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Temperature Measurements from Oxygen Isotope Ratios of **Fish Otoliths**

Abstract. Measurements have shown that the temperature of a fish's habitat can be deduced from the oxygen isotope ratio of its otoliths (ear bones). Isotope ratios obtained from fossil otoliths indicate a water temperature which agrees wiht that found by isotope measurements on associated benthonic foraminifera.

Oxygen isotope measurements on calcium carbonate laid down by certain marine animals in the form of a shell or skeleton can give the temperature of the water in which the animals lived. Since the original work in oxygen isotope measurements by Urey et al. (1) many different forms of marine fossil carbonate, including belemnite guards, molluscan shells, and foraminiferal tests, have been used for this purpose.

Although fish form no suitable skeleton, their otoliths (ear bones) are composed of calcite and they have often been preserved as fossils in sediments. Otoliths from both living and fossil fish were studied, and the results given in this report indicate that fish form their otoliths in isotopic equilibrium with the sea water in which they live and that the temperature of their habitat can be deduced from the oxygen isotope ratio of these otoliths.

Suitably sized living fish that had lived under conditions of controlled temperature were not available for study, so samples were obtained of species that had known habitats, where the temperature range was also known. The isotopic "temperature" determined from an otolith is an average of the temperature fluctuations from winter to summer-possibly a weighted average if the calcite was not laid down at an even rate all year round.

Results of oxygen isotope measurements on fossil otoliths contained in Tertiary sediments from New Zealand gave temperatures close to the bottom temperature, based on similar measurements on associated mollusk shells and benthonic for aminifera tests (2).

Fossil otolith samples were cleaned in an ultrasonic bath for 2 minutes; they were then dried and ground to a powder. Otoliths of living fish were powdered and soaked in a 5 percent aqueous solution of sodium hypochlorite for 3 days to remove any organic material incorporated in the calcite. (I found that sodium hypochlorite treatment is unnecessary with fossil otolith material.) After soaking, the samples were rinsed with distilled water and dried. Powdered samples were reacted with 100 percent phosphoric acid under vacuum in a glass reaction tube at 25°C. Oxygen and carbon isotope ratios of the resulting carbon dioxide were measured on a Nuclide Analysis Associates 60°, 6-inch (15-cm), double collector mass spectrometer.

The results are given in Tables 1 and 2. Oxygen isotope ratios are given in the δ notation, that is,

$$\delta O^{18} = \left[\frac{(O^{18}/O^{16})_{sample}}{(O^{18}/O^{16})_{PDB}} - 1 \right] 1000$$

with respect to PDB (carbon dioxide prepared by phosphoric acid evolution, Peedee belemnite carbonate from the University of Chicago; it is an international standard for carbon and oxygen isotope measurements). The analytical error in each δO^{18} measurement is \pm 0.2 (3). The temperature was calculated from the result of the oxygen isotope measurement by using the equation of Epstein et al. (4, 5). The expected temperature for living fish is the mean of summer and winter temperatures of the water in the area where the fish lived (6).

The fossil otoliths that I examined were from various Pliocene and MioTable 1. Results of oxygen isotope measurements on otoliths from living fish. Analytical error in each δO^{18} measurement is ± 0.2 (3).

Sample	δO ¹⁸ (with respect to PDB)	Temperature (°C)		
		Calc. iso- topic	Ex- pected	
Tarakihi (C	heilodactylı	is macro	pterus)	
1st specimen				
1st otolith	2.1	8.3	8 to 10	
2nd otolith	2.1	8.3	8 to 10	
2nd specimen				
1st otolith	2.0	8.7	8 to 10	
Butterfi	sh (Corido	dax pullu	is)	
1st specimen		•	-	
2 otoliths				
combined	1.4	10.9	12 to 13	
2nd specimen				
2 otoliths				
combined	1.1	12.0	12 to 13	
Snapper	(Chrysoph	rys aurat	us)	
1st specimen			-	
1st otolith	0.2	15.7	15 to 16	

cene sediments and were thought to have been deposited at depths of a few hundred meters. Unsuccessful attempts were made to identify the fossil otoliths from published data and by comparison with otoliths from Recent species. However, before the powdered samples were prepared the otoliths from each sediment sample were grouped arbitrarily on the basis of their shape. Each group was clearly distinct but may have contained otoliths from several species of fish. Each powdered sample was made up of one complete otolith, and this normally weighs 5 to 10 mg. A typical series of results for one sediment sample is given in Table 2.

Results (Table 2) obtained from measurements on benthonic foraminifera indicate that the bottom water temperature was 8° to 9°C. The otolith

Table 2. Results of oxygen isotope measurements on fossil otoliths contained in an Upper Miocene sediment from Bells Creek, Wairarapa, New Zealand. Analytical error in each δO^{18} measurement is ± 0.2 (3).

δO ¹⁸ (with respect to PDB)	Calc. isotopic temp. (°C)
c foraminifera	
2.2	8.0
1.8	9.4
toliths	
2.2	8.0
2.0	8.7
2.0	8.7
1.7	9.7
1.4	10.9
1.3	11.3
	δQ ¹⁸ (with respect to PDB) c foraminifera 2.2 1.8 toliths 2.2 2.0 2.0 1.7 1.4 1.3

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results suggest that the species of fish represented by group A lived at or near the bottom of the water column, whereas those of group B lived above the bottom for at least part of their lives. From foraminiferal and molluscan paleoecological evidence, the depth of deposition of this sediment is estimated to be about 500 m.

Extension of the oxygen isotope technique to the study of fish ecology has several possibilities. Valuable information on fish migration may be forthcoming if oxygen isotope temperatures differ from the water temperature where the fish are collected. Small differences may reflect seasonal movement to waters of different latitude or depth.

Calcium carbonate laid down in equilibrium with fresh water has a markedly different oxygen isotope from that laid down in equilibrium with sea water. This difference is reflected in anomalously high isotopic temperatures. Anomalous results obtained from a fish's otolith may indicate that migration into rivers or estuaries is an important phase in the life-span of the fish.

Results of isotope measurements on otoliths may also be useful in determining the bottom temperature for studies in paleoecology. In deep-water sediments, benthonic fossil carbonate is often scarce. From results given in Table 2 it is reasonable to conclude that the lowest temperature obtained from a number of otoliths from a sediment sample is generally the bottom temperature. This has been demonstrated for sediments deposited at a depth of at least 500 m but may not necessarily be the case for sediments deposited at very great depths.

There is only a slight probability of a fish's remains being deposited in an area of bottom temperature warmer than that of its normal habitat. Of 40 otoliths from eight Tertiary sediments of different ages, not one recorded a temperature lower than the bottom temperature for the locality, as indicated by isotope measurements on other fossil forms.

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- 3. The δO^{18} of the mass spectrometer CO_2 standard has been determined to ± 0.05 by comparison with international standards. Duplicate runs on the same sample are normally (90 percent) within 0.10 on δO^{18} . The total

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analytical error on each single result is therefore estimated conservatively at ± 0.2 . Results given in the tables have been rounded off to the nearest 0.1.

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- 5. No major influx of fresh water occurs near the areas where the fish lived, and, although the oxygen isotope ratios of the waters have

not been measured, salinity values indicated open ocean water; hence no corrections have been applied to the results.

- I acknowledge the assistance given by J. Moreland, Dominion Museum, and L. Paul, Fisheries Research Laboratory, in selecting samples and supplying the ecological data necessary for this study.
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Flaxedil (Gallamine Triethiodide): Evidence for a Central Action

Abstract. A central action of gallamine triethiodide has been demonstrated in cats with permanently implanted electrodes that permit direct repetitive stimulation and recording of afterdischarges from cerebral cortex. Systemically administered gallamine produced a consistent and reproducible augmentation of duration of afterdischarge at doses just sufficient to produce skeletal muscle paralysis. Simultaneous examination of expiratory carbon dioxide, blood pressure, body temperature, blood glucose, and direct cortical response to brief single stimuli failed to reveal any consistent peripheral change to which the centrally observed effect could be attributed.

Gallamine triethiodide [1,2,3,-tri(2diethylaminoethoxy) benzene triethiodide, Flaxedil] is a synthetic tubocurarine substitute first described by Bovet (1). It produces paralysis by blocking the action of acetylcholine at muscle end-plate receptors. Because of a belief that gallamine has no central action when administered in paralyzing doses, it is used, together with infiltration anesthesia, to immobilize animals for experiments involving the central nervous system. During such an experiment we noticed that afterdischarge obtained from the cerebral cortex of cats immobilized with gallamine was of much

longer duration than that from comparable freely roving animals. We designed some experiments to determine the effect of gallamine on mechanisms of cortical afterdischarge.

Platinum electrodes held in a plastic plate were fixed over the pial surface of the suprasylvian gyrus of cats $(2\frac{1}{2}$ to 5 kg) of both sexes (2). Preparation of these animals for "acute" experimentation was done under ether anesthesia. Arterial, venous, and tracheal cannulae were fitted, and after primary closure all wounds were infiltrated with long-acting local anesthetic (Efocaine, Fougera). The animals were made com-



Fig. 1. (A) Afterdischarges at a threshold current of 1.5 ma are shown before the administration of any drug. *I*, Expiratory CO₂, calibration is 5 percent CO₂ (see spontaneous respirations); *II*, marks for one second; *III*, abdominal aortic blood pressure, calibration 0 to 100 mm-Hg; *IV*, electrocorticogram showing stimulus artifact ($2\frac{1}{2}$ seconds) and afterdischarge, calibration 1.0 mv. (B) The sequence and the stimuli are the same as in (A) except that (B) was obtained 15 minutes after intravenous administration of gallamine triethiodide (6.25 mg/kg). The animal was mechanically ventilated to near its level of carbon dioxide before administration of the drug, and the afterdischarge was of longer duration.