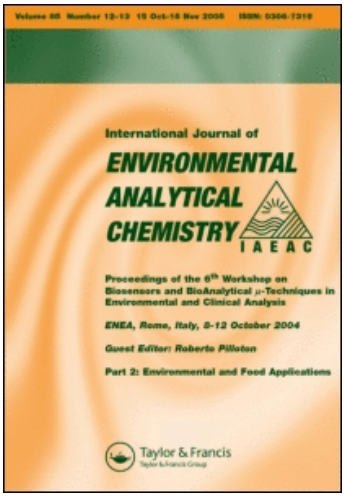

International Journal of Environmental Analytical Chemistry
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713640455

Determination of Cadmium in the Livers and Kidneys of Puffins by Carbon Furnace Atomic Absorption Spectrometry

J. M. Ottaway[a]; W. C. Campbell[a]
[a] Department of Pure and Applied Chemistry, University of Strathclyde. Cathedral Street, Glasgow, Scotland

To cite this Article Ottaway, J. M. and Campbell, W. C.(1976) 'Determination of Cadmium in the Livers and Kidneys of Puffins by Carbon Furnace Atomic Absorption Spectrometry', International Journal of Environmental Analytical Chemistry, 4: 3, 233 — 239
To link to this Article: DOI: 10.1080/03067317608071117
URL: http://dx.doi.org/10.1080/03067317608071117

Intern. J. Environ. Anal. Chem., 1976, Vol. 4, pp. 233–239

Printed in Great Britain

Determination of Cadmium in the Livers and Kidneys of Puffins by Carbon Furnace Atomic Absorption Spectrometry

J. M. OTTAWAY and W. C. CAMPBELL

Department of Pure and Applied Chemistry, University of Strathclyde, Cathedral Street, Glasgow G1 1XL, Scotland

(*Received August 11, 1975*)

KEY WORDS: Carbon furnace, atomic absorption spectrometry, cadmium, biological species.

A carbon furnace atomic absorption procedure is described for the determination of cadmium in the livers and kidneys of puffins, fratercula arctica. Samples are dried and weighed and 2 to 100 mg are dissolved in sulphuric and nitric acids. These solutions are analysed directly in the carbon furnace against aqueous standards and provide accurate results in the range 0·1 to 100 μg/g dry weight. The method is simple and rapid and requires much less of the small total sample than would be required for flame atomic absorption.

INTRODUCTION

The breeding populations of the Puffin, Fratercula Arctica in the south-west of England, the Irish sea area and south-west Scotland are known to have undergone a dramatic decrease since approximately the turn of this century. Colonies which previously held massive populations now have no puffins or negligible numbers. In 1969–70, ornithologists in the U.K. carried out a survey of the populations of seabird colonies. The results of the survey which was known as "Operation Seafarer", were recently published[1] and indicate that the serious decline in puffin populations was also affecting the major colonies in the north-west of Scotland at St. Kilda, Shiants, Clo Mor (Sutherland), etc. The counting of puffin populations at large colonies is extremely difficult due to inaccessibility of nest sites, the uncertainty as to

which burrows are occupied at any colony and the variation from hour to hour and day to day of attendance of birds at the colony.[2] Despite these limitations on the value of the data reported, it seems clear that the puffin is now in serious decline in the north-west of Scotland and recent reports not only support this conclusion but suggest that the decline has been continuing, if not accelerating, since 1969.[2–4] Although, populations of puffins at most smaller colonies on the east coast of Scotland and northern England, notably the Isle of May, have been increasing in recent years, the overall effect is of a dramatic decline in puffins breeding at colonies in the U.K.

The cause or causes of this dramatic and apparently systematic decline in puffin populations remain unexplained but a number of suggestions have been put forward.[1–6] Introduction of rats and predation by gulls, notably great black backed gulls may have affected some colonies but the decline is too widespread for these to be more than contributory factors. If the earlier decline in the south and west of the British Isles is linked to that in north-west Scotland, a continuous, systematic change can be postulated spreading over many years along the south and west coasts. This might suggest that the cause was linked to a change in climatic environment either changing the availability of food or changing the diet of the birds. Where the availability was decreasing, birds might be expected to move to areas where food supply was more plentiful and there is some evidence for this since recent increases in puffins in east Scotland are greater than can be accounted for by natural means.[4] A change of diet might affect the health of the birds or might increase susceptibility to particular pollutants. It is well known for example that certain toxic metals are concentrated from sea water by plankton and certain species of fish at very high concentration factors.[7,8] Pollution on its own appears an unlikely cause of the decline of puffins largely because of the long term nature of the decrease. Oil spills may have had serious effects on the population at some colonies but relatively few puffins are found washed up on beaches in an oiled condition in contrast to other auks notably guillemots. Parslow *et al.*[8] have shown that a number of recovered dead puffins had very low levels of pollutants such as DDE and PCB's found in other birds. Of the heavy metals investigated,[8] mercury, lead, cadmium, copper and zinc only the cadmium levels showed any significant difference between birds collected during the winter and those collected during the breeding season. Although there is too little data on which to reach even tentative conclusions, the puffins found on the west coast also had much higher levels of cadmium in their livers than those birds collected on the east coast. Of the pollutants measured this is the only distinct variation corresponding to the changes in puffin colonies and further work on cadmium is in progress at the present time.[9]

As with other animals, the puffin collects and concentrates cadmium in the liver and kidney, particularly the latter. The puffin is a small bird, total body weights being reported[8] in the range 234 to 394 grams, and the size of the livers varies in the range 5 to 20 grams and kidneys 3.5 to 6.0 grams. These samples are very small, when one considers the number of different analyses which will be of interest and the need for replicates. There will therefore be an advantage from the use of a small amount of sample in any determination provided the portion is representative and the distribution of the analyte within the sample is not too inhomogeneous.

The analysis of metals in samples of this type will normally be carried out by atomic absorption spectrometry and flame methods have been used to date.[8,9] The sensitivity of the flame necessitates relatively large portions of the sample which must then be ashed before analysis. Carbon furnace atomic absorption is generally about 10 to 100 times more sensitive for cadmium.[10] Atomisation of solutions made up in the nitrate and/or sulphate media frequently used for wet ashing or dissolution, has also been shown to give reproducible results which are free from matrix interference from a wide range of materials.[11] In this paper, we report a simple procedure, developed for the determination of cadmium in the livers and kidneys of puffins, using carbon furnace atomic absorption. This technique could be applied to the determination of other metals and could make a substantial contribution to research in this area.

EXPERIMENTAL

Apparatus

The instrument used for all measurements was a Perkin–Elmer, Model 306, atomic-absorption spectrometer equipped with an HGA 72 heated graphite atomiser and deuterium arc background corrector and coupled to an Electronik 194 strip chart recorder. A Perkin–Elmer Intensitron Hollow-Cathode lamp was used as the source.

The design and operation of the HGA-72 which is similar to the HGA 70 has been described in detail elsewhere.[12,13] Samples are atomised in a graphite tube, 5.3 cm long and 1 cm in diameter under an argon atmosphere. Samples were transferred to the centre of the tube by means of a 50 μl Eppendorf micropipette. The instrument has variable time and temperature selectors for sequentially drying, charring and atomising the samples and, once set this sequence of operations proceeds automatically. Signals are measured on the recorder, which for analysis purposes, is switched on only during the atomization stage at a rate of 3 cm min^{-1}.

Reagents

All reagents were of the highest available purity. Deionised water was used in the preparation of all solutions. A stock cadmium solution containing 1000 μg/ml cadmium was prepared from cadmium sulphate. This was diluted to 10 μg/ml for a working stock solution as required.

Procedures

1) *Preparation of calibration solutions* Dilute 5 ml of the working stock solution to 1 litre with water. This solution should be freshly prepared every day. Transfer 0, 2.0, 4.0 and 6.0 ml of this solution into 100 ml calibrated flasks, add 5 ml AnalaR sulphuric acid to each flask and dilute to the mark with water. These solutions contain 0, 0.001, 0.002 and 0.003 μg/ml cadmium in 5% v/v sulphuric acid. Under these conditions the calibration graph was found to be linear up to 0.003 μg/ml cadmium.

2) *Preparation of sample solutions* It is usual to express heavy metal concentrations in animal tissue as a function of the dry weight of the tissue. Solutions were therefore prepared as follows: Dry a suitable portion of the sample for two hours at 110°C. Weigh 2 or 10 mg of the dried sample and transfer to a PTFE beaker. A weight of 2 mg was found suitable for the cadmium concentration range 30 to 100 μg/g dry weight and 10 mg was used for samples containing less than 30 μg/g dry weight.

To the sample in the beaker add 5 ml sulphuric acid and place the beaker on a low temperature hot plate. After 5 minutes add nitric acid dropwise until all visible reaction has ceased. Heat the solution for a further 5 minutes before cooling. Transfer the cooled solution to a 100 ml calibrated flask and dilute to the mark with water. The dissolution procedure takes approximately 20 minutes.

3) *Instrument Operation* The operating conditions should be set up as follows:

Wavelength, nm	228.8
Lamp current, mA	10
Spectral band width, nm	0.7
Drying conditions	45s at 100°C
Ashing conditions	30s at 350°C
Atomisation conditions	10s at 2000°C
Volume of sample solution, μl	50
Scale expansion	×3
Argon flow	1.5 l min^{-1} at 40 lb in^{-2}

Sequentially inject 50 μl aliquots of standard and sample solutions into the

graphite tube and atomize using the above conditions. The deuterium arc background corrector should be operated and carefully matched to the hollow cathode light beam. After each atomisation, the maximum temperature button should be depressed to clear the tube. Derive the concentration of cadmium in the samples from a calibration graph obtained from the standards.

RESULTS AND DISCUSSION

The dissolution procedure was designed to be as simple and rapid as possible. Complete oxidation of the organic material is not necessary as the ashing stage can be completed in the carbon furnace. If sulphuric or nitric acids were used alone, a heavy precipitate was formed on dilution with water. Using a mixture of both acids, as described, sufficient oxidation is attained to allow dilution with no precipitation and reproducible 50 μl aliquots of the sample solution can then be added to the carbon furnace. Ashing was carried out in the carbon furnace at 350°C as at higher temperatures cadmium is volatilised and low results are obtained. The ashing procedure is essential to remove the smoke produced from the sulphuric acid. This gives a non-specific absorption signal at the cadmium wavelength of greater than 0.6 absorbance units and the background corrector does not completely compensate for it. In Figure 1, two signals from a typical liver sample are shown. Despite the background corrector being in use and correctly aligned a small non-specific absorption signal is obtained during the ashing stage. Cadmium recovery tests and measurement with a non-absorbing line were carried out to ensure that this signal was not even in part due to cadmium.

Four dead puffins were supplied by M. P. Harris of the Institute for Terrestial Ecology in Banchory. On analysis of the livers and kidneys from these puffins, very low results were obtained of approx. 0.1 μg/g dry weight in the livers and 0.2 μg/g dry weight in the kidneys. Recovery tests gave 100% recovery of cadmium added to the sample after weighing. Subsequently samples of livers and kidneys of puffins were received from J. L. F. Parslow of the Monks Wood Experimental Station of the Institute of Terrestial Ecology in Huntingdon. These samples had been analysed at Monks Wood by flame atomic absorption spectrometry. The results obtained using the carbon furnace procedure are given in Table 1, together with the results reported by Monks Wood. Within the limitations of the experiment samples were analysed at different times etc., the results appear to be in acceptable agreement with the exception of the kidney sample 4112. Repeat analyses carried out on different days using the carbon furnace were in good agreement and all the results suggest that cadmium is homogeneously distributed

within the samples. No significant error arises from the small sample size used in the carbon furnace method.

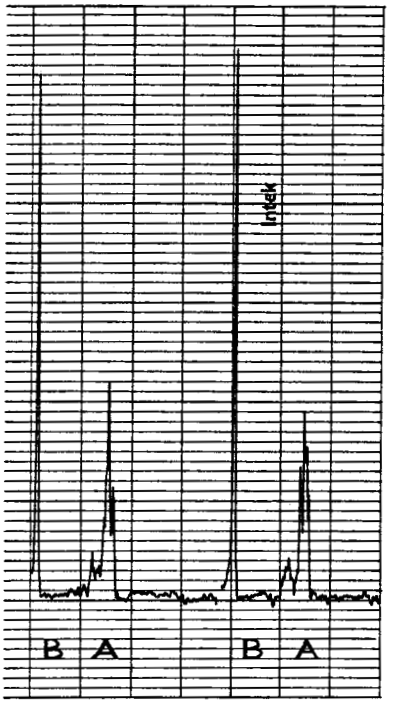

FIGURE 1 Recorder chart for the ashing and atomisation stages in the analysis of a typical liver sample. (A) Signal during ashing at 350°C (B) cadmium atomic absorption signal. Recorder chart speed 3 cm min^{-1}.

CONCLUSION

Carbon furnace atomic absorption spectrometry offers a simple and rapid method for the determination of cadmium in the livers and kidneys of puffins. The small portion of the sample used, allows faster dissolution and allows replicate analyses to be performed where necessary, without the consumption of too much sample. The method could be used with advantage for investigations of the cadmium toxicity of puffins at different locations, for the examination of trace metals in other species of birds, for the investigation of other toxic metals or for tests for the distribution of trace metals in different parts of any particular organ. The method makes use of the sensitivity and freedom from interference of the carbon furnace technique.

TABLE I

Analysis of puffins liver and kidney samples
Cd concentrations (μg/g dry weight)

Sample	Monks Wood Value	Strathclyde Value
3307 Liver	32	20,20
3981 Liver	27	24,29
3982 Liver	14	14,17
3983 Liver	19	12,13
3466 Liver	57	62,55
3987 Liver	15	15,13
4112 Liver	12	9,8
4368 Liver	13	13,12
3981 Kidney	90	93,89
3982 Kidney	86	76,73
3983 Kidney	90	90,97
4112 Kidney	33	58,51
4367 Kidney	14	17,14
4368 Kidney	96	81,87

Acknowledgements

We would like to thank M. P. Harris, J. L. F. Parslow and C. Lloyd for the provision of samples and helpful comments on this project. The work was made possible by the award to J. M. O. of a research equipment grant from the Royal Society for the purchase of the HGA 72.

References

1. S. Cramp, W. R. P. Bourne and D. Saunders, *The Seabirds of Britain and Ireland* (Collins, London, 1974).
2. M. de L. Brooke, *Bird Study* **19**, 1 (1972).
3. J. J. M. Flegg, *Bird Study* **19**, 7 (1972).
4. C. Lloyd (*Annual Seabird Census, Royal Society for the Protection of Birds, Sandy, Bedfordshire*), personal communication.
5. J. L. F. Parslow, *British Birds* **60**, 2 (1967).
6. W. R. P. Bourne, *New Scientist* **52**, 8 (1971).
7. J. H. Steel, A. D. McIntyre, R. Johnston, I. G. Baxter, G. Topping and H. D. Dooley, *Mar. Poll. Bull.* **4**, 153 (1973).
8. J. L. F. Parslow, D. J. Jefferies and M. C. French, *British Birds* **19**, 18 (1972).
9. J. L. F. Parslow, to be published.
10. G. F. Kirkbright, *Analyst* **96**, 609 (1971).
11. J. M. Ottaway, *Proc. Soc. Analyt. Chem.* **12**, 176 (1975).
12. D. C. Manning and F. J. Fernandez, *Atom. Absorption Newsl.* **9**, 65 (1970).
13. F. J. Fernandez and D. C. Manning, *Atom. Absorption Newsl.* **10**, 65 (1971).