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Revised technique for calculation of calcareous nannofossil accumulation rates

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ABSTRACT: A rapid and easy settling technique designed to estimate quantitative abundances of calcareous nannofossils is described. Compared with other methods, the time spent in weighing and other time-consuming steps are compensated by the possibility of standardizing and producing large series of samples at the same time. Formulas for the calculation of absolute abundances and accumulation rates are presented. A test in which the examination of several samples where the amount of sediment as well as other parameters have been changed demonstrates a random distribution of nannoliths across the slide. Variations in the calculated accumulation rates were minimal, suggesting high reliability.

INTRODUCTION

Coccolithophores, and probably most of the calcareous nannoplankton, are primary producers whose distribution in the oceans is controlled by several parameters. Sea surface temperature (SST), related to climatic zoning, is one of the most important limiting parameters e.g.: different species in varying abundances characterize equatorial to subpolar regions (McIntyre et al. 1970; Haq 1980; Winter et al 1994). At both local and global level, biolimiting nutrient distribution is another important factor related to coccolithophore abundance (e.g. Molfino and McIntyre 1990; Young 1994). Changes in SST or in nutrient concentrations in the oceans are related to paleoceanographic changes and knowledge of coccolithophore (nannofossil) abundance in sediments could therefore help to understand the evolution of the world's oceans.

The aim of the present study was to test an easy and rapid technique for estimating nannolith abundance per gram of sediment. This technique allows one to convert the amount of nannofossils into nannofossil fluxes when precise age models and sediment densities are available.

Several techniques have been proposed to estimate the absolute abundances of nannofossils (Wei 1988; Beaufort 1991; Okada 1992; Henriksson 1995). Some of these techniques are very time-consuming, or involve instrumental problems in the preparation of well spread surfaces for nannofossil counting, often meaning than many visual fields (nannofossils) must be counted. In other cases, the proposed techniques allow only afford an estimation of the true abundance, as for example in Backman and Shackleton (1983).

PROCEDURE

The aim of the technique proposed here is to obtain slides on which the nannoliths are distributed homogeneously, and to achieve the possibility of standardizing the procedure in order to save time. Our experience is that with this procedure it is possible to prepare sets of up to one hundred samples at one time; i.e., a rate of around 50 samples per day. This technique was tested using both deep-sea sediments, where nannofossils were the most important component, and sediments, in which the calcareous nannofossil component was diluted by organic or inorganic material.

The steps below describe the procedure, and should be followed carefully.

Step 1. Sample drying. A small amount of sediment is dried in a heater at a temperature of about 50°C.

Step 2. Weighing of an amount of dried sediment. This amount depends on the richness of the samples in nannofossils, as well as on the parameters discussed below. For routine analysis we used masses between 0.1 to 0.3 g.

Step 3. The weighed sediment is placed in a bottle, and a varying volume of distilled water is added (we used 10 to 30 ml). The mixture is shaken and, if necessary, placed in an ultrasound apparatus for a few seconds. For samples rich in organic matter, the distilled water can be substituted with H_2O_2 . It is advisable to leave the mixture for a few hours under conditions of regular





Scheme showing: a) tools and procedure used in this technique; b) way followed to observe 40 visual fields (1250x)

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TEXT-FIGURE 2

Number of nannoliths (asteroliths) counted in a given number of fields of view (magnification 1250x), in three duplicated samples. Starting weight and volume extracted with a micropipette where modified in each sample.

manual or automatic shaking. This is done in order to obtain a well disaggregated sample.

Step 4. After the mixture has been shaken, a fraction of the mixture is extracted with a micropipette (between 100 to 300 μ l). This volume is taken from the middle part of the bottle after waiting for a few seconds to allow the settling of possible aggregates and/or large particles.





Step 5. A coverslide is placed on the bottom of a Petri disk (with a diameter of 40 to 60 mm) and a mixture (0.05g/l) of distilled water and unflavoured gelatin ("Gold Gelatin") is added (to reduce natural surface tension). The volume extracted with the micropipette is then dropped onto the Petri disk and the fluid is mixed by pumping several times with the pipette, untill a uniform distribution is achieved (text-fig.1).

Step 6. The mixture is kept on a stable horizontal surface at a temperature of about 20 $^{\circ}$ C for 12 to 24 hours. After this time, the fluid is withdrawn from the Petri disk using short strips of filter paper placed at the edge of the disk (text-fig. 1). To avoid removing settled particles, this withdrawal must be very slow.

Step 7. After the disk has been dried a cover slide is mounted with Canada balsam or any other mounting media adequate for the calcite refraction index.

A preliminary check on a conventional smear slide (Perch-Nielsen 1985) can help to estimate nannofossil abundance in cores or stratigraphic sections and thus help to establish the standard (weight, volume...). The concentration per v.f. should permit easy counting.

NANNOFOSSIL PER GRAM OF SEDIMENT AND NANNOFOSSIL ACCUMULATION RATE

In order to estimate the absolute abundance of nannofossils counting was performed in random visual fields (v.f.) (text-fig. 1). The number of nannoliths counted was between 300 and 500, which according to Dennison and Hay (1967), for a probability of failure of 0.005, suggests that all species or groups over 1% were considered. The results obtained after the visualization of several fields of view are shown in text-figure 2. Using the following formula, the number of nannoliths per gram of sediment allowed us to estimate the amount of either one species or the total abundance of nannofossils in a given sample:

$$N = n * R^2 * V * r^{-2} * g^{-1} * v^{-1}$$

TABLE 1

Summary of the counting asteroliths in 40 visual fields in 3 duplicated samples, and the calculated absolute abundances in the sample (ODP 138, 852C-5H-3, 110-111cm).

samples	sum	mean	standard deviation	
g/µl	Σx	x	SD	nannofossils/g
0.1/300 a	500	12.5	3.81	1.4 4e+ 09
0.1/300 b	514	12.85	3.309	1.48e+09
0.2/150 a	489	12.23	3.591	1.41e+09
0.2/150 b	484	12.1	2.898	1.39e+09
0.2/100 a	327	8.175	2.934	1.41e+09
0.2/100 b	336	8.4	2.458	1.45e+09
		nannofossils/g	nannofossils/g	
		1.43e+09	2.93e+07	

where N is the number of nannofossils per gram of dry sediment; n the number of nannofossils counted in a random scanned area; R the radius of the Petri disk used; V the volume of water added to the dry sediment in the bottle, r the radius of the visual field used in the counting; g the dry sediment weight, and v the volume of mixture withdrawn with the micropipette.

The Nannofossil Accumulation Rate (NAR) was calculated following the procedure indicated by Mayer, Pisias, Janecek et al. (1992) as:

NAR= N * d * S

Where NAR is the Nannofossil Accumulation Rate (nannofossils * cm^{-2} * ky^{-1}); d is the estimated dry density (g * cm^{-3}) and S is the linear sedimentation rate (cm * ky^{-1}).

REPRODUCIBILITY

Several authors have studied the reproducibility and random distribution of particles, using different techniques to prepare microfossil slides (Moore 1973; Maher 1981; Laws 1983). To test our technique, we used an ODP sample (Leg 138, 852C-5H-3, 110-111cm) of early Pliocene age (Raffi and Flores 1995) that was rich in calcareous nannofossils. A commonly recorded and easily distinguished group of species in the sample was the asteroliths. For this reason we chose these forms for the experiment to test reproducibility and distribution across the slide. In order to obtain slides with different densities we changed the initial weight as well as the volume extracted with the micropipette. We used the same type of Petri disks (diameter: 47 mm) and bottle volume was also kept constant (10 ml). We also used the same microscope and magnification (diameter of v.f.) throughout the experiments.

We considered 3 samples in which the starting weight and volume of mixture was changed (Table 1). These samples were duplicated, and a total of 6 subsamples was studied. Text-figure 1 show the variability in the number of asteroliths counted in 40 random v.f. in duplicate slides (A and B). After these data had been converted into nannoliths per gram of sediment, only small discrepancies were observed (Table 1 and text-fig. 3).

In this way, it is possible estimate the abundance per gram of sediment (convertible into NAR) of one species or group of species. It is also possible to calculate the absolute abundance of other taxa following the same procedure or, in order to save time, proceed as follows:

1) After calculating the number of nannoliths per gram of a given species or group of species (N), the ratio between n and other taxa a (n/a) is estimated. In our experience, this counting

can be done in 5 to 15 v.f. (1250x), depending on the nannofossil concentration on the slide.

2) The absolute abundance (A) or NAR of these taxa a can be calculated by the following expression:

$$A = n/a * N$$

These results show that with random examination and with a significant number of nannofossils considered, the proposed technique affords absolute abundances of nannofossils with small margins of error. These absolute abundances can be directly converted into NAR with the same error, if the age models and density data are precise.

ACKNOWLEDGMENTS

The authors wish to express thanks to Dr. Monique Bonnemaison and Dr. Anders Henriksson for their valuable suggestions and critical comments on this study. Jesús Roncero is also acknowledged for his help in sample processing. Thanks are also due to N. Skinner for proofreading.

This study was supported by research grant CICYT ANT94-0277.

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- Manuscript received December 11, 1996 Manuscript accepted December 18, 1996