If blinding was not possible, describe why OR explain why blinding was not relevant to your study.*

Did the study involve field work? ☐ Yes ☐ No

Field work, collection and transport

Field conditions	*Describe the study conditions for field work, providing relevant parameters (e.g. temperature, rainfall).*
Location	*State the location of the sampling or experiment, providing relevant parameters (e.g. latitude and longitude, elevation, water depth).*
Access & import/export	*Describe the efforts you have made to access habitats and to collect and import/export your samples in a responsible manner and in*

Access & import/export	*compliance with local, national and international laws, noting any permits that were obtained (give the name of the issuing authority, the date of issue, and any identifying information).*
Disturbance	*Describe any disturbance caused by the study and how it was minimized.*

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

n/a	Involved in the study
☒	☐ Antibodies
☒	☐ Eukaryotic cell lines
☒	☐ Palaeontology and archaeology
☐	☒ Animals and other organisms
☒	☐ Clinical data
☒	☐ Dual use research of concern
☒	☐ Plants

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☒	☐ Flow cytometry
☒	☐ MRI-based neuroimaging

Animals and other research organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research, and Sex and Gender in Research

Laboratory animals	1. D. salina were obtained from Shanghai Guangyu Biological Technology Co., Ltd. 2. The cysts of brine shrimp were purchased from the Institute of Marine Sciences, Shandong and stored at 4 °C before experiments. 3. The adult marine medakas (>3 months old, body lengths of 31.9 ± 1.4 mm) were purchased from Shanghai Feixi Biotechnology.
Wild animals	No wild animals were used in the study.
Reporting on sex	The findings were supposed to apply to both female and male marine medakas. This study is on the accumulation and digestion process of 3H in organism and the result should sex-independent.
Field-collected samples	No field-collected samples were used in the study.
Ethics oversight	All animal experiments were strictly performed under the guidelines of the Chinese Council for Animal Care, approved by the Animal Care Committee of the Laboratory Animals in Nanjing University.

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Plants

Seed stocks	*Report on the source of all seed stocks or other plant material used. If applicable, state the seed stock centre and catalogue number. If plant specimens were collected from the field, describe the collection location, date and sampling procedures.*
Novel plant genotypes	*Describe the methods by which all novel plant genotypes were produced. This includes those generated by transgenic approaches, gene editing, chemical/radiation-based mutagenesis and hybridization. For transgenic lines, describe the transformation method, the number of independent lines analyzed and the generation upon which experiments were performed. For gene-edited lines, describe the editor used, the endogenous sequence targeted for editing, the targeting guide RNA sequence (if applicable) and how the editor was applied.*
Authentication	*Describe any authentication procedures for each seed stock used or novel genotype generated. Describe any experiments used to assess the effect of a mutation and, where applicable, how potential secondary effects (e.g. second site T-DNA insertions, mosiacism, off-target gene editing) were examined.*