Application of Multifrequency Bioelectrical Impedance Analysis Method for the Detection of Dehydration Status in Professional Divers

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Key words: *multifrequency bioelectrical impedance analysis; dehydration; diving; divers; heliox.*

Summary. *Background and Objective. The level of dehydration has been known to be a predisposing factor for the development of decompression sickness in divers. The aim of this study was to determine the level of dehydration in divers who dove with heliox and to determine whether the source of this dehydration was intracellular and/or extracellular by means of multifrequency bioelectrical impedance analysis (MF-BIA).*

Material and Methods. Eleven male professional divers were enrolled in the study. In order to determine the level of dehydration, MF-BIA was carried out (at 5, 50, and 100 kHz) and capillary hematocrit (Hct) was measured two times: one before diving and the other after leaving the pressure room.

Results. When prediving and postdiving parameters were compared, significant increases in the resistance at 5 kHz ($P<0.001$), 50 kHz, ($P<0.001$), and 100 kHz ($P<0.01$) and Hct ($P<0.01$) were observed after the diving. Similarly, a statistically significant fluid shift was found: total body water, –1.30 L ($P<0.001$), extracellular water, –0.85 L ($P<0.001$); and intracellular water, –0.45 L ($P=0.011$).

Conclusions. Our results showed that mild dehydration occurred both in the intracellular and extracellular compartments in divers after deep diving. This study also indicates that MF-BIA could be a reliable new method for determining the dehydration status in divers.

Introduction

Dehydration is known to develop both in diving and under pressure conditions in divers. This effect is greater in cold water. Colder conditions stimulate peripheral vasoconstriction, which is followed by cold diuresis (1–3). Divers can also lose fluids through the skin, sweat, vomiting, or diarrhea. However, breathing causes the most loss of water in divers (4). When a diver is dehydrated, there is a reduced blood volume in the body as a decrease in plasma volume causes increases in hemoglobin, hematocrit (Hct), and serum proteins. A reduction in blood volume may affect blood viscosity and the dynamics of blood flow (1, 4–7). Moreover, some authors argue that hemoconcentration resulting from postdiving dehydration could also precede and promote the development of decompression sickness (DCS) by reducing a tissue blood perfusion rate and finally by elimination of the inert gases (particularly nitrogen) (8–14).

In addition, dehydration and hypothermia can adversely affect divers in other ways. Since there is a reduced volume of blood circulating through the body, it tends to lower blood pressure. Cardiac output is decreased because of bradycardia. The heart has to work harder and pump faster in order to meet the body's demands. This leads to decreased endurance and increased fatigue, which diminish the diving performance (4, 15–17). Therefore, it is very important to maintain fluid balance in divers.

Currently, it is well known that dehydration may increase the risk of DCS in divers (12, 15, 18). However, the results of animal studies are conflicting. Fahlman and Dromsky (19) studied two groups of pigs; 31 were hydrated and 26 were dehydrated. The hydration status at the time of decompression significantly influenced the incidence and time to the onset of DCS. Dehydration significantly increased the overall risk of severe DCS and death in this model. However, the results of experiments in another animal study on rats by Skogland et al. (20) did not support the idea that dehydration increased circulatory venous gas emboli and DCS.

Hct, however, is an estimation of intravascular hydration only and does not differ between fluid loss from the body and fluid shift between compartments. Being a ratio, its value reflects changes both in red cell mass and plasma volume (21). Hct values have been frequently used as the indicators of dehydration levels in divers. However, the results obtained in these studies are contradictory. Moreover, there is no consensus concerning the compartment from which fluid loss occurs during diving (12, 21).

Nowadays, the bioelectrical impedance analysis (BIA) method is widely used in many clinics to

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measure body compartments in sports medicine and weight loss programs. There are also numerous clinical studies on the use, efficacy, and reliability of the BIA method in estimating body composition in acute fluid loss (22–24). Lukaski (25) summarized methods for the determination of total body water (TBW), which is calculated as follows: TBW = extracellular water (ECW) + intracellular water (ICW). Thus, these studies have indicated that BIA is a very good compromise between cost, ease of operation, and reliability.

Two types of BIA systems are currently available in medical research.

Single-frequency BIA (SF-BIA) generally at 50 kHz is passed between surface electrodes placed on the hand and foot. Some BIA instruments use the electrodes of other locations, such as foot-to-foot (26, 27) or hand-to-hand. At 50 kHz, BIA does not measure TBW, but a weighted sum of ECW and ICW resistivity (~25%). SF-BIA permits us to estimate fat-free mass (FFM) and TBW, but cannot determine the differences in ICW. BIA results are based on a mixture of theories and empirical equations (28).

Multifrequency BIA (MF-BIA) is similar to SF-BIA; MF-BIA uses empirical linear regression models, but includes impedances at multiple frequencies. MF-BIA utilizes different frequencies (0, 1, 5, 50, 100, and 200–500 kHz) to evaluate FFM, TBW, ICW, and ECW. At the frequencies of less than 5 kHz and more than 200 kHz, poor reproducibility has been noted especially for reactance at low frequencies (19).

According to Patel et al. (29), MF-BIA is more accurate and provides less biased estimates than SF-BIA for the prediction of ECW, whereas SF-BIA as compared with MF-BIA is more accurate and provides less biased estimates for TBW in critically ill subjects. Hannan et al. (30) noted that the application of MF-BIA as compared with bioelectrical spectroscopy (BIS) led to better prediction of TBW and same prediction of ECW in surgical patients. Utter and Lambeth (31) demonstrated that MF-BIA was able to detect changes in the body composition of wrestlers.

The aim of the present study was to determine whether dehydration occurred during diving performed with heliox and, if dehydration occurred, to ascertain whether it was intracellular and/or extracellular by means of the MF-BIA method.

Material and Methods

The study was conducted in accordance with the Declaration of Helsinki and was approved by the Ethics Committee of the Istanbul Faculty of Medicine at Istanbul University. Informed consents were provided by participants before the study.

Participants. In this study, 11 male volunteers with a mean age of 30.1 years (SD, 3.4) were enrolled. The participants were healthy professional divers who worked in the undersea construction of Duzce/Akcakoca Natural Gas Piping System and used surface-supplied heliox to prevent nitrogen narcosis. Heliox is a mixture of 79% helium and 21% oxygen and is often used for commercial deep diving. The divers were first screened by physical examination. Divers whose a history of diving showed previous DCS and/or arterial gas embolism were excluded from the study, as they could be susceptible to DCS, which may affect the MF-BIA results.

Hematocrit Measurement. In order to determine the level of capillary Hct, the divers were examined two times: one before diving and the other after leaving the hyperbaric chamber. Capillary blood samples were taken from the fingertip of divers by Na-heparinized hematocrit capillary tubes. The tubes were centrifuged (Hettich-Hematocrit 27, D-78532, Tuttlingen, Germany) for 5 minutes at 10 000 rpm before Hct estimation on a standard visual plate reader.

Diving Procedure. Water temperature during the study period was 16°C±2°C at the surface, falling to 10°C±2°C over the depths at which the divers were active. Although the participants were wearing dry suits with protective clothing and underwear to decrease heat loss, hypothermia may also be caused by body heat loss from breathing and bare hands in longer heliox dives. The divers included in this study did not use any hot-water suits, and the breathing gas was not heated. The divers performed dives using MK37-type diving gear with surface-supplied heliox. During the dives, the divers performed the regular pipe installing job-requiring moderate-to-heavy workloads. The U.S. Navy Standard Surface Supplied Mix Gas Diving Decompression Tables were used during the dives (Fig. 1). The divers dived only once a day and took rest for a minimum of 18 hours before the next dive. MF-BIA measurements were performed within an hour before diving and within 10 minutes after leaving the decompression chamber. All divers were asked to adhere to their standard diet supplied on the vessel. Additionally, they were instructed to refrain from strenuous exercise, caffeine consumption, and alcoholic beverages, and medications were prohibited during the total dive period. In addition, each diver emptied his bladder before and after the dives. The temperature of extremities was also controlled to have the normal (34.0°C±1.0°C) temperature by means of a Braun ThermoScan-IRT 4020 digital thermometer (Braun GmbH, Kronberg, Germany).

Conditions of Multifrequency Bioelectrical Impedance Analysis. The Nutriguard M Phase Sensitive Multifrequency Impedance Analyzer (Data-Input GmbH, Darmstadt, Germany), utilizing the sinusoi-

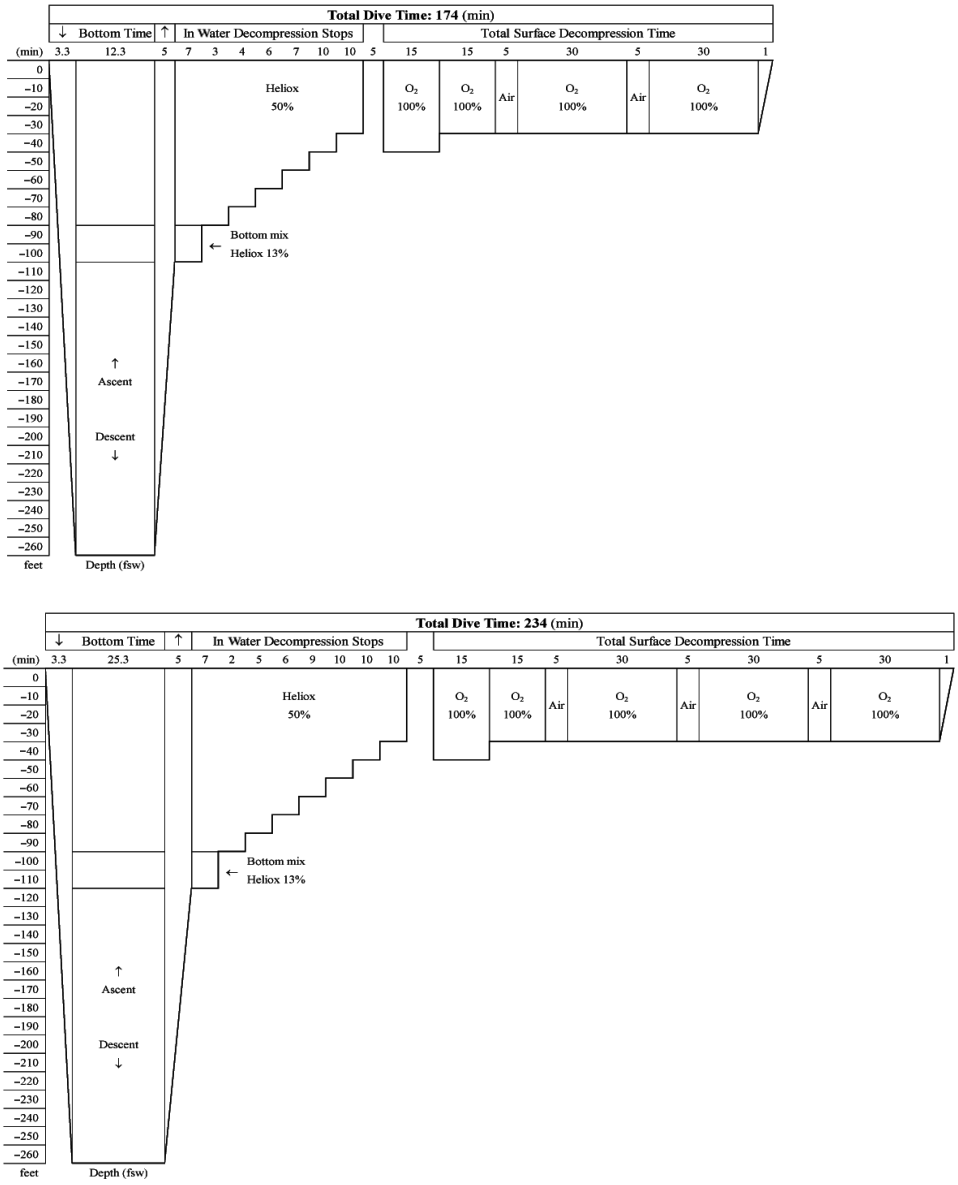


Fig. 1. Decompression tables (range of total dive time, 174 and 234 minutes)

dal frequencies of 5, 50, and 100 kHz, was used to measure the distribution of ECW and ICW. The divers were instructed to remove all metals from their clothing and body before the measurements, and were kept lying in a relaxed horizontal position on a couch. In the MF-BIA measurements, the contact between the legs or arms and the trunk shortens the path of the current and leads to highly falsified results. Therefore, the legs of the divers were kept apart at an approximate angle of 45° so that the thighs did not touch each other. Similarly, the arms were kept apart at an approximate angle of 30° from the median plane to avoid any body contact. All the extremities, hands and feet, were positioned to be at the same height as the body during measurements, because if the extremities are not positioned at the same level (affecting distribution of the fluid content unfavorably), then falsified results may be obtained (Figs. 2A–C).

All the subjects were right handed. For standardization reasons, the measurements were always carried out on the dominant side (right hand and right foot). The right hand and the right foot were unclothed for measurements.

Electrode Placements. Adhesive Ag/AgCl Bianostic Classic® electrodes (Data-Input GmbH, Darmstadt, Germany) were used for tetrapolar and ipsilateral measurements. It is very important that the electrodes are positioned accurately. The precise placement of the electrodes is crucial for ac-

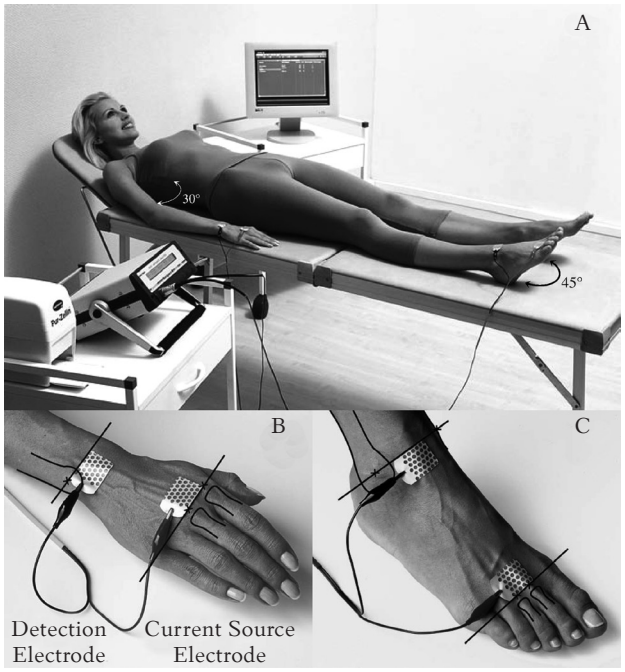


Fig. 2. MF-BIA measurement station (A) and hand (B) and foot electrode placement (C)

Table 1. Demographic Characteristics and Diving History of Divers (n=11)

Characteristic	Value
Age, mean (SD), year	30.1 (3.4)
Body weight, mean (SD), kg	80.8 (8.6)
Body height, mean (SD), cm	175.2 (5.8)
Body mass index, mean (SD), kg/m^2	26.3 (2.4)
Diving experience, range, years	7–10
Average depth, mean (SD), meters of sea water	77.2 (1.3)
Bottom time, mean (SD), min	20.5 (4.2)
In water decompression stops, range, min	47–59
Total surface decompression time, range, min	101–136
Total dive time*, range, min	174–234
No. of oxygen periods†, range	2–4
MF-BIA measurement prediving‡, mean (SD), min	31.4 (9.3)
MF-BIA measurement postdiving§, mean (SD), min	5.16 (1.95)

*Total dive time (surface-to-surface) = bottom time + in water decompression stops + total surface decompression time.
†The number of oxygen periods comprised 30 min each, in which divers inhaled 100% oxygen in the decompression chamber with 5-minute air breaks in between.
‡MF-BIA measurements of divers were performed before leaving from the surface.
§MF-BIA measurements of divers were performed after coming out of the decompression chamber.

curate measurements. For MF-BIA measurements of body composition, two electrodes were attached to the hand and two to the foot. The voltage was fed in through the two outer electrodes at the hand and foot. The resistance was measured over the two interior electrodes (Figs. 2A–C).

Statistical Analysis. The NutriPlus© (Data-Input GmbH, Darmstadt, Germany) software was used for the analysis of body composition in divers. The SPSS version 16.0 was used for statistical analysis. Means and standard deviations of the data were calculated, and their suitability for normal distribution was confirmed using the Kolmogorov-Smirnov test. Comparison of prediving and postdiving data of the groups was performed by the nonparametric Wilcoxon test, and $P<0.05$ was considered the level of statistical significance.

Results

The subjects included in the study were 11 male professional divers with a mean age of 30.1 years (SD, 3.4), mean weight of 80.8 kg (SD, 8.6), mean height of 175.2 cm (SD, 5.8), and mean body mass index (BMI) of 26.3 kg/m^2 (SD, 2.4). The demographic characteristics and diving history of the divers are presented in Table 1. The data of the present study were obtained from 26 dives performed by the 11 divers. In our study, 6 divers dived 3 times, 3 divers dived 2 times, and 2 divers dived 1 time.

MF-BIA, utilizing the frequencies between 5 and 50 and 100 kHz, is used to measure TBW and the distribution of ECW and ICW. The level of deflection on or penetration into the tissue cell membrane is dependent on which ranges of MF-BIA frequencies are used. For example, at 5 kHz, no penetration into the tissue cell membrane occurs (full deflection onto the cell membrane and measurement of ECW); at 50 kHz, slight deflection onto the tissue cell membrane (measurement of ICW and body cell mass [BCM]), and at 100 kHz, no deflection to the tissue cell membrane (full penetration into the cell and measurement of TBW).

In the first stage of our study, resistances before and after diving as the indicators of dehydration were compared. A significant increase in the resistances at 5 kHz ($P<0.001$), 50 kHz ($P<0.001$), and 100 kHz ($P<0.01$) was recorded after diving (Table 2).

It is generally agreed that the reference limits of TBW range from 50% to 60% of total body weight for men and from 55% to 65% for women. The distribution of TBW into ECW and ICW were 43% (lymph, interstitial, transcellular, plasma) and 57%, respectively. Significant differences in the TBW, ECW, and ICW levels comparing the values before and after diving were documented: –1.30 L for TBW ($P<0.001$), –0.85 L for ECW ($P<0.001$), and –0.45 L for ICW ($P=0.011$) (Table 2).

Increased Hct is a commonly used proxy measure for dehydration. Any decrease in the volume of plasma causes an increase in Hct, despite an increased red blood cell count (32). For divers with dehydration during postdiving, fluid lost from the intravascular space makes the blood more concentrated with possible increases in Hct. Hct levels before and after diving for each diver were determined, and an increased Hct level by 1.9% after diving was recorded ($P<0.01$) (Table 2).

Table 2. Comparison of Prediving and Postdiving Variables in 11 Divers (n=11)

Variable	Prediving	Postdiving	*z*	*P**	Fluid Shift
Resistance at 5 kHz, W	559.4 (62.6)	586.9 (65.2)	3.825	<0.001	ECW
Resistance at 50 kHz, W	471.2 (54.1)	490.2 (54.9)	3.825	<0.001	ICW+BCM
Resistance at 100 kHz, W	444.7 (48.1)	457.8 (51.9)	3.059	<0.01	TBW
TBW, L	46.2 (4.0)	44.8 (3.8)	−3.750	<0.001	−1.30
ECW, L	18.4 (2.5)	17.9 (2.2)	−3.500	<0.001	−0.85
ICW, L	27.3 (1.7)	26.9 (1.5)	−2.530	0.011	−0.45
Hct, %	45.8 (1.3)	47.7 (1.2)	4.062	<0.01	+1.9

Values are mean (standard deviation).
TBW, total body water; ECW, extracellular water; ICW, intracellular water; Hct, hematocrit.
*Wilcoxon test.

In the second stage of our study, 6 divers who dived sequentially 3 times were the most affected, and their fluid shifts were recorded after each period. Results of the first and second stages were similar. All results were independent from the average depth (msw) and bottom time (min). After each period, there was a marked loss in TBW (P=0.024), ECW (P=0.025), and ICW (P=0.036). In line with all these findings, there was a relevant linear marked increase in the resistance at 5 kHz, (P=0.020), 50 kHz (P=0.018), and 100 kHz (P=0.032) after diving (Fig. 3A). However, the increases in Hct were not as pronounced as the increases in the resistance at all frequencies. There was a slight increase in Hct comparing the values before and after diving (P=0.045) (Fig. 3B).

Discussion

Adequate hydration is crucial in maintaining optimal physiological functioning, and the need for the fast and reliable assessment of dehydration status in diving and hyperbaric medicine research has become increasingly important in divers (15, 19, 33). Although there are many methods in literature, which have been used to determine fluid loss during dives, the commonly used method includes monitoring of the changes in Hct levels.

In a recent study conducted on scuba (self-contained underwater breathing apparatus) divers who dive into tropical waters, a significant increase in Hct levels with an increase in the diving period was demonstrated, and a significant, but weak, correlation was demonstrated between diving depth and Hct levels. However, because of the error rates in the Hct measurement techniques, it was suggested that these outcomes are not reliable enough for fluid replacement protocols to be applied to divers who experience decompression (12, 21). These conflicting outcomes, which were determined between the level of dehydration and the change in Hct levels, have prompted studies that aimed to determine the degree of hydration, which would occur in biological systems under various conditions with the use of more reliable techniques. In this regard, the BIA method that is an economic, portable, accurate, reliable, and noninvasive method for measuring body composition has gained importance.

Although there are many studies on the level of dehydration and Hct postdiving in divers (12, 34, 35), to the best of our knowledge, this is the first study that has employed the MF-BIA method for the determination of dehydration level in divers. Newton et al. (12), Blanc et al. (34), and Boussuges et al. (35) reported that hemoconcentration with the Hct values of 48% or greater increased the risk of neurological deficits in recreational scuba divers with DCS. Our study showed that the results of the first and second stages were similar; the postdiving Hct values of the professional divers were approximately 48% (47.7% [SD, 1.2%] and 48.4% [SD, 0.9%], respectively) (Table 2 and Fig. 3B). Therefore, in line with these results, we may suggest that professional divers who participated in our study could also have a risk of DCS.

Although an increase in the Hct level shows especially a loss of intravascular fluid, it does not give information about losses of ECW and ICW. Dehydration in divers can be shown by both methods. However, we observed that the MF-BIA method was a more accurate and reliable way to measure dehydration in divers than Hct.

The meta-analysis by Martinoli et al. (36) concluded that SF-BIA and BIS significantly overestimated TBW in healthy individuals, whereas there was no overestimation in case of MF-BIA. MF-BIA seems to be a more accurate method for determining the TBW compartment in healthy and obese adults and in persons with chronic renal failure. In the MF-BIA method, the frequency intervals of 0, 1, 5, 50, 100, and 200–500 kHz are used. Previous studies have shown that the repeatability rate of MF-BIA analyses performed at a frequency of less than 5 kHz and more than 200 kHz is quite low (37, 38). Considering these findings, the measurements at the frequencies of 5, 50, and 100 kHz were performed. Evaluation of the outcomes of the present study demonstrated that mild dehydration developed subsequent to deep dives performed by professional divers using heliox.

Our results concerning MF-BIA showed that dehydration occurred both in the intracellular and extracellular compartments in divers after deep dives. Putten (18) reported in his study that body fluid

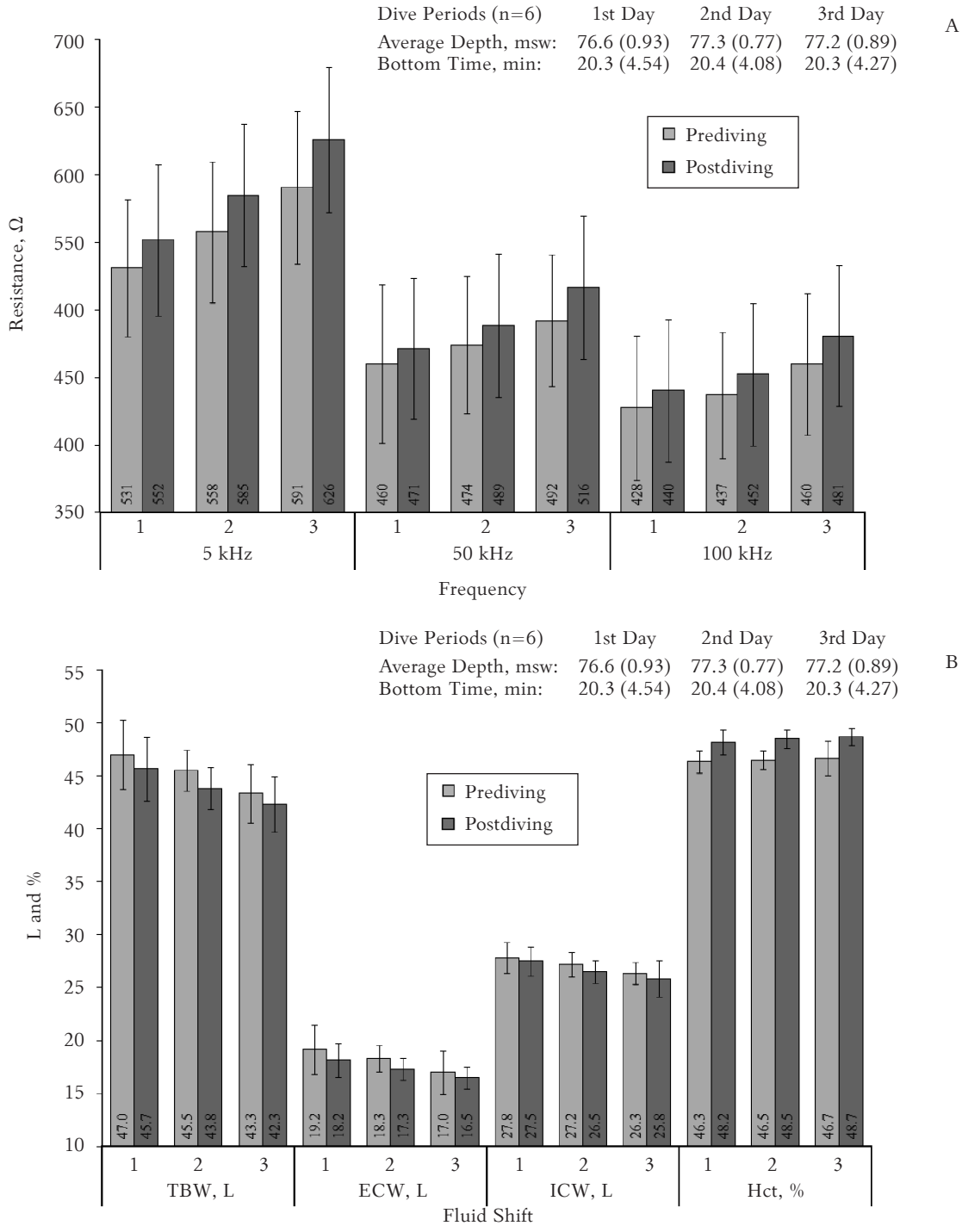


Fig. 3. Changes in the resistance at 5, 50, and 100 kHz (A), total body water (TBW), extracellular water (ECW), intracellular water (ICW), and hematocrit (Hct) (B) in 6 divers (1st, 2nd, and 3rd days)

losses in caisson workers were mainly from the intracellular compartment, which is similar to our BIA results. On the other hand, Leni et al. (39) showed that pre-existing extracellular dehydration induced functional motor spinal cord injury after severe decompression in rabbits.

Furthermore, it was surprising to note a significant dehydration for a total dive time (range) of 174 and 234 minutes, although the mean time spent at the bottom was as short as 20.5 minutes (SD, 4.2; range, 12.3–25.3). Our results indicate that the acute phase of mild dehydration developed in the divers, and it is not recovered easily despite the long surface decompression time (Table 2, Figs. 3A and B).

In the second stage of our study, it was observed that dehydration increased more and did not com-

pletely return to normal level, but it became worse during further dehydration analyses performed on divers who dived for 3 sequential days (Figs. 3A and B). Marked increases were detected at all frequencies in MF-BIA due to dehydration; thus, the prominent decreases in TBW, ICW, and ECW were also detected.

Fluid shifts from the intravascular compartment to the extravascular space could also potentially cause an increased Hct in divers (21). Experimental studies have shown that the Hct level increases during decompression in the first hour and decreases slowly to normal values after a 24-hour delay (40). Determination of mild increases in Hct related to intravascular plasma volume loss has indicated that MF-BIA is a more sensitive and reliable method than the Hct measurements especially in the detection of dehydration during repetitive dives.

This indicates that special attention should be paid to fluid balance during longer heliox dives. Therefore, before diving, the divers should abstain from activities that would lead to dehydration. They should also avoid caffeine consumption and alcohol intake, and should not dive in cases of diarrhea and vomiting. Accurate hydration of divers should be ensured after dives. It is known that a 1-day break at least particularly after the sequential 4-day diving with heliox for protection against pulmonary oxygen toxicity may be essential for recovery from dehydration (33).

Conclusions

Our results showed that dehydration occurred both in the intracellular and extracellular compartments in divers after deep dives. The results of this study also indicate that MF-BIA could be a reliable method for determining the dehydration status in divers. By carrying out similar studies on heliox, air dives, and comparing outcomes of the present study, one can better understand dehydration, which could occur after diving, as well as determine the fluid replacement method that would be required to minimize the risk of decompression sickness resulting from dehydration.

Acknowledgments

The authors would like to thank Evin Isgor, MD (Freelance Medical Writer Istanbul/Turkey), for the editorial supervision and Julius Epie, MD, for the language editing contributions to the manuscript. The authors also thank Kornelia Henne (Data-Input GmbH, Darmstadt, Germany) for BIA images and documents.

Statement of Conflict of Interest

The authors state no conflict of interest.

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Received 26 October 2011, accepted 16 April 2012