ISSN 1061-9348, Journal of Analytical Chemistry, 2010, Vol. 65, No. 13, pp. 1356–1363. © Pleiades Publishing, Ltd., 2010.
Original Russian Text © T.A. Velivetskaya, A.V. Ignat'ev, S.A. Gorbarenko, 2010, published in Mass-spektrometria, 2010, 6(3), pp. 214–220.

ARTICLES

An Installation for the Preparation of Foraminifera Micro Samples for the Isotopic Analysis of Carbon and Oxygen

T. A. Velivetskaya[a], A. V. Ignat'ev[a], and S. A. Gorbarenko[b]

[a] *Far East Geological Institute, Far East Division, Russian Academy of Sciences, pr. Stoletiya Vladivostoka 159, Vladivostok, 690022 Russia.*

[b] *Pacific Institute of Oceanology, Far East Division, Russian Academy of Sciences, ul. Baltiyskaya 43, Vladivostok, 69004 Russia*
e-mail: velivetskaya@mail.ru

Received December 10, 2008; in final form, March 30, 2009

Abstract—A technical modification of the traditional method of decomposition of carbonates in phosphoric acid was proposed for the determination of $\delta^{13}C$ and $\delta^{18}O$ in organogenic carbonate samples weighing 10–30 μg with an accuracy of 0.05%. The extraction of CO_2 was carried out under a vacuum at 95°C in 105% phosphoric acid. The isotopic composition of CO_2 was measured by CG–IRMS. The used feed-motion of samples to the reactor provides a consecutive delivery of the samples from the sample holders to the acid. This sample feeding method prevents the contamination of the acid with impurities from the surface of the sample, obviates the necessity of removing the sample holders from the acid, and allows the use of the same acid for performing a very large numbers of analyses. The accuracy and reproducibility of the $\delta^{13}C$ and $\delta^{18}O$ values was estimated by measuring international standards and comparing with the $\delta^{13}C$ and $\delta^{18}O$ values for organogenic carbonate samples obtained by the proposed method of analysis at a microgram level and the traditional method at a milligram level. The proposed technology was successfully used to study the isotopic composition of oxygen and carbon in the plankton and benthos foraminifers in order to reconstruct the Okhotsk Sea palaeotemperatures.

Keywords: foraminifera, stable isotope carbon and oxygen analysis, microanalysis

DOI: 10.1134/S1061934810130083

INTRODUCTION

Variations of carbon and oxygen stable isotopes in carbonates have been investigated for more than 50 years; the majority of researchers solve tasks and problems in palaeoclimatology. For the determination of palaeotemperatures and the construction of palaeclimate reconstructions, the high reproducibility ($\leq \pm 0.05‰$) of the results of measurement of the isotopic composition of carbon and oxygen is required. Organogenic carbonate and, in particular, foraminiferas are the basic objects of the isotopic analysis of carbon and oxygen. The analysis of the $^{13}C/^{12}C$ and $^{18}O/^{16}O$ isotope ratios in carbonates is based on the method of CO_2 extraction from carbonates in reaction with phosphoric acid. The use of phosphoric acid for isotopic analysis is preferable to other acids (for example aqueous solutions of HCl and H_2SO_4), because the product of the reaction, CO_2, does not exchange oxygen isotopes with phosphoric acid. The method of the decomposition of carbonates by phosphoric acid was suggested in 1950 [1] and is used in various modifications up to the present time. According to this method, extraction and purification of CO_2 are carried out under vacuum. A sample of carbonate (30–100 mg) and 100% H_3PO_4 (2 mL) are loaded in separate vials, pumped out, and thermostatted (25 ± 0.1°C). Then, the vial with the acid is turned over and the acid flows down to the sample. The reaction products, CO_2 and H_2O, fill the vacuum line, where their cryogenic separation takes place. Purified CO_2 is collected in a glass ampoule and analyzed on a mass spectrometer with a double inlet system. The reproducibility of the results of analysis is ±0.1‰ for $\delta^{18}O$ and $\delta^{13}C$. The method is labor-intensive, unproductive, and also demands a considerable amount of the analyzed sample. The improvement of the method proposed in a number of subsequent works was aimed at increasing the productivity and reducing the size of the sample. The decrease in time of the reaction by increasing temperature to 50°C and using the direct connection of the extraction system to the system of introduction into the mass spectrometer allowed for a considerable reduction in the time of analysis [2, 3]. The increase in productivity of the method through the simultaneous connection of several (from 12 to 24) reactors with acid to the vacuum system was described in [4]. In each reactor (glass flask with a ground valve), a portion of acid (10–20 mL) is poured; the reactors are placed in a water thermostat (50°C). The sample (2–5 mg) is loaded in a spoon soldered to the core of the valve. All reactors together with the tests are pumped down simultaneously to a pressure of 1.33×10^{-3} Pa. Then,

we turned the valve of each reactor, the reactor was thus removed from the vacuum system, and the sample dropped from the spoon to the acid. After the termination of the reaction, each reactor was connected to the vacuum line, and purification and collection of CO_2 for the subsequent mass spectrometric analysis were carried out. In this modification of the method, the same portion of acid was used to carry out a great number of analyses of carbonates. The authors analyzed about 1000 samples with the use of the same portion of acid; the reproducibility of the results of analysis was 0.07‰ for $\delta^{18}O$ and $\delta^{13}C$.

An alternative method was described in [5]. In this method, a reactor with acid is connected with a "roundabout" multisample device for the feed-motion of samples. Carbonate samples are placed in holders and loaded into roundabout cells along a circle. After pumping down, holders with samples are consistently dropped in the acid (90°C). The extracted CO_2 is continuously taken away from the reactor in a trap cooled with liquid nitrogen. After performing a series of analyses, sample holders are removed from the acid. The constant renewal of the acid in the reactor is required in this method.

As a result of improvements of the initial method, the high reproducibility (±0.05‰) of the results of the determination of $\delta^{18}O$ and $\delta^{13}C$ was achieved and the weight of the analyzed sample was decreased to 1–2 mg. The further decrease in the weight of samples and, respectively, the amount of analyzed carbon dioxide is limited by the fulfillment of the condition of the viscosity of the leakage of gas in the ion source of the mass spectrometer working in the mode of measurements with a double inlet system. With the development and application of the new technology of a gas inlet into the mass spectrometer and measuring the $^{13}C/^{12}C$ and $^{18}O/^{16}O$ ratios in a continuous flow of helium (continuous-flow isotope ratio mass spectrometry, CF–IRMS) [6], it became possible to analyze very small amounts of gas [7]. The use of this technology and the elaboration of the GasBench II, CTC Combi-PAL automated system of sample preparation (Thermo Finnigan, Bremen, Germany) allowed analysts to considerably increase the productivity of the method and decrease the weight of samples considerably. In this method, samples of carbonates (usually 100–450 μg) are loaded into test tubes of volume 10 mL and sealed with septum from butyl rubber. Test tubes with samples (72 pieces) are placed in a temperature-controlled aluminum sample holder. An automatic purge of test tubes with helium with the help of a specialized needle introduced into the test tube through the septum is provided for the removal of atmospheric air from the test tubes. After the purge, phosphoric acid (0.1 mL) is added into each test tube. After the termination of the reaction, the needle is again introduced into the test tube and the mixture of extracted gases is blown out with a flow of helium into the system of gas purification and input into the mass spectrometer. Water is removed from the helium flow on a Nafion dehumidifier. CO_2 is separated from the other gases on a chromatograph column and then enters the mass spectrometer with the helium flow through the system of an open flow splitter. Some problems and also the conditions of analysis at which this method gives results with the minimum error are described in a number of papers. In one of them [8], the authors conducted the reaction of carbonates (400 μg) with 100% H_3PO_4 at 26°C for 24–54 h, depending on the type of the analyzed samples; the reproducibility of the results for $\delta^{18}O$ and $\delta^{13}C$ was 0.1 and 0.2‰, respectively. Thus, the long (for more than 24 h) storage of CO_2 in test tubes with a needled rubber septum leads to the deterioration of reproducibility because of the fractionation of CO_2 and pollution with atmospheric air entering through the septum [9] in the test tube. In another work [10], 104% H_3PO_4 was used at a temperature of 72°C, which allowed one to decrease the time of the reaction to 1.5 h and reach higher reproducibility (0.06 and 0.08‰ for $\delta^{13}C$ and $\delta^{18}O$, respectively) for samples of a weight of 50–150 μg. However, at the decrease in the weight of samples from 50 to 10 μg, there was a decrease in the values of $\delta^{13}C$ and $\delta^{18}O$ (by 1.5 and 3.0‰, respectively) and the deterioration of the reproducibility of the results, on average, to 0.2‰. The analysis of small samples with high reproducibility is always a great problem and demands special carefulness and purity. Even trace amounts of atmospheric air remained after purging the test tubes [9] or the type and quality of the material of which test tubes [11] were made can affect the results. Experiments aimed at improving the reproducibility and productivity of isotopic analyses using the GasBench II equipment [12, 13] continue to be conducted, but problems of the analysis of microgram (10–30 μg) amounts of samples with high reproducibility, which is necessary for the determination of palaeotemperatures and climatic research, have not been solved yet. The analysis of foraminiferas as the main object of research for the construction of palaeoclimate reconstructions requires the determination of $\delta^{18}O$ with reproducibility not worse than 0.05‰ for sample weights of 10–30 μg.

In the present work, we proposed a technical modification of the classical method for the extraction of CO_2 and measurement of the isotopic composition with a reproducibility of 0.05‰ for samples weighing 10–30 μg. The method unites the classical principles of the vacuum extraction of CO_2 from carbonates and the up-to-date procedure of measuring micro amounts of gas in a constant helium flow. The combination of the classical extraction method with the up-to-date procedure of measurements allows high-precision measurements of the isotopic composition of carbonates for palaeoclimate reconstructions.

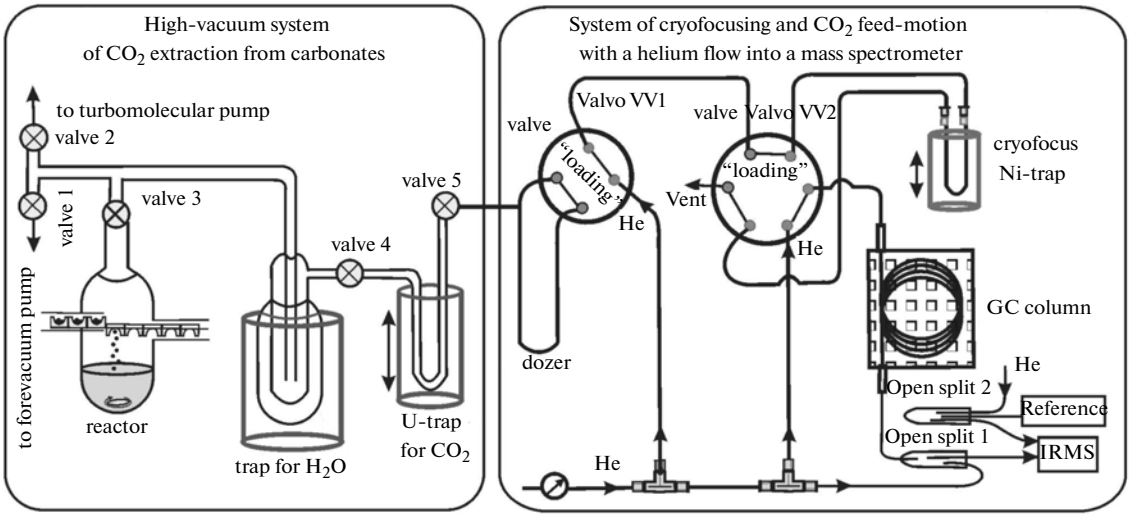

Fig. 1. Scheme of installation for the preparation of microgram samples of carbonates for the isotopic analysis of oxygen and carbon. The installation unites a vacuum system for CO_2 extraction from carbonates and a system of cryofocusing and feed-motion of CO_2 into the mass spectrometer with a helium flow.

EXPERIMENTAL

Equipment and materials. The scheme of installation for the preparation of microgram samples of carbonates and measurement of the isotope composition of carbon and oxygen is shown in Fig. 1. The installation includes a vacuum system for the extraction of CO_2 from carbonates and a system of cryofocusing and feed-motion of CO_2 with a helium flow into a mass spectrometer. The system of CO_2 extraction is made of glass and consists of a vacuum pumping line, a reactor for the decomposition of carbonates, and two cryogenic traps for H_2O and CO_2. A system of sample preparation is based on the traditional method of CO_2 extraction from carbonates, but has a number of technical modifications for improving the reproducibility of the results analysis of microgram carbonate samples. In particular, the reactor for the decomposition of carbonates has an original design of the feed-motion device, which allows consecutive delivery of samples from sample holders to the acid (Fig. 2). The reactor is made as a flask from a Pyrex tube 30 mm in diameter. Two tubes with an inner diameter 10 mm and length 15 cm are sealed into the reactor. The tubes enter the inner part of the flask to a depth of 1 cm so that the ends of the tubes are located close to the middle of the reactor. The centers of the tubes are shifted from each other to a height of 1.5 cm. One tube is intended for loading sample holders with samples (tube no. 1, Fig. 2); the other tube serves to take empty sample holders (tube no. 2, Fig. 2). A core from stainless steel 1.5 mm in diameter is placed into the tubes. Sample holders from stainless steel with samples are consistently steeled at the core. The tube for taking sample holders is vacuum sealed with a screw ring. The sample-feed device is connected to the tube with the samples via a connector with a viton seal. The sample-feed device includes a pusher (a core with a screw thread) and a rod of the pusher from stainless steel with a diameter of 1.5 mm. The pusher rod enters into the tube with samples through a sealing ring in the case of the connector. The rotation of a handle leads to the smooth forward movement of the rod inside the tube. The rod advances a series of sample holders along the core in the tube, shifting them outside the tube edge. The forced aside sample holder turns over on the core. The sample drops into the acid; and the sample holder remains on the core.

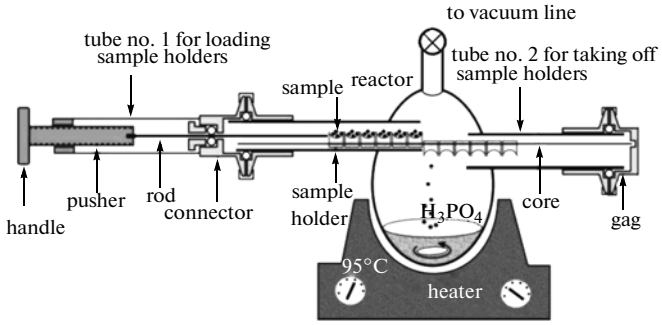

Fig. 2. Scheme of reactor with sample feed method to the acid.

The reactor contains 5 mL of 105% phosphoric acid prepared by the method described in [14]. The reactor is thermostatted at a temperature of 95 ± 0.2°C.

Traps for H_2O and CO_2 are consistently connected to the reactor (Fig. 1). The cryogenic trap for water is made from glass. The trap is vacuum-isolated with a

glass case sealed to the trap from which air is pumped out. The distance between the trap walls and the case is about 2 cm. A vessel with liquid nitrogen is put onto the cryogenic trap, the temperature in the trap achieves –(60–70)°C, which provides the purification of the extracted CO_2 from water. Pure CO_2 is condensed in a U-trap at the temperature of liquid nitrogen.

The vacuum system for the extraction of CO_2 is connected to the system of cryofocusing and feed-motion of CO_2 with a helium flow into the ion source of the mass spectrometer (Fig. 1).

The system of cryofocusing and gas feed-motion into the mass spectrometer includes the following components: a four-way valve VV1 (Valco Instruments Co. Inc., Houston, TX); a dozer of the volume 100 μL from a corrosion-proof tube of the diameter 1.6 mm; a six-way valve VV1 (Valco Instruments Co. Inc., Houston, TX); a cryofocusing Ni-trap from a corrosion-proof tube of the diameter 1.6 mm with a nickel wire inside (Thermoquest Finnigan); a chromatograph capillary column Poraplot-Q of the length 25 m and diameter 0.32 mm (Agilent Technologies); and an open splitter of gas flows no. 1 and no. 2 for the injection of the analyzed and standard gas into the mass spectrometer, respectively. The components are connected with quartz capillaries, and the helium flow rate is 2 mL min^{-1} in the system at a helium input pressure of 0.8×10^5 Pa.

Measurements of $\delta^{13}C$ and $\delta^{18}O$ were carried out on a Finnigan MAT-252 mass spectrometer (ThermoFinnigan, Bremen, Germany) with the use of the Finnigan Isotope Data acquisition program (ISODAT).

Procedure of analysis. Sample holders with samples are loaded into the reactor by stringing them on a core. A series of 16 samples can be loaded in the developed system. A sample feed device is connected to a tube with the samples. The reactor and the whole system are pumped down to 1.33×10^{-3} Pa for 2 h. Valves 2–5 of the vacuum system are opened, and valve VV1 is set in the loading position. Valve VV2 is set in the input position. A Dewar vessel with liquid nitrogen is placed on the H_2O-trap.

Before starting the reaction, a U-trap was placed in the vessel with liquid nitrogen, and then valve 2 was closed. The sample-feed device is put into action by rotating the pusher handle. The rod moves a series of sample holders along the core; the first sample holder leaves the edge of the tube and turns over. The sample drops to the acid. Water and carbon dioxide formed in the reaction are continuously frozen in the H_2O- and U-traps, respectively. Five minutes is sufficient for completing the reaction and collecting the extracted carbon dioxide in the U-trap. After that, we closed valve 4 and removed nitrogen from the U-trap. Carbon dioxide fills the dosing loop, then valve 5 is closed, and the dosing loop with a dose of CO_2 is isolated from the vacuum system. The vacuum system is connected to the pumping-out line.

To obtain a strongly focused peak of CO_2 in an ion source, CO_2 from the dosed volume is concentrated in a Ni-trap (Fig. 1). For this purpose, the Ni-trap is dipped into the Dewar vessel with liquid nitrogen, valve VV2 is switched to the trap in the position "loading", and valve VV1 is switched to the position "input". A helium flow enters the dosed volume and transfers CO_2 to the Ni-trap. One minute is enough to concentrate CO_2 from the dosed volume in the Ni-trap. After that, value VV2 is switched to the input position and nitrogen is removed from the Ni-trap. The helium flow carries defrozen CO_2 through a chromatograph column, in which the additional purification of CO_2 is carried out; then, CO_2 with the helium flow comes to the open splitter of the gas flow and, as a short impulse, to the ion source of the mass spectrometer. Valve VV1 is switched to the position loading and valves 5 and 2 of the vacuum system are opened for pumping out helium from the dosed volume.

The procedure of collecting CO_2 in the Ni-trap and gas input into the mass spectrometer takes about 5 min. For this time, the vacuum system was pumped out to high vacuum and is ready to perform the following reaction. The analysis of 16 samples takes nearby 3 h. After all samples are analyzed, empty sample holders are taken out. For this purpose, valve 3 is closed, the plug is removed from the tube for taking out sample holders, and the sample holders are taken out. Another set of sample holders with samples is loaded into the reactor and the system is prepared for performing a new series of analyses as described above.

RESULTS AND DISCUSSION

The accuracy and reproducibility of the values of $\delta^{13}C$ and $\delta^{18}O$ obtained by the proposed procedure of sample preparation was estimated by the results of the analysis of standard samples and by comparison of the results of the analysis by the proposed method and by a traditional method of CO_2 extraction from carbonates (Table 1). For this purpose, international standards NBS-18 ($\delta^{13}C_{V\text{-}PDB} = -5.01‰$, $\delta^{18}O_{V\text{-}PDB} = -23.0‰$), NBS-19 ($\delta^{13}C_{V\text{-}PDB} = +1.95‰$, and $\delta^{18}O_{V\text{-}PDB} = -2.2‰$) [15], as well as laboratory standard Coral-1 ($\delta^{13}C_{V\text{-}PDB} = -0.28‰$ and $\delta^{18}O_{V\text{-}PDB} = -3.69‰$), were used. The laboratory standard (aragonite of recent coral *Porites lutea*, New Caledonia) is calibrated by the NBS-18, NBS-19, and CO-8 international standards using the traditional procedure of CO_2 extraction in phosphoric acid at 25°C and the measurement on a double inlet system of a FLOOR-MAT 252 mass spectrometer.

The weight of the samples analyzed by the proposed procedure varied from 28 to 32 μg. Samples were weighed in sample holders on an ME-5 balance (Sartorius, Goettingen, Germany); the error was

Comparison of the results for $\delta^{13}C$ and $\delta^{18}O$ by the proposed and traditional methods of carbonate analysis

Sample	Proposed method. Sample weight is 28–32 μg		Traditional method. Sample weight is 2000 μg	
	$\delta^{13}C_{V\text{-}PDB} \pm 1\sigma$	$\delta^{13}O_{V\text{-}PDB} \pm 1\sigma$	$\delta^{13}C_{V\text{-}PDB} \pm 1\sigma$	$\delta^{13}O_{V\text{-}PDB} \pm 1\sigma$
NBS-19	1.90 ± 0.04	–2.28 ± 0.04	1.93 ± 0.01	–2.19 ± 0.03
NBS-18	–5.00 ± 0.02	–22.99 ± 0.06	–5.03 ± 0.01	–23.07 ± 0.02
Coral-1 standard	–0.28 ± 0.03	–3.68 ± 0.04	–0.28 ± 0.05	–3.69 ± 0.05
Ammonite-A*	0.84 ± 0.03	0.31 ± 0.06	0.89 ± 0.05	0.35 ± 0.06
Ammonite-A/1*	–0.84 ± 0.04	0.16 ± 0.07	–0.92 ± 0.05	0.10 ± 0.05
Ammonite-M4**	1.43 ± 0.07	0.27 ± 0.09	1.28 ± 0.06	0.16 ± 0.07
Ammonite-M5**	0.16 ± 0.04	–1.18 ± 0.08	0.19 ± 0.05	–1.15 ± 0.05
Elf-bolt-8D***	0.22 ± 0.05	–0.84 ± 0.05	0.20 ± 0.05	–0.81 ± 0.04

Notes: * Alaska, Lower Cretaceous;
** Madagaskar, Lower Cretaceous;
*** Pacific, Magellan Rise, Campanian Maastrchtian.

0.1 μg. The $\delta^{13}C$ and $\delta^{18}O$ values found for the international and laboratory standards had equally good reproducibility ≤ ±0.05‰ (Table 1). The difference between the measured and true values $\delta^{13}C$ and $\delta^{18}O$ for NBS-19 and NBS-18 [15] was within the permissible measurement error. The measured values of $\delta^{13}C$ and $\delta^{18}O$ for the laboratory standard also coincided with its accepted values.

The results of measurements of ammonite and elf-bolt samples are characterized, on the average, by slightly greater errors than those for standards, the maximum error was 0.07 and 0.09‰ for $\delta^{13}C$ and $\delta^{18}O$, respectively (Table 1). To estimate the reliability of the results obtained, the same samples were analyzed by the traditional method of CO_2 extraction in phosphoric acid at a temperature of 25°C [4]. The weighed portions of samples were 2–2.5 mg. The reproducibility of the results was, on the average, 0.05‰ for $\delta^{13}C$ and $\delta^{18}O$. A comparison of the values obtained by the traditional method of analysis at the milligram level and the proposed method at the microgram level shows good convergence of the results obtained by the two methods. The data in Table 1 show that the proposed procedure for the analysis of microgram amounts gives authentic results with good reproducibility for samples of about 30 μg.

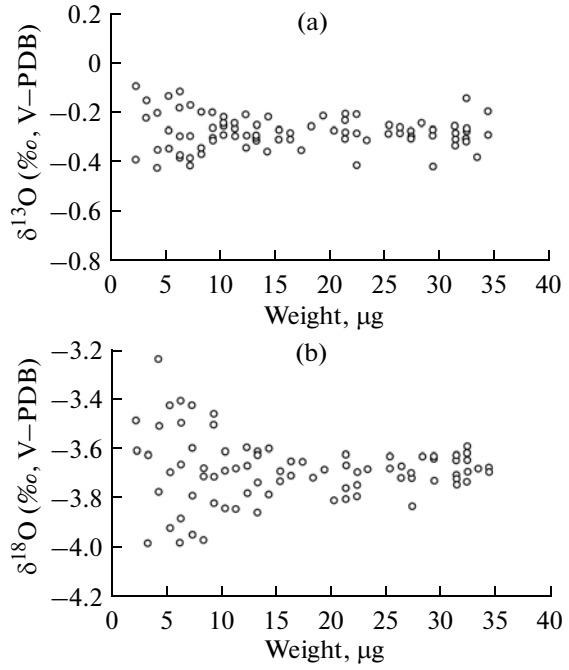

Fig. 3. $\delta^{13}C$ and $\delta^{18}O$ values of the laboratory standard Coral-1 as functions of sample weight.

Analyses of samples of the Coral-1 standard weighing from 30 to 2 μg were carried out for the determination of the minimum weight at which samples can be analyzed on the proposed installation with equal reproducibility. The results of analyses are presented in Fig. 3. The average value of the isotopic composition of carbon and oxygen remains virtually constant in the whole range of the weights of the analyzed samples. However, the errors of the mean values differ between the samples of weights less than 10 μg and more than 10 μg. For samples weighing from 10 to 30 μg, the error of measurements is $1\sigma \le 0.05$‰; for samples weighing less than 10 μg, it increases to 0.12‰. The increased influence of the blank experiment on the results of analyses can be a reason for the deterioration of the reproducibility of the results of samples of decreasing weight. The value of the blank experiment in our system was about 0.1% of the amount of the extracted gaseous CO_2 from carbonate samples weighing 30 μg. The isotopic composition of CO_2 in the blank experiment in our system was –16.4‰ and +5‰ for $\delta^{13}C$ and $\delta^{18}O$, respectively. If the blank experiment had made an appreciable contribution to the results of the analyses, the average value of the isotopic composition of $\delta^{13}C$ should regularly decrease with decreasing

weight. In our case, the average value virtually does not change; therefore, we can suppose that the deterioration of reproducibility is connected with the isotopic heterogeneity of the standard at analyzed weights less than 10 μg.

Thus, the proposed modification of the classical method of the extraction of CO_2 in combination with the up-to-date procedure of the measurement of micro amounts of gas in a constant flow of helium allows high-precision studies of the isotopic composition of samples weighing 10–30 μg. Using this procedure, we carried out in our laboratory about 1500 analyses of carbonates from deposits for half a year. Analyses of samples were alternated with analyses of the Coral-1 standard. Each series of analyses included 14 samples of carbonate and 3 samples of the Coral-1 standard weighing 30 μg. The statistics of the results of analyses of the Coral-1 standard for $\delta^{13}C$ and $\delta^{18}O$ from July until September 2008 is given in Fig. 4. According to the data in Fig. 4, the average values, –0.29 and –3.69‰ for $\delta^{13}C$ and $\delta^{18}O$, are calculated with a standard deviation of $1\sigma = 0.05$‰ for both results. It should be noted that there is a total scatter of $\delta^{13}C$ and $\delta^{18}O$ is 0.2‰; however, the scatters in each separate series of measurements seldom exceeded 0.1‰. Shifts between the series lead to an increase in the total scatter to 0.2‰. The reason for shifts between series has not been found yet.

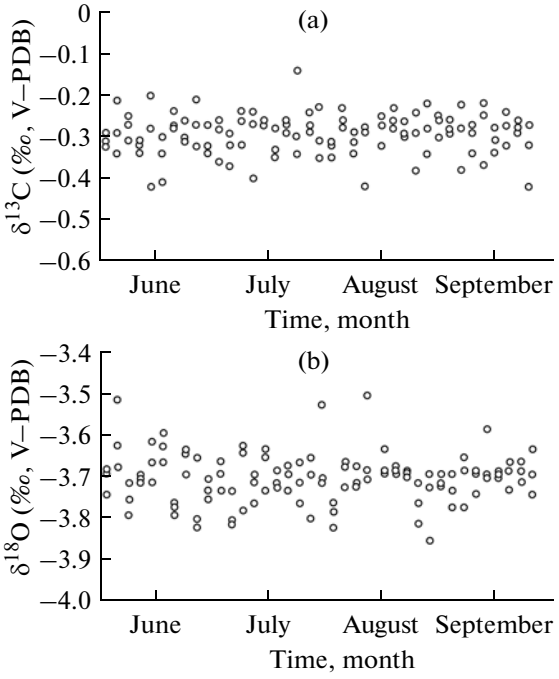

Fig. 4. Changes in values of $\delta^{13}C$ and $\delta^{18}O$ for the laboratory standard Coral-1 from June until September, 2008. The weight of analyzed samples was 20–30 μg.

The procedure proposed in this work was used to perform palaeotemperature studies based on the determination of $\delta^{18}O$ of foraminiferas. As an example of such studies, below we present the results of studying the isotopic composition of oxygen and carbon of planktonic and benthos foraminiferas for the reconstruction of palaeotemperature conditions of the Sea of Okhotsk for the last glacial and holocen periods with a resolution of 100–300 years. The object of the study was a column of LV 28-40-5 bottom deposits from the central part of the Sea of Okhotsk (the depth of the Sea is 1312 m and the length of the column is 801 cm). The deposits of the studied column formed over the last 80000 years. The age model of deposits was constructed on the basis of the results of measuring the carbon ^{14}C isotope by the method of accelerator mass spectrometry (AMS), isotope-oxygen chronostratiography, and regularities of the variation of magnetic susceptibility [16]. Samples of planktonic (*Neogloboquadrina pachyderma* s.) and benthos (*Uvigerina auberiana* and *Uvigerina parvacostata*) foraminiferas were selected from each 2.5 cm of the column length. The weight of the analyzed samples was 15–30 μg. Isotopic trends $\delta^{13}C$ and $\delta^{18}O$ were constructed based on the results of isotopic analyses (Fig. 5). The $\delta^{18}O$ trends for bentos and planktonic foraminiferas show virtually identical behavior in time; thus, the average values of $\delta^{18}O$ for benthos and planktonic foraminiferas differ on the average by 1.5‰ among themselves. The high reproducibility of the results of analysis by proposed method allows us to distinctly monitor the specific features of the behavior of the trends. The increase in the values of $\delta^{18}O$ for planktonic and benthos foraminiferas was observed in a range of 10000–40000 years in comparison to a range of 40000–80000 years. However, the values of $\delta^{18}O$ increase to different degrees and are 0.8‰ and 0.5‰, on the average, for planktonic and benthos foraminiferas, respectively. In going from the maximum of the last glacial period to the holocen, the values of $\delta^{18}O$ for planktonic and benthos foraminiferas decrease and, in the record of $\delta^{18}O$ for benthos forms, this decrease appears with a delay in time in the comparison with the planktonic form. The values of $\delta^{13}C$ for planktonic foraminiferas in the holocen time are 0.9‰ higher than in the glacial period, and the primary increase in the values of $\delta^{13}C$ for foraminiferas occurs simultaneously with the decrease in the isotopic composition of oxygen on the border of these periods. Individual nuances in the records of $\delta^{18}O$ and $\delta^{13}C$ for benthos and planktonic foraminiferas are revealed by high-precision isotopic measurements of $\delta^{18}O$ for samples selected from each 2.5-cm layer of deposits containing no more than 7–15 pieces of foraminiferas. The obtained records of $\delta^{18}O$ and $\delta^{13}C$ allow the reconstruction of climate changes over periods of a thousand years and less.

CONCLUSIONS

The development of a mass spectrometric procedure for measuring small amounts of gases in a con-

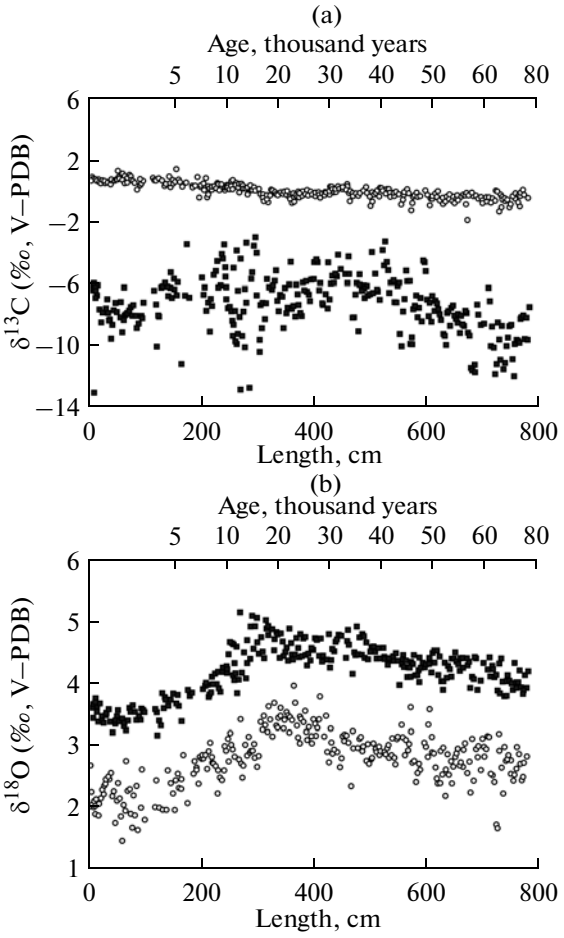

Fig. 5. Values of $\delta^{13}C$ and $\delta^{18}O$ for planktonic (*Neogloboquadrina pachyderma s.*) and benthos (and *Uvigerina parvacostata*) foraminiferas from bottom deposits of the LV 28-40-5 column. Benthos and planktonic foraminiferas are shown by black squares and white circles, respectively.

stant flow of helium favors the elaboration of a new procedure for sample preparation of small amounts of samples. In particular, the isotopic analysis of carbon and oxygen in carbonates is now performed with high reproducibility for samples weighing 100–50 μg. In this work, a technical modification of the traditional method of the decomposition of carbonates in phosphoric acid is proposed, which allows the analysis of samples of organogenic carbonate weighing 10–30 μg with reproducibility 0.05‰ for $\delta^{13}C$ and $\delta^{18}O$. The method combines the traditional system of CO_2 extraction from carbonates in a vacuum and the up-to-date procedure of measuring micro amounts of gas in a constant flow of helium. The system of CO_2 extraction has a number of technical modifications aimed at improving the reproducibility of results of analysis of microgram amounts of carbonates. In particular, the used feed-motion of samples provides the consecutive delivery of samples from sample holders to the acid. Such method of the feed-motion of samples has a number of advantages. Thus, the sample holder does not drop into the acid together with the sample, which prevents the pollution of the acid with substances from the surface of the sample holder; and the same portion of acid can be used for a very large number of analyses.

The combination of the classical method of vacuum extraction with the up-to-date procedure of measuring small amounts of gas in a constant flow of helium allowed the analysis of samples weighing 10–30 μg with reproducibility of 0.05‰ for $\delta^{13}C$ and $\delta^{18}O$. The accuracy and reproducibility of the $\delta^{13}C$ and $\delta^{18}O$ values was estimated by the results of measurements of international standards and verified by comparison of the results obtained by the proposed method at the microgram sample level with results of analyses by the traditional method at the milligram level.

Although the proposed technology in productivity ranks below the commercial automated devices for the preparation of carbonate samples, it reliably ensures the high reproducibility of the results of analysis of $\delta^{13}C$ and $\delta^{18}O$ for samples weighing 10 μg. This technology was used to study the isotopic composition of oxygen and carbon planktonic and benthos foramin-

iferas to reconstruct the palaeotemperature conditions of the Sea of Okhotsk.

REFERENCES

1. McCrea, J.M., *J. Chem. Phys.*, 1950, vol. 18, no. 6, p. 849.
2. Shackleton, N.J. and Opdyke, N.D., *Quat. Res*, 1973, vol. 3, no. 1, p. 39.
3. Matthews, R.K., Curry, W.B., Lohman, K.C., Sommer, M.A., and Poore, R.Z., *Nature*, 1980, vol. 283, no. 5747, p. 555.
4. Ignat'ev, A.V., *Cand. Sci. (Geol.-Mineral.) Dissertation,* Moscow, 1979.
5. Swart, P.K., Burns, S.J., and Leder, J.J., *Chem. Geol.*, 1991, vol. 86, no. 2, p. 89.
6. Matthews, D.E. and Hayes, J.M., *Anal. Chem.*, 1978, vol. 50, no. 11, p. 1465.
7. Werner, R.A. and Brand, W.A., *Rapid Commun. Mass Spectrom.*, 2001, vol. 15, no. 7, p. 501.
8. Revesz, K.R. and Landwehr, J.M., *Rapid Commun. Mass Spectrom.*, 2002, vol. 16, no. 22, p. 2102.
9. Paul, D. and Skrzypek, G., *Rapid Commun. Mass Spectrom.*, 2006, vol. 20, no. 13, p. 2033.
10. Spotl, C. and Vennemann, T.W., *Rapid Commun. Mass Spectrom.*, 2003, vol. 17, no. 10, p. 1004.
11. Nelson, S.T., *Rapid Commun. Mass Spectrom.*, 2000, vol. 14, no. 4, p. 293.
12. Paul, D. and Skrzypek, G., *Int. J. Mass Spectrom. Ion Proc.*, 2007, vol. 262, no. 3, p. 180.
13. Martineau, F., Fourel, F., Lecuyer, C., Toth, E., and Gorog, A., *Proc. JESIUM Conf.*, France, 2008, p. 38.
14. Ghosh, P., Patecki, M., Rothe, M., and Brand, W.A., *Rapid Commun. Mass Spectrom.,* 2005, vol. 19, no. 8, p. 1097.
15. Coplen, T.B., Brand, W.A., Gehre, M., Groning, M., Meijer, H.A.J., Toman, B., and Verkouteren, R.M., *Anal. Chem.*, 2006, vol. 78, no. 7, p. 2439.
16. Gorbarenko, S.A., Goldberg, E.L., Kashgarian, M., Velivetskaya, T.A., Zakharov, S.P., Pechnikov, V.S., Bosin, A.A., Psheneva, O.Y., and Ivanova, E.D., *J. Oceanography*, 2007, vol. 63, no. 4, p. 609.