ABSTRACT

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Repeated body composition analyses were performed on 30 male volunteers aged 18-25 yrs. The Ss were measured both before and after hydration by hydrostatic weighing (HW) and bioelectrical impedance analysis (BIA). Validity measures of the BIA were examined relative to HW. Sensitivity of the BIA was measured in response to an overhydration volume of 1.5% of body weight in tapwater. The validity of the BIA for % BF was found to be sig ($p < 0.05$) overestimated (2.6 - 3.1%). Neither the BIA nor HW were sensitive to overhydration as they were not found to be sig ($p < 0.05$) for % BF or LBM. Future studies could include larger fluid volumes and, a larger, more, homogenous sample in an effort to statistically emphasize the physiological effect of hydration and the BIA's potential ability to detect such changes.

Validity and Sensitivity Measures of the BIA in Response to Overhydration

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Human Performance: Exercise Physiology

by

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CHAPTER I

INTRODUCTION

The assessment of human body composition continues as a topic of popular interest. Bioelectrical impedance analysis, a relatively new technique, is presently receiving a great share of this attention. Currently, the bioelectrical impedance analysis (BIA) is under widespread scrutiny due to its potential as a practical, noninvasive measurement technique. The potential use of this procedure exists not only for sports and fitness purposes, but also for the medical and nutritional domains.

The concept behind the BIA is based on the potential electrical conductivity of the body. Electrical conductance is known to be enhanced by body fluid volumes and electrolyte concentrations (Lukaski, Bolonchuk, Hall, & Sider, 1986). Knowing that lean body mass (LBM), or more accurately fat free weight (FFW), contains greater amounts of both water and electrolytes than body fat (Presta, Segal, Gutin, Harrison, & Van Itallie, 1983), it follows that FFW has greater electrical conductivity than body fat. With greater conductivity then, FFW is measured by the BIA as creating less impedance (or resistance), therefore signifying a lower percentage of body fat in the final body composition assessment. The actual measure of the BIA is of both total body conductance, as well as resistance, with the desired response showing that the conductance of FFW is greater than that of fat mass (Segal, Gutin, Presta, Wang, & Van Itallie, 1985).

Since there is no exact method for determining body composition other than direct dissection, some techniques are based, in part, on estimated total body water content. Loeppky, Myhre, Venters, and Luft (1977) measured the water content of LBM in a research group of 35 men to be 73.5%, and 73.2% through mammalian dissection. The BIA (BIA-103, RJL Systems, Detroit, Michigan) uses the latter value in its formula for determining LBM, and thus percent of body fat. Total body water (TBW) is a measure which may fluctuate for any one individual over time, and may almost certainly vary between individuals (Girandola, Wiswell, & Romero, 1977). For practical, noninvasive purposes, however, the assumption is made that this value must be considered a constant.

The attempt to determine total body water based on LBM and its ability to allow electrical conduction, however, is also dependent upon other factors. The principle behind the BIA assumes that both the electrical signal and the configuration of the system involved, namely the human body, are also constant. The potential electrical impedance depends on the length of the conducting system and its cross-section, or conducting volume (Hoffer, Meador, & Simpson, 1969). It is not entirely clear how influential the variability of body size and dimension can be, but the assumption that all human body geometrics are comparable may, in fact, prove to be erroneous. Research continues in attempt to refine these types of problems.

Need for the Study

As with all measurement devices, and especially those involved with measuring biological parameters, a degree of error is inevitable. It was the purpose of this study to attempt to gather more data regarding the sensitivity of this body composition technique. Due to the relative infancy of the BIA, the majority of research efforts to date have concentrated primarily on validation of the technique and refinement of its underlying equations, rather than measures of sensitivity.

Statement of the Problem

The purposes of this study were to: 1) compare the related baseline and post fluid ingestion measures of hydrostatic weighing (HW) with baseline and post fluid ingestion measures of the BIA in attempt to examine the validity of the BIA's measurements; and, 2) to examine the sensitivity of the BIA to an increase in total body water from the ingestion of a volume of fluid which represented 1.5% of each subject's total body weight.

Hypotheses

The major hypotheses of this study were:

1. The validity measures of lean body mass (LBM) and percent body fat (% BF) as obtained by bioelectrical impedance analysis will not vary significantly from those measured by hydrostatic weighing.

2. The ingestion volume of tapwater representing 1.5% of a subject's total body weight will not create a significant change in the percent of body fat as measured from baseline to post ingestion by the BIA technique.

Assumptions

The following assumptions were relative to this study:

1. The HW technique is a valid and reliable reference technique.
2. Test procedure administration is consistent.
3. The subjects performed the residual volume (RV) and HW procedures to the best of their abilities.
4. All subjects adhered to the suggested pre-test guidelines.
5. All subjects were in good health during their testing period.
6. The fluid ingestion volume representing 1.5% of total body weight was a standard measure for all subjects.
7. The delay time of 50 minutes following the fluid ingestion period of 15 minutes represented a standardized absorption time among subjects, and between test phases.
8. The marking of electrode placement sites with permanent ink reduced variability of electrode placement between testing phases.

Limitations/Delimitations

1. Subjects were male college age volunteers available from the University of Wisconsin-La Crosse.
2. Exact assessment of total body water (TBW) was beyond the scope of this study.
3. Variabilty of body fat percentage among subjects did not represent or include those possible in more extreme samples.

Definition of Terms

Selected terms as interpreted and applied in the content of this study are as follows:

Bioelectrical Impedance Analysis (BIA) - method of percent fat determination based on total body resistance measured with a four electrode arrangement that introduced a painless electrical current of 800μA at 50 kHz. Resistance read at 0-1,000Ω and reactance read at 0-200Ω (RJL BIA-103, RJL Systems, Inc. Detroit, Michigan).

Hydrostatic Weighing (HW) - reference method for determining body density based on the Archimedes principle with residual volume (RV) accounted for. Percent body fat was calculated from body density as established by Brozek, Grande, Anderson, and Keys (1963).

Archimedes Principle - a solid body immersed in a liquid is bouyed up by a force equal to the weight of the liquid displaced.

Residual Volume (RV) - the volume of air remaining in the lungs following maximal forced expiration. This volume represents a percent of total lung capacity and is measured in liters of air.

Vital Capacity - the maximal volume of gas that can be expelled from the lungs following a maximal inspiration.

Dry Weight - body weight (measured to nearest 0.25 lb. and converted to kg.), attained prior to HW, measured in swim attire only.

Fluid Ingestion - a specified amount of tapwater measured in mililiters based on 1.5% of total body weight as represented by daily dry weight (lbs. converted to kgs.). The water was chilled to 0-9° C.

Fat Free Weight (FFW) - body weight representing muscle mass, bone, water, connective tissue, organ and other lean body mass tissues, exclusive of nonessential fat.

Nonesssential Fat - lipid storage found primarily within adipose tissue.

Essential Fat - includes lipids incorporated into the organs and tissues such as nerves, brain, heart, liver, and mammary glands.

CHAPTER II

REVIEW OF RELATED LITERATURE

Bioelectrical Impedance Analysis

Early research with the BIA concentrated on the establishment of valid measurement parameters. Hoffer et al. (1969) demonstrated the relationship of total body water (TBW) to total body weight through measurements recorded by the bioelectrical impedance analysis method. Their recognition of the inherent problems in accepting the involved assumptions, led them to examine various other equations. The square of body height divided by resistance (T^2/Z) demonstrated the best relationship with total body water (TBW) for determining percent of body fat (% BF). This was supported by a correlation coefficient (r) of 0.92 (Hoffer et al., 1969). Two areas of concern, however, are associated with this equation, as well. First, as it is not practical to attempt to measure fluid distribution throughout the body, its uniformity remains unknown. Second, the inability to accurately, yet non-invasively, measure the volume of a human body persists. These two factors, although recognized as potential sources of inaccuracy, are included in the agreed assumptions.

The thrust of much of the BIA research has thus confronted the question of validity and reliability in regards to its ability to determine percent of body fat based on the estimation of TBW. Again, as all *in vivo* estimates of human body composition are in fact, indirect, absolute accuracy is improbable. Several known standards of measure, however, are accepted for the purpose of comparison. One such technique is the measure of total body potassium (TBK) determined via radioisotope tracing. Lukaski, Johnson, Bolonchuk, and Lykken (1985) compared the validity of the BIA to total body potassium (TBK) in estimating total body water (TBW). Their findings suggested an average precision variation (validity) of the impedance method to be 2.0% (range of 0.9 - 3.4%) while reliability testing indicated a correlational coefficient of 0.99 (Lukaski et al., 1985).

In 1986, Lukaski and associates, expanded upon their earlier work (Lukaski et al., 1985) by further testing BIA validity. In this study, LBM, indirectly measured from anthropometric and skinfold measurements, was compared with those measures from the BIA and HW. Their findings again supported the concept that conductance was a valid and reliable predictor of LBM not only in the assessment of male subjects, but also for the assessment of female subjects (Lukaski et al., 1986). Specifically, Lukaski et al. (1986) recorded the BIA standard error of estimate (SEE - 2.7%) for calculating percent body fat (% BF) to be lower than that of standard anthropometric techniques (SEE - 3.9%).

Although the BIA offers promise, results most often indicate the need for more research, or more accurate equations for measurement. Segal et al. (1985) compared densitometry, TBW, TBK, anthropometry, total body electrical conductance (TOBEC), and BIA. Their results for the BIA demonstrated its promise as an evaluative tool, but more importantly, their research indicated its weaknesses in measuring LBM as the percent of body fat increased. Their results suggested that the BIA systematically overestimated LBM for extreme samples on the body composition continuum (Segal et al., 1985). Numerous studies support this pattern of overestimating the LBM percentage of the overall body composition analysis.

Miranda, Lombardi, Troxel, and Menon (1986) indicated that the BIA significantly overestimated percent body fat for females by an average of 2.6%, while it simultaneously underpredicted percent body fat for males by an average of 2.1%. Renk, Breitlow, and Sander (1986) also suggested that the BIA provided different testing reliability for male versus female subjects. Additionally, Keller and Katch (1986) reported overprediction of % BF for females, and although to a lesser degree, underprediction of % BF for males, as well. Keller and Katch (1986) further examined underfat, normal fat, and overfat subjects in attempt to gain information on the sensitivity of the BIA. Repeated error in prediction of percent body fat led them to report only moderate validity for the BIA (Keller & Katch, 1986). Furthermore, Miles and Stevens (1986) also obtained questionable validity with the BIA, as their studies likewise demonstrated its insensitivity at the extremes of the body composition range.

In further examination of the sensitivity of the BIA, recent research has begun to include a multitude of additional potentially influential parameters. Katch, Keller, and Solomon (1986), for example, indicated that the BIA underpredicts percent body fat in cardiac and pulmonary patients, while overpredicting for both black male and female subjects. Furthermore, Lukaski and Bolonchuk (1986) examined the sensitivity and validity of the BIA to athletes in relation to various states of hydration, exercise, and food consumption. Their conclusions implicated the need for controlled conditions among subjects in order for valid responses with the BIA when estimating both fat free mass and percent body fat (Lukaski & Bolonchuk, 1986). Shore and Taylor (1986) prohibited the use of caffeine, alcohol, smoking, exercise, and food for five hours prior to BIA measurement, yet found significant overestimates of percent body fat for their young adult male subjects. Finally, Lawlor, Crisman, and Hogdon (1986) after comparing HW, BIA, and body circumference measures, controlled for the factor of age, gaining only a moderate improvement for males and no significant changes in the correlation coefficient for females (.779 to .826, and .728 to .815, respectively). They concluded that their relative measures of correlation for body circumferences (males = .881, females = .815) were more precise in predicting percent body fat than was the BIA (Lawlor et al., 1986).

In summation, it is clear that, although promising, the BIA is in need of continued research. Most important is the development of valid measurement for the multitude of variables which are present in non-steady state and untraditional subject situations. Among these variables are the effects of overhydration, dehydration, and the accompanying electrolyte changes. This study primarily examined the associated influences of overhydration on the body composition analysis as established by the BIA.

At present, minimal data exist which specifically measure any of these effects. It is, however, recognized that hydration factors do play an important role in establishing improved regression equations for the measures at the extremes of the body composition scale for the BIA (Shore & Taylor, 1986; Cohn, 1985; Nash, 1985).

Hydrostatic Weighing

In attempting to measure the accuracy and sensitivity of the BIA in determining lean body mass and percent of body fat, a reputable reference technique was needed. Hydrostatic weighing (HW) or underwater weighing as it is commonly known, is currently accepted as the most reliable method for the estimation of body composition (Mendez & Lukaski, 1981). Using the concept defined by the Archimedes Principle, body volume and an estimated body density are then calculated from an individual's underwater weight.

The University of Wisconsin-La Crosse's Human Performance Laboratory follows the densitometric values established by Brozek, Grande, Anderson, and Keys, 1963. These figures are based on the body composition analysis of actual cadaver dissection. Brozek and his associates (1963) established a "reference body" based on such analysis of three male cadavers. Their prediction equation is based on two components: the density of this reference body to be 1.064, and the density of what has been labled as "obesity tissue" to be 0.938 (Brozek et al., 1963). From these standardized densitometric values, percent of body fat can be estimated from various equations. Unfortunately, however, even research as detailed as that of Brozek et al. (1963), suffers from numerous undefined parameters.

In order to proceed with body composition analysis, one must accept several assumptions throughout the entire process. With densitometric values calculated as best supported by research, the potential still exists for two sources of error to interfere with accurate body composition measurements. Katch and Katch (1980) described these potential sources of error as: biological variation and experimental error. They explain that "biological variation" includes variances in total body water, bone density, the density of adipose tissue itself, and fat tissue content. "Experimental error" according to Katch and Katch (1980) includes measurement error from multiple trials, error in determining residual volume, and erroneous estimation of intestinal gas volume. As is evident, body composition analysis is not yet an exact science. Nevertheless, we accept the values currently established and continue to attempt to quantify body composition into the basic components of lean body mass and percent of body fat.

Fortunately, these densitometric scores as determined by hydrostatic weighing offer encouraging measures of reliability. Katch and Katch (1980) suggest examining three parameters in support of this: body weight, underwater weight (body volume), and residual lung volume (RV). Without the influence of overhydration or dehydration, body weight fluctuates only minimally within a 1-2 hour testing period (Katch & Katch, 1980). This suggests, then, that any changes incurred from pre- to post-measurements of body weight are likely to be due to the inconsistency or imprecision of the scale apparatus, observer error, or within subject variation. Underwater weight, on the other hand, can vary noticeably. This measurement, which is directly related to the underwater residual lung volume measure, undoubtedly affects the estimation of body density.

Katch in his research from 1969, suggested performing an average of 10 trials before determining a true underwater weight. The most representative underwater weight should be estimated from the "trend line" of scores received from multiple trials (Katch, 1969). In contrast, to choose the highest or lowest weight from a series of trials would introduce more error than choosing a score which represented the subject's mean or median score (Katch, 1969). According to his previous research from 1968, Katch noted that the progressive increase in underwater weights throughout a series of trials was directly related to the subject's "learning effect". As subjects become more accustomed to making a maximal forced expiration while submerging themselves underwater, they progressively increase their underwater weights. Katch (1968) cited the body density values between trial 1 and trial 10 as varying by as much as .001-.003 density units. This variance represents a body fat percent difference of 0.4% to 1.2% (Katch, 1968). This seemingly small value could be interpreted as a relevant amount of difference in the cases of weight loss, weight gain, conditioning/ training programs, or diseased states.

The third influential parameter on density determination suggested by Katch and Katch (1980) is residual lung volume. These authors stated that error in residual volume measurements "can contribute substantially to changes in computed density." They, as a result, support a two trial method of residual volume measurement in which a final averaged value can be used for further calculations. By so doing, a standard error of measurement (SEM) of "usually less than 60 ml" can be obtained (Katch & Katch, 1980). In further calculations illustrating the effect of residual volume error, Katch and Katch (1980), cite a body density change of 0.08 density units, 8.0 percentage units of body fat, and 12 kg of lean body weight from a 600 ml difference in residual volume. Morrow, Jackson, Bradley, and Hartung (1986) support the importance of accurate RV measurement, but report greater possible variation between the two trials (177 and 160 ml for men and women prior to immersion; 195 and 118 ml for post-immersion). Morrow et al. (1986) also notes the 0.08 $gm\text{-}cm^{-3}$ impact on body density as a result of a 600 ml error in RV determination as cited by Katch and Katch (1980). Morrow et al. (1986) claim this to be in error, and instead suggest the resultant error effect to be perhaps 0.008 $gm\text{-}cm^{-3}$, indicating that possibly even smaller errors in RV measurement can have a major impact on the estimation of body density and percent body fat. The conclusion is apparent: specific attention to detail is warranted when attempting to measure RV if dependable scores are to be obtained. The lack of an exact measurement technique for RV further compounds the numerous assumptions inherent in present body composition analysis techniques.

Fluids and Electrolytes

As described for hydrostatic weighing, values for fat-free weight, total body water, and fat weight are also essential body composition components for bio-electrical impedance analysis. Being based on the principles of conductivity and resistance, the BIA is even more dependent upon the estimations of both lean body mass and total body water. Knowing that LBM contains the majority of TBW, it is here that electrolyte concentration must also be examined. This, however, is with the assumption that the percent of body water in the LBM does, in fact, represent the standard accepted value of 73.2%. Furthermore, these assumptions refrain from addressing the possible incidence of unequal distribution of body water within various body compartments, as previously mentioned.

Hoffer et al. (1969) recognized the possible importance of including electrolyte concentration data. They looked at serum sodium (Na_s), serum potassium (K_s), and water concentrates and entered them into the following equation: ionic mass = TBW $(Na_s + K_s)/\%H_2O$ which resulted in a correlation of 0.92 when compared with T^2/Z (Hoffer et al., 1969). As this was comparable to the less complicated comparison of total body water (TBW) versus T^2/Z (the accepted formula for estimating percent body fat as predicted from TBW values), no further exploration was performed at that time.

Lukaski and Bolonchuk (1986), in their study of validity measures on athletes, examined the BIA results for percent body fat as found in a group uncontrolled for preceding exercise or hydration level, and also in a controlled group having consumed a light meal within two hours of testing. The results were compared to HW, which was chosen as the accepted reference body composition technique. Their conclusions, although not specific to hydration volume or electrolyte concentration, did indicate that the uncontrolled measures were less accurate for percent body fat predictions than were the controlled measures (Lukaski & Bolonchuk, 1986).

When attempting to study the influence of hydration level and the associated ionic concentration changes, it is important to quantify tolerable, yet measurable volumes for ingestion, and also an appropriate time period for gastric emptying and absorption. Fordtran and Saltin (1967), in their study on gastric emptying, intestinal absorption, and endurance exercise, showed that gastric emptying was dependent upon the type of solution ingested and its ionic concentration. While water was most easily absorbed when compared to saline solutions, an isotonic saline solution actually emptied with more ease (Fordtran & Saltin, 1967). Furthermore, the addition of glucose was found to not only inhibit the emptying process, but to actually hinder the emptying of both water and sodium, as well (Fordtran & Saltin, 1967). Also in support of the use of an isotonic saline solution for rapid gastric emptying were Hunt and Pathak (1961), as their study indicated the same type of inhibition on gastric emptying with saline solutions which were either more dilute or more concentrated than plasma. The practical application of this information becomes limiting, however, as the ingestion of a sufficient volume of isotonic saline solution is difficult for most subjects.

Gastric emptying, as suggested by Fordtran and Saltin (1967), is dependent upon the type of solution ingested and its ionic concentration. It follows that absorption time required for equilibration into the tissues is also dependent upon this information. Cooper, Levitan, Fordtran, and Inglefinger (1966) offer relevant data on absorption speed and equilibration time periods recommended. They measured water and solute absorption via collecting tubes inserted directly into the small intestine. Their conclusions suggested that a 50-minute equilibration period provided more accurate accounts than did 30-minute measures for their isotonic electrolyte test solution (Cooper et al., 1966). They further stated that the precision of one hour measures also limited the possibility of irregular delivery due to possible variable patterns of oral ingestion.

Costill and Saltin (1974) demonstrated that gastric emptying was also influenced by gastric volume, or more accurately gastric pressure, and by test solution temperature. Their findings demonstrated a rapid rate of gastric emptying with a test solution temperature of 5° C, while Shapiro and Stoner (1968) showed reduced emptying rates with temperatures ranging from 2.5-9.0° C. Costill and Saltin (1974) further discussed the effect of gastric volume, stating that increased gastric pressure due to increased gastric volume did increase gastric motility and, therefore, emptying, but that this appeared to be effective only up to volumes between 600-750 ml., after which emptying again required more time. With a one liter test solution at a temperature of 5° C, Costill and Saltin (1974) found the gastric emptying rate to be approximately 290 ml/15 minutes. Further calculations suggest that an average volume of approximately 967 ml would be emptied within a 50 minute absorption time period.

Various volumes of fluid ingestion have been investigated with regard to gastric emptying, as well as, with dehydration in relationship to endurance exercise. Yet, research specific to fluid volume ingestion capable of registering with the BIA is minimal. Research of this nature is limited for HW, as well. Normal and extreme fluid volume changes undeniably vary among healthy individuals both at rest and when exercising. Fluid volume changes are also likely in various diseased states. Specifically, the measurement of hydration levels for athletes in a practical, yet precise manner will likely become increasingly essential in this age of maximized training and performance techniques.

Conclusion

The importance of body water is undeniable in general health, as well as, for fitness or athletic concerns. By determining an estimated value for percent of body water in free fat weight (FFW), total body water (TBW) can then be predicted. With the acceptance of the involved theoretical assumptions, percent body fat also becomes deducible information. The appearance and promise of the BIA in facilitating these measures further enhance the likelihood of establishing body composition analysis techniques that are reliable, rapid, safe, noninvasive, easily operated, and portable.

CHAPTER III

METHODS

Subjects

Thirty male subjects from the University of Wisconsin-La Crosse, ranging from 18-25 years of age, volunteered to participate in this study. Volunteers were accepted without fitness level assessment, or designated body fat percentage as criteria. Subjects were also requested to be medication free and in good health.

Preparatory Procedures

Subjects were personally contacted on an individual basis and briefed regarding testing preparations, conditions, and objectives (see Appendix A). A 12-14 hour fast was agreed upon prior to the scheduled testing period. Subjects were informed to eat properly and normally prior to fasting, but to avoid caffeine, nicotine, alcohol, medications, drugs, and foods that are known producers of flatulence. Subjects were also requested to avoid strenuous exercise for at least 5-6 hours prior to testing to minimize the occurrence of potential dehydration. Finally, subjects were requested to bring two pair of light weight shorts or swim trunks of comparable weight for wet and dry weight measurements.

Test Measurement Procedures

Upon arrival at the UW-L Human Performance Laboratory, subjects were presented with informed consent materials (see Appendix B) and given a brief explanation of testing procedures. Detailed explanation of each aspect of the testing process was provided as the subject was guided through the testing procedure.

Vital Capacity and Residual Volume

Subjects were first requested to perform one trial of a maximal forced expiration using a Collin's 9 liter Vitalometer to obtain an approximate vital capacity value. This information was valuable primarily when a subject's vital capacity was equal to or greater than the bag volume of the residual volume apparatus. After acquiring vital capacity, trial one of residual volume was performed. Subjects were given a noseclip, asked to inhale deeply, make a tight seal around the mouthpiece, then exhale forcefully until no more air could be expelled. At that moment, the valve allowing the subject to inhale pure oxygen was opened and the subject was coached through the mixing phase. Inhalations were expected to be fairly deep and somewhat rapid in an attempt to finalize the equilibration of the body's nitrogen with the premeasured bag volume of oxygen within approximately 6-10 respiratory exchanges.

The UW-L Human Performance Laboratory uses a closed circuit method of oxygen dilution for residual volume determination. With known values for room air, and by measuring the amount of nitrogen left in the lungs following maximal expiration (alveolar nitrogen-AN_2), and also by recording a value for impurity within the system (impurity nitrogen-IN_2), residual volume was determined. This was done by measuring the IN_2 mixed with the oxygen until equilibrium was obtained (equilibrium nitrogen-EN_2 and final nitrogen-FN_2). This process was measured by a

Collins electronic gas analyzer attached to a 6 liter Collins Vitalometer driven by a pulley and chain, and recorded by pen and recording paper on a spirometry bell. The UW-L Human Performance Laboratory determines residual volume as described based on the research and techniques of Wilmore (1969). The residual volume and underwater weighing data sheet is presented in Appendix C.

At this point in the testing procedure, the subject was directed to void body wastes, change into swimwear and return for dry weight and height as measured on a Health-O-Meter Scale (Continental Scale Corporation, Chicago). Measurements were recorded to the nearest 0.25 pound (lb.) and 0.25 inch, respectively. The body weight measures were then converted to kilograms (kg). Upon completion of this aspect of the procedure, the subject returned to complete RV trial number two. Two trials for RV determination are suggested by Katch and Katch (1980) in order to gain a reasonably dependable score. Both "trial one" and "trial two" for the RV determination included two actual RV measures. Data were considered acceptable if the value for impurity (IN_2) was below 2.0% and if FN_2 and EN_2 readings were comparable from one effort to the next (see Appendix D). These two measures were averaged and that value was recorded as the actual score for either trial one or trial two as appropriate.

With dry weight, height, and residual volume data collected for the preliminary phase of the testing procedure, the subject was then advanced to the Bioelectrical Impedance Analysis phase of the testing procedure.

Bioelectrical Impedance Analysis

The BIA was determined by the RJL Systems, Inc., (Detroit, Michigan). The model BIA-103B analyzer was used for this data collection. A four electrode method was used with two electrodes placed on the right hand and two on the right foot while the subject was supine (see Appendix E). All electrodes were cut into halves before being placed on the subject. The exact placement site was marked with permanent ink and maintained from the baseline phase of the procedure to the post-water ingestion phase. Electrode placements were sited on the dorsal surfaces of the hands and feet at the distal metacarpals and metatarsals between the distal prominances of the radius and the ulna and also between the medial and lateral malleoli of the ankle (Lukaski et al., 1985). Subjects were requested to lie so that their arms were not touching their sides and their legs were also not touching each other. Height, weight, age, and sex were entered into the computer program of the BIA. Resistance (R) was measured at 0-1,000Ω, and reactance (XC) at 0-200Ω from a constant current of 800μA at 50 kHz. Fat mass, lean mass, body fat, percent of body water, liters of body water, and a ratio of lean mass to fat mass were all determined from the equations provided by the manufacturer of the BIA (see Appendix F). This study examined the results recorded in the categories of percent body fat, lean body mass, percent of water, and liters of water only.

Hydrostatic Weighing

Following data collection with the BIA, subjects were directed to change into underwater weighing attire, and instructed to shower and wash their hair using soap/shampoo in order to remove excess body oils. With the shower completed, the subjects were then asked to enter the hydrostatic weighing tank. As the subject entered the tank, they were asked to immerse until their shoulders were just under the water surface to record the weight of the seating apparatus. After each subject was seated, they were instructed to sweep their hands over their arms, legs, chest, back, and swimsuit to remove any excess or trapped air bubbles. Water temperature of the HW tank was recorded at this time. A noseclip was provided for use at the subject's preference. It was requested that each subject forcefully exhale as completely as possible before gently lowering below the water surface into a tucked position. Any additional air was released underwater after turning the head slightly to the side so as not to trap any air in the chest/neck area. This tucked position was maintained until the recorder had received a measurement from the scale or until the subject no longer was comfortable underwater. Eight to 10 trial immersions and weight recordings (see Appendix C) were performed after which the subject was allowed to shower and return to street clothes. The average value of the "trend line" of scores was used for the final value for the underwater weight. No data were shared with the subjects at this time. The hydrostatic weighing process, as described, was performed in the UW-L Human Performance Laboratory Hydrostatic Weighing tank with the use of a Chatillion autopsy scale (15 kilos x 25 grams, Model #8-2096). The formula for the estimation of percent body fat was based on densitometric information as formulated by Brozek et al. (1963).

Fluid Ingestion

Upon completion of the initial (baseline) phase of the BIA and the HW procedure, subjects were given a volume of tap water (chilled between 0-9° C.) which represented 1.5% of their total body weight. Fifteen minutes were allowed for the ingestion period, and an additional absorption period of 50 minutes was required. Subjects were instructed to dress so that they would remain warm and dry during this manditory waiting period. They were reminded that no additional fluid or food should be ingested, and that no volume of body fluid or wastes should be eliminated until completion of the entire testing procedure. Subjects were restricted to two immediate study areas for the absorption period and were, therefore, under frequent observation during this time.

Post-fluid ingestion measures of dry weight and the two trials of residual volume were initiated at approximately 40 minutes into the absorption time period. As the 50 minute period ended, each subject repeated the process previously explained for the BIA. Following the BIA measures, subjects then repeated the entire testing process as described for HW. All test information was recorded on the data collection form found in Appendix G. Calculations for densitometry from the HW figures were then performed using the equation from Brozek et al. (1963). This procedure was used in both of the testing phases as described for the fluid ingestion, and the baseline data collections. Subjects were then debriefed with the data received.

Statistical Treatment

Raw data can be found in the tables of Appendix H. Standard descriptive charactistics were compiled separately (see Appendix I) before application of statistical analysis. A two by two analysis of variance (ANOVA) with repeated measures was performed. The variables of lean body mass (LBM) and percent body fat (% BF) were examined as recorded in both the pre- and post-fluid ingestion measures for the BIA and the HW.

Dependent "t" tests for dependent variables were calculated on the two trials (pre and post) of residual volume as recorded for the HW, and on the two trials (pre and post) of resistance (R), conductance (XC), percent of water (%H_20), and liters of water (LH_20) as recorded for the BIA.

Significance was established at the 0.05 level of confidence, and post-hoc Scheffe' tests were performed where appropriate.

CHAPTER IV

RESULTS AND DISCUSSION

Introduction

The purpose of this study was twofold. By comparing both pre- and post-fluid ingestion measures of the bioelectrical impedance analysis (BIA) with those from hydrostatic weighing (HW), BIA validity was examined. Secondly, and of more importance to BIA research, the sensitivity of the BIA to fluid ingestion was determined.

Subjects

Thirty college age males volunteered as subjects for repeated body composition analysis. Table 1 presents the descriptive characteristics of these individuals. All subjects were measured by HW, as well as BIA, prior to ingestion of 1.5% of their body weight in tapwater. After a mandatory 50 minute absorption period all subjects were then remeasured by both the HW and BIA methods of body composition determination.

Pilot Study

The volume of tapwater representing 1.5% of a subject's body weight was a value chosen primarily through the results of the pilot study. A volume was needed which would be comfortably ingested in a short period of time, easily absorbed in a reasonable period of time, and minimally stressful to the gastrointestinal system, yet potentially measurable by the apparatus in question. Six male subjects (age 22-25 yrs) were given fluid volumes ranging from 0.15%-3.36% of their body weight for ingestion

Knowing that a small sample size such as this would not offer any precise values of tolerance, the subjective responses of these subjects were used for the purpose of establishing a practical volume for the ingestion phase. At a volume of 2.0% of body weight or greater, immediate discomfort and decreased cooperation were apparent. A volume of 1.5%-2.0% of body weight appeared to have varying effects on different individuals. Therefore, the volume of 1.5% of body weight was administered. This was equal to a range of 869-1,424 mls ingested for the involved subjects (see Table 2).

Table 1. Descriptive characteristics of test population. (N = 30)

Variable	Mean	SD	SE	Range
Age (yrs)	21.3	1.77	0.322	18 - 25
Height (in)	70.3	2.99	0.546	62.5 - 75.7
Weight (kg)				
Pre-Hydration	72.4	9.40	1.716	57.9 - 94.9
Post-Hydration	73.5	9.50	1.733	59.1 - 95.9

Comparatively, Girandola and his associates (1977), looked at body composition changes resulting from both fluid ingestion and dehydration. Their 10 subjects ingested water according to their individual capacity and tolerance, consuming anywhere from 1.2-2.4 liters (Mean = 1.8 L). Although their values were not represented as a percentage of the subject's body weight, these values were at least within a comparable range to the volume consumed in the present study.

Table 2. Volumes of fluid ingested representing 1.5% of body weight.

Subject	Weight (kg)	Volume (ml)
01	80.24	1,204
02	58.14	872
03	68.86	1,033
04	57.94	869
05	75.86	1,138
06	63.73	956
07	59.64	895
08	78.93	1,184
09	61.69	925
10	68.27	1,024
11	74.05	1,111
12	77.68	1,165
13	81.19	1,218
14	69.06	1,036
15	74.84	1,055
16	68.15	1,022
17	60.10	902
18	64.75	971
19	66.79	1,003
20	85.96	1,290
21	68.95	1,034
22	89.36	1,340
23	84.60	1,269
24	76.32	1,146
25	68.83	1,033
26	68.15	1,022
27	94.92	1,443
28	77.34	1,160
29	68.83	1,033
30	79.04	1,186
Mean	72.41	1,084

Measure of Validity

Measures of percent body fat (% BF) as established by the BIA compared to HW for both pre- and post-hydration were significantly ($p < 0.05$) higher. These results indicated that the BIA measurements predicted a higher % BF than those obtained by HW, with a mean difference from pre-BIA to pre-HW of 2.60% higher, and a mean difference from post-BIA to post-HW of 3.10% higher (see Table 3). As would be expected, the BIA values for lean body mass (LBM) were correspondingly lower than those from HW in both the pre-BIA to pre-HW comparison, and the post-BIA to post-HW comparison (see Table 3). These LBM values, however, were not significantly ($p > 0.05$) lower. As it was proposed that the variance between techniques for the estimation of % BF would not be significantly different, this finding necessitated the rejection of that hypothesis.

Yet, these findings were consistent with those reported by several other authors. Miles and Stevens (1986), Keller and Katch (1986), and Nash (1985), all suggested that the BIA repeatedly overestimated % BF in subjects nearest the leaner range of the body composition continuum, and underestimated the % BF of those subjects who tended toward the fatter values on such a continuum. Although the definitions of "leaner" and "fatter" were not provided, it would seem reasonable to include body fat percentages of less than 20% for males in the "nearer the leaner end" category of a body composition scale. The subjects in this study ranged in initial % BF as determined by HW from 1.94-19.29%, with a mean of 10.25% (see Appendix J). This sample of college age males may well have represented a comparatively lean sample group, which would be consistent with the direction of the results for % BF. Furthermore, Shore and Taylor (1986) in their study on males, also reported the BIA as significantly ($p < 0.001$) overpredictive for % BF. The estimated % BF values for their subjects for HW and BIA, were 16.7% and 18.8%, respectively.

Table 3. Mean and SD for pre- and post-ingestion of % BF and LBM between the BIA and HW techniques. (N =30)

	BIA			HW		
Variable	Pre-	Post-	Δ	Pre-	Post-	Δ
% BF	12.9[a] 3.34	14.1[b] 3.13	1.20	10.3 4.50	11.0 4.17	0.80
LBM (kg)	63.3 8.07	63.0 7.59	0.30	64.9 8.05	65.3 8.28	0.40

a = Pre-BIA/Pre-HW: 2.60%
($p < 0.05$)
b = Post-BIA/Post-HW: 3.10%
($p < 0.01$)

In comparison to the results found in this study, other researchers simply concluded that the validity of the BIA for determining % BF was inconsistent and at best could be considered more dependable than skinfold measurements (Renk, et al., 1986; Lawlor, et al., 1986; Katch, et al., 1986). Lukaski et al. (1986) examined the measure of LBM specifically. Unlike others, they stated that the principle of conductance was both reliable and valid for either males or females, however, they further concluded only that BIA measurements were more valid than anthropometric measures. In a separate work, Lukaski and Bolonchuk (1986) offered more directional results. They cited the tendency for the BIA to overestimate LBM and underestimate % BF. These findings were opposite of the findings of this study, but this could be attributed to their uncontrolled variables of hydration, exercise, and food consumption as was suggested by Lukaski and Bolonchuk (1986).

Other researchers also reported results opposite to the findings of this study. Miranda and associates (1986) cited a variance from HW for the BIA of 2.1% body fat for males, but in contrast to the present study, found this to be in the direction of underprediction (HW = 14.1%, BIA = 12.0%). Keller and Katch (1986) also reported the underprediction of % BF for males when determined by the BIA. Furthermore, Segal et al. (1985) examined LBM measures and reported similarly that as % BF increased, the accuracy of the BIA decreased in such a way that the LBM was also overestimated. The subjects in the study by Segal et al. were measured at 28.53% ± 14.23 for HW, and 26.24% ± 10.56 for BIA.

The undeniable conclusion is that the research currently available on the validity of the BIA is undoubtedly equivocal. The results of the present study are certainly in keeping with that of comparable research efforts. Consequently, the use of the BIA for valid body composition analysis of % BF must be done with prudence.

Sensitivity to Fluid Ingestion

After initial measures of % BF and LBM by both the BIA and HW, each subject consumed 1.5% of his dry body weight in chilled (0 - 9^{o}) tapwater and then waited 50 minutes for the mandatory absorption period.

Following this procedure, all subjects were remeasured by both the BIA and HW. These values represent the potential sensitivity of the technique to fluid ingestion.

Percent Body Fat

The results of this study found the treatment effect of hydration on % BF to be statistically insignificant ($p > 0.05$) in the measures provided by the BIA (see Table 4). This necessitated the acceptance of this study's number 2 (null) hypothesis that the ingestion of fluid would not create a significant change in the percent of body fat as measured by the BIA. The mean value for % BF as measured before water ingestion for the BIA was 12.90%. After hydration, this increased to a mean % BF of 14.10%.

Although this 1.20% change was not significant, it was a consistent change. The range for the treatment effect on the BIA for 29 out of 30 subjects was 0.3-6.8% body fat. This indicated that the effect of the overhydration was perhaps physiologically significant, although not statistically significant ($p < 0.05$).

In an attempt to minimize any change from pre- to post-hydration measures unrelated to the treatment, electrode placement sites were marked with permanent ink at the initial BIA measurement. The electrodes were then placed at those marks for the post-hydration measure. Lukaski et al. (1985) reported that the reproducibility of measuring total body water (TBW) and LBM, from which % BF is determined, would compare to a .99 test-retest correlation coefficient if the electrodes were placed at designated landmarks. Only one value out of the 30 subjects tested demonstrated a change of direction which indicated a decrease in % BF following hydration as measured by the BIA (see Appendix K).

The treatment effect of hydration on HW also produced an increase in % BF (0.70%), but this too was not significant at the 0.05 level (see Table 4). Since the subjects were weighed in water for HW, the volume of fluid ingested should have had a minimal effect on their underwater weight. As Girandola et al. (1977) reported, body density is minimally influenced from hydration changes since the density of body water and the water in the HW apparatus are essentially of the same density. This creates a situation where the water weighs relatively nothing in water. Therefore, the change from pre- to post-hydration for HW cannot knowingly be attributed to the hydration treatment, but may in fact, be the result of biological or experimental variation and error.

Overall, the response to the hydration treatment on % BF was not significant ($p > 0.05$) for either the BIA or HW. The difference between the two techniques in their responses to the treatment also was not significant ($p > 0.05$) with a difference between technique responses being only 0.50% (see Table 5). This lack of significance suggests that the two techniques responded in similar ways to hydration. Both techniques, did however, indicate a trend toward increased % BF following the ingestion of a volume of water which represented 1.5% of a subject's body weight (see Appendix K and L).

Lean Body Weight

Of interest in relation to the results for % BF, are the results for LBM following hydration treatment. It was expected that when % BF was a higher value, LBM would be of a lesser value. The treatment effect on the BIA did produce such a response (see Table 4). As the changes in % BF following hydration for the BIA were not significant, it was logical that the changes for LBM also were not significant. The change in LBM from pre- to post-hydration for the BIA was very slight (-0.40 kg) and could only best be considered the result of biological variance or experimental error.

Table 4. Mean and SD representing the treatment effect of hydration on % BF and LBM as measured by BIA and HW. (N = 30)

	BIA		HW		Δ	
	% BF	LBM (kg)	% BF	LBM (kg)	% BF	LBM (kg)
Pre-Hyd.	12.90	63.4	10.30	65.0	2.60	1.60
	3.34	8.07	4.50	8.05		
Post-Hyd.	14.10	63.0	11.00	65.3	3.10	2.30
	3.13	7.59	4.17	8.28		
	+1.20	-0.40	+0.70	+0.30	0.50	0.70

Similarly, the treatment effect on LBM as measured by HW was not significant (see Table 4). These values, however, did not follow the expected pattern. As the mean % BF value increased from pre- to post-hydration (as measured by HW), so did the values for LBM. As mentioned, the expected response was for LBM to decrease as % BF increased. Since this value for the treatment effect was only +0.30 kg., and did not represent significance at the $p < 0.05$ level, justification of this change is limited. In either case, the mean treatment effect was very slight and it would be risky to project whether these results suggested a trend or were merely the response to biological variation or experimental error.

Other Variables

Additional variables were examined following the initial two by two ANOVA (with repeated measures) and the subsequent Scheffe' post hoc statistical treatment. Dependent "t" tests were performed on residual volume (RV), resistance (R), conductance (XC), percent of water ($\%H_20$), and liters of water (LH_20).

Residual Volume

As previously mentioned, residual volume is often a substantial factor in the determination of body density (Katch & Katch, 1980; Morrow et al., 1986). In this study the subjects performed RV once before hydration and again after hydration. No practice opportunity was provided prior to these measures. Only six of the 30 subjects had previous experience with RV or HW. Consequently, a dependent "t" test was calculated to determine if a significant difference appeared between the two RV measures. The mean RV values did show a mean increase of 0.083 liters from trial 1 to trial 2 (pre-post hydration) indicating that the subjects were forcing more air out on trial 2, but this difference was not significant at the 0.05 level (see Table 5).

Table 5. Variability between trials for residual volume (RV).

	Mean (L)	SD	SE
Trial 1 (pre-hydration)	1.70	0.444	0.081
Trial 2 (post-hydration)	1.62	0.438	0.080

Other variables were looked at specific to the BIA. As mentioned before, the results for % BF and LBM indicated that the BIA was able to detect a consistent change from pre- to post-hydration status. Although this change was not significant ($p < 0.05$), further examination of the sensitivity of the BIA to the hydration change through other variables was needed.

Resistance

Resistance (R) and conductance (XC) measure the ability of the electrical current to travel through the tissues from one electrode to the other. These measures are directly affected by the components of total body water, lean body mass, and nonessential fat. Presta et al. (1983) reported that fat-free weight (FFW), or lean body mass (LBM) as it is commonly referred to, contains more water and the associated electrolytes than does body fat. As Lukaski et al. (1986) reported, conductance is enhanced by both body water and the electrolyte concentration. Consequently, a leaner person has a proportionately greater volume of total body water with the appropriate concentrations of electrolytes. Resistance would, therefore, be lower and conductance would be greater than for a person with a high percent of body fat. The results of this study indicated that the direction of change in resistance from pre-hydration to post-hydration was significantly greater ($p < 0.05$), as expected (see Table 6). This was due to the dilutional effect of the additional water upon the existing electrolyte concentration. The addition of water to the system increased the resistance by a mean value of 8.50Ω.

Table 6. Mean and SD of hydration effect on BIA variables of resistance, conductance, percent of water, and liters of water.

	Pre-Hydration		Post-Hydration		Δ
	Mean	SD	Mean	SD	
Resistance (R) 0-1,000Ω	453.77	49.04	62.27*	43.95	+8.50*
Conductance (XC) 0-200Ω	64.7	9.01	68.4*	5.83	+3.70*
Percent of Water	65.04	4.55	63.56*	3.63	-1.48*
Liters of Water	46.88	4.67	46.46*	4.62	-0.42*

* = Sigificance at $p < 0.05$

Conductance

Conductance (XC), on the other hand, would be expected to decrease following the addition of water to the body. Although the addition of water would seemingly lead to responses similar to those of a leaner person, this was not the case. The increased water volume without an accompanying electrolyte increase, only served to dilute the existing electrolytes. Thus, the conductance speed decreased, and the resistance values increased significantly ($p < 0.05$). These expected changes were consistent for resistance, but not for conductance (see Table 6).

Body Water

Furthermore, percent of water and the resultant value of liters of water were examined. Contrary to the expected response, both values decreased following the increase in total body water from the ingested tapwater (see Table 6).

Implications/Summary

This research project was designed to examine the validity of the BIA as compared to HW, and to examine the sensitivity of the BIA to overhydration. The results regarding validity measures were consistent with those of other researchers in the field. The measures for % BF were significantly ($p < 0.05$) overestimated as compared to those determined by HW. The BIA overpredicted % BF by as much as 2.60-3.10%. The corresponding values for LBM were lower than those estimated by HW by 1.60-2.30 kg, but were not significantly lower. In testing the sensitivity of the BIA to overhydration, no statistical significance was found for either % BF or LBM. The values for both % BF and LBM indicated a possible trend toward demonstrating the BIA's ability to sense an increase in % BF following increased total body water. This pattern was present in 29 out of 30 subjects for % BF and 20 out 30 subjects for LBM (see Appendix K and M).

The implications from these results are twofold. As with other studies, the validity measures from the BIA demonstrated a need for improved precision. The fluid sensitivity measures only suggested the possibility that the BIA was capable of detecting relatively small changes in what it represented as % BF. This, of course, was erroneous. Body fat did not increase, but total body water did. This distinction was not clear from the results the BIA provided. The values measured for %$H_2$0 and L$H_2$0 were both perceived as significantly changed at 0.0000 and 0.038, respectively. These changes, however, were perceived as decreases in total body water. The equational procedure for the BIA measures % BF based on the values given for resistance and conductance. As these values would be disturbed due to the dilutional effect of the ingested water on the electolyte concentration, it follows that the water values could also be erroneaously perceived. This suggests then, that not only was the ingested fluid volume absorbed into the tissues as desired, but that the BIA may indeed be sensitive to this type of change in TBW. The problem remains in the interpretation of this information. Finally, the results may possibly have been more directional if a larger volume of water had been ingested by the subjects.

CHAPTER V

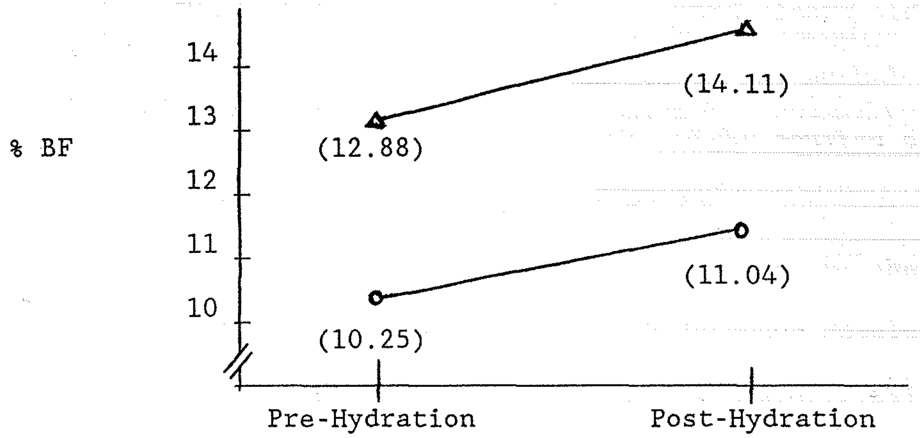
SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

This study primarily was designed to examined the sensitivity of the BIA to overhydration in the assessment of % BF in body composition analysis. In using hydrostatic weighing as a reference technique, validity measures were also studied.

Thirty college age male volunteers from the University of Wisconsin-La Crosse, agreed to undergo repeated body composition assessment. Initial testing included hydrostatic weighing followed by bioelectrical impedance analysis. The subjects were then given a volume of tapwater representing 1.5% of their total body weight to consume. Following the required 50 minute absorption period, all subjects then repeated both the HW and BIA testing phases.

The results for validity testing on the BIA indicated a significant ($p < 0.05$) overestimation of % BF (see Figure 1). Comparably, lean body mass (LBM) is generally expected to decrease as % BF increases. This was, in fact, what was found. With the addition of a mean weight gain of 3.38 kg following overhydration, the BIA demonstrated a mean increase in % BF of 1.20%. The simultaneous mean increase in LBM was 0.40 kg. Neither the increase in % BF or the decrease in LBM following overhydration were statistically significant ($p < 0.05$).

Figure 1. Mean difference between HW and BIA in the assessment of % BF both before and after overhydration.



* Note: BIA = △
HW = ○

Although the treatment effect of overhydration was not statistically significant ($p < 0.05$), a definite trend was displayed for % BF. Twenty-nine out of 30 subjects were measured as increasing in % BF from the pre- to post-hydration phases. Percent of body fat did not actually increase, but the BIA was sensitive to the decrease in conductivity and/or increase in resistance which accompanied the additional TBW volume. The dilutional effect of this additional water on the existing electrolyte concentration was ultimately perceived as an increase in % BF. Although not refined in assessment, the BIA does offer promise that it can detect changes in body composition of this nature.

Recommendations/Conclusions

Overall, the results of this study offer promise. As was expressed previously, equation refinement must constantly be underway in order to improve the validity of the measures found by the BIA for % BF. Sensitivity measures, on the other hand, need continued research. Sensitivity regarding not only hydration, but dehydration, food ingestion, age groups with varied bone densities, the influence of racial variances, disease states, the effect of decreased temperature, nondominant side for electrode placement versus dominant side, the use of the actual electrical pathway versus the present equation of body height squared divided by resistance, and numerous others could be helpful.

Recommendations specific to this study would include the use of larger volumes of water, a larger and more homogenous sample, and also the use of a saline solution to see the resultant electrical effects more clearly and comparatively.

Many assumptions must be accepted at the present in body composition analysis. The more refined any one component can become, the more refined the entire process has the potential to be. The BIA offers such promise in complimenting hydrostatic weighing in the domain of body composition determination. In its current form, the BIA primarily offers potential as a technique for measuring changes in body composition such as for weight loss or weight gain programs where major health concerns are not a factor.

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APPENDICES

APPENDIX A

PRETEST GUIDELINES

BIA HYDRATION SENSITIVITY

Pre-test Guidelines

The following guidelines are important to the success of an accurate measure of your percent body fat. Please make all necessary efforts to adhere to them. If for any reason you are unable to meet these requests, please inform me (Bev Davison) prior to any test measurements.

1. Fast for 12 hours prior to actual testing.
2. Avoid gas producing foods: beans, chocolate, cabbage, etc.
3. Avoid caffeine.
4. Avoid nicotine.
5. Avoid strenuous exercise six hours prior to actual testing.
6. Eat regular, nutritional meals daily prior to fast episode.
7. Take no drugs or medications, unless previously discussed with the tester.
8. Consume fluids as needed on a minimal basis during fast episode.
9. Urinate, allow bowel movement, and void flatus immediately prior to testing.

APPENDIX B

INFORMED CONSENT FORM

INFORMED CONSENT FORM

The University of Wisconsin-La Crosse
La Crosse, Wisconsin

VALIDITY AND SENSITIVITY MEASURES OF THE BIA IN RESPONSE TO OVERHYDRATION

(Bev Davison)

I, ______________________________, agree to participate in the body composition study being conducted in the Human Performance Laboratory at the University of Wisconsin-La Crosse. I understand that the participation in this study will involve one visit to the Human Performance Laboratory. At the testing time I will have residual lung volume determined, body composition determined by both the underwater (hydrostatic) weighing and the bioelectrical impedance analysis (BIA) techniques, and that I will be asked to consume a fluid volume representing 1.5% of my total body weight. I am aware that the fluid solution will consist only of tap water. I also understand that I may withdraw from the study at anytime.

In any type of testing situation some potential risk is involved. When working in a water environment these risks may include infection, accident, and possible drowning. When working with an electrical apparatus, potential risks include possible shock or accidental electrocution. There has, however, never been an accident or report of serious infection as a result of the hydrostatic weighing or bioelectrical impedance analysis procedures at the University of Wisconsin-La Crosse Human Performance Laboratory.

I am also aware that feelings of fullness, nausea, or other gastrointestinal discomforts may occur when consuming a fluid volume which represents 1.5% of my total body weight. Other possible discomforts may occur as a result of voluntary overnight fasting, permanent marker ink, or electrode adhesive.

The actual testing will be conducted by Bev Davison, a graduate student in the Human Performance: Exercise Physiology program at the University of Wisconsin-La Crosse. She will be under the supervision of Nancy Kay Butts, Ph.D.

I, ______________________________, approve of the procedure as explained for the body composition study at the University of Wisconsin-La Crosse's Human Performance Laboratory and agree to participate. I have read the foregoing and I understand it. Any questions which may have occurred to me have been fully answered to my satisfaction. The potential risks have been explained to me and I fully understand their implications. I hereby acknowledge that no representations, warranties, guarantees, or assurances of any kind pertaining to the procedures have been made to me by the University of Wisconsin-La Crosse, the officers, administrators, employees, or by anyone acting on behalf of them.

Signed:______________________________ Date:__________

Witness:______________________________ Date:__________

APPENDIX C

RESIDUAL VOLUME AND UNDERWATER WEIGHT DATA FORM

RV AND UNDERWATER WEIGHING DATA FORM

BODY COMPOSITION - HUMAN PERFORMANCE LAB

Subject's Name:______________________________ Age:____ Sex: (1=male, 2=female)

Tester's Name:_______________________________ Date: (mo/day/yr):______________

Prior Tests? (yes/no):______ Reason for Test:__________________________________

Comments:____

****************************INFORMED CONSENT AGREEMENT***************************

The procedures involved in underwater weighing, residual volume measurement, and anthropometric measurements (skinfolds, girths and diameters), have been explained clearly to me and I understand them. To my knowledge I have no physical handicaps or medical conditions that would prevent participation in these tests. I agree to hold harmless the University of Wisconsin-LaCrosse and its employees for any accidental injury that might occur as a result of these tests, which I have agreed to take.

Signed:____ Date:_______________

****

A. Vital Lung Capacity: #1______liters (to .1), #2________liters (to .1)

B. RV Determinations:

	Trial 1:	Trial 2:
Bag Volume of Oxygen (BV)	_______liters (to .01)	_______liters (to .01)
Alveolar nitrogen (AN)	_______% (to .1)	_______% (to .1)
Impurity nitrogen (IN)	_______% (to .1)	~~_______% (to .1)~~
Equilibrium nitrogen (EN)	_______% (to .1)	_______% (to .1)
Final nitrogen (FN)	_______% (to .1)	_______% (to .1)

C. Direct subject to dressing room to put on swimsuit. NO SHOWER YET!

D. Anthropometry (mean of 3 readings):

Male subjects:

Skinfolds:
Triceps _________mm
Biceps _________mm
Subscapular _________mm
Suprailiac _________mm
Thigh _________mm
Pectoral _________mm
Midaxillary _________mm
Umbilical _________cm

Girth: Suprailiac _________cm

Diameter: Wrist _________cm

Female subjects:

Skinfolds:
Triceps _________mm
Biceps _________mm
Subscapular _________mm
Suprailiac _________mm
Thigh _________mm

Girths:
Forearm _________cm
Abdominal _________cm
Buttocks _________cm

Diameter: Wrist _________cm

E. Direct subject to toilet to void any solid, liquid or gas possible.

F. Dry Weight____________lbs (to 1/4)   Height___________inches (to 1/4)

G. Direct subject to shower to wash hair and shower completely. Jewelry and contact lenses should be removed before underwater weighing!

H. Densitometry:

Water Temp ____degrees C.   Immersed Weight of Apparatus (MY)________(to .1)

MX: (1)_________ (2)_________ (3)_________ (4)_________ (5)_________
(6)_________ (7)_________ (8)_________ (9)_________ (10)_________

Select the heaviest reproducible weight (MX) from the trials above:
Immersed weight of subject + apparatus (MX)________Kg (to .1).

I. Results:

J. Equations: *RV = 1.1 X [BV X (EN - IN) - DS] / (AN - FN)

**BD = MA / { [(MA - MW) / DW] - RV - .1 liter }

***FAT% = (457 / BD) - 414.2

K. References: *Wilmore, J.H. 1969. A simplified method for determination of residual lung volumes. JOURNAL OF APPLIED PHYSIOLOGY 27: 96 - 100.

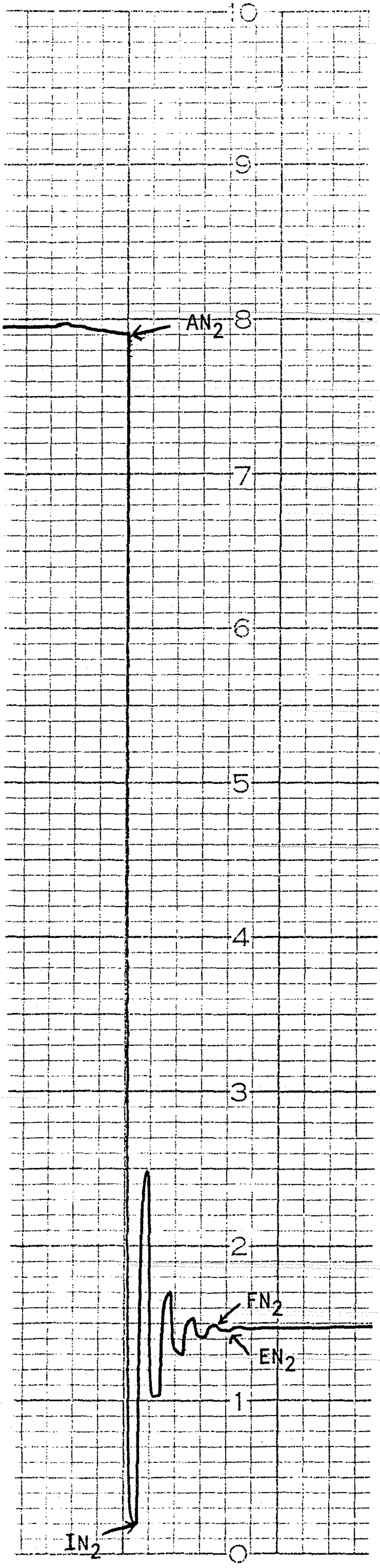
**BusKirk, E.R. 1961. Underwater weighing and density: a review of procedures. In: "Techniques for Measuring Body Composition". Edited by J. Brozek and A. Henschel. National Academy of Sciences - National Research Council. Washington D.C.

***Brozek, J., A. Henschel, J.T. Anderson, and A. Keys. 1963. Densitometric analysis of body composition: revision of some quantitative assumptions. ANNALS OF THE NEW YORK ACADEMY OF SCIENCES. 110: 113 - 140.

APPENDIX D

RESIDUAL VOLUME CHART-PAPER EXAMPLE

RESIDUAL VOLUME CHART-PAPER EXAMPLE



AN$_2$ = Alveolar Nitrogen

IN$_2$ = Impurity Nitrogen

EN$_2$ = Equilibrium Nitrogen

FN$_2$ = Final Nitrogen

APPENDIX E

ELECTRODE PLACEMENT DIAGRAM

ELECTRODE PLACEMENT DIAGRAM

DETECTING ELECTRODE

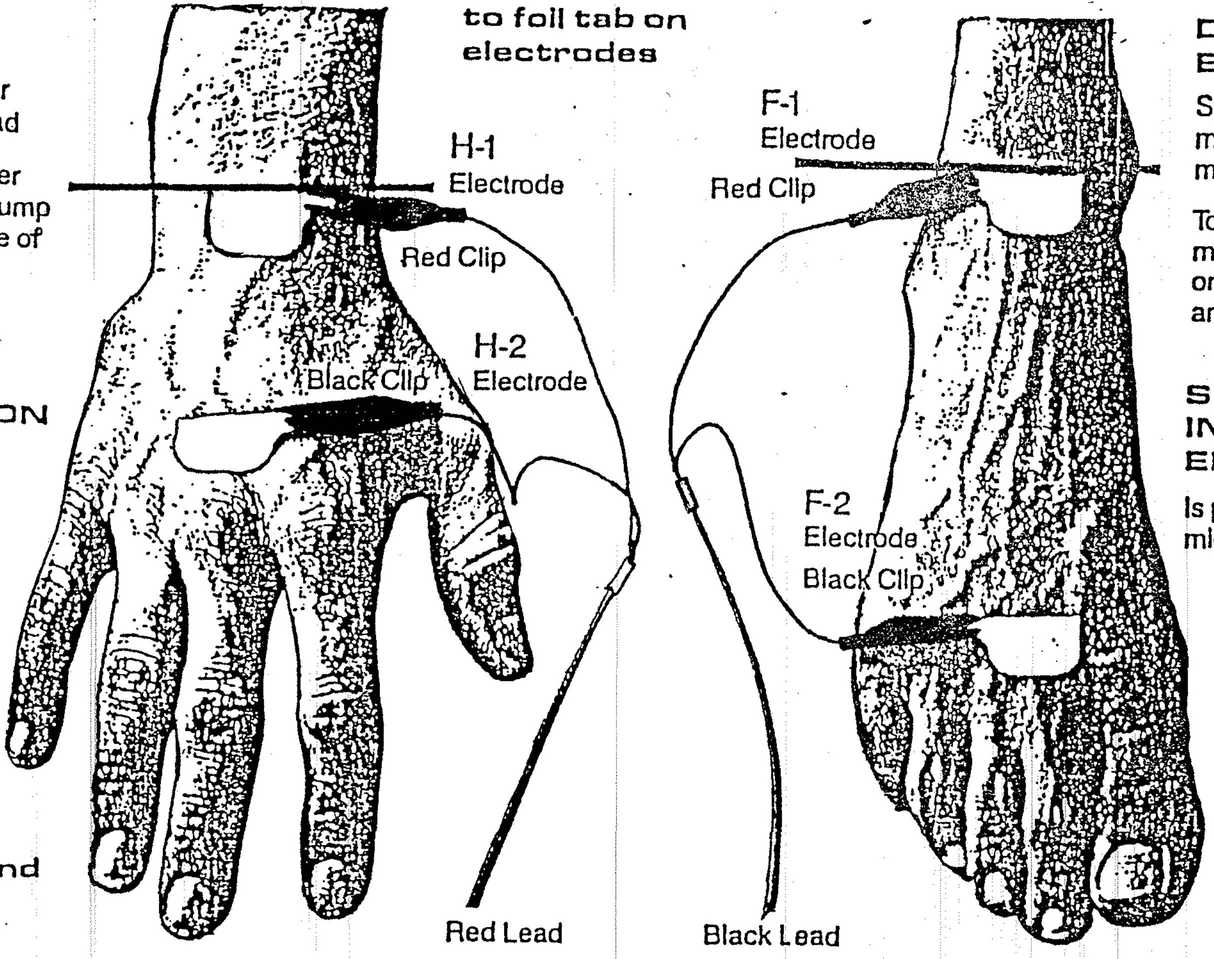
Superior linear border must bisect ulnar head

Top straight line border must cut in half the bump on the little finger side of the wrist

SIGNAL INTRODUCTION ELECTRODE

Is placed just behind middle finger

Clips are Attached to foil tab on electrodes



DETECTING ELECTRODE

Superior linear border must bisect the medial malleolus

Top straight line border must cut in half the bump on the big toe side of ankle

SIGNAL INTRODUCTION ELECTRODE

Is placed just behind the middle toes

APPENDIX F

BIOELECTRICAL IMPEDANCE ANALYSIS EQUATIONS

BIA EQUATIONS

Males: BD = 1.1554 - 0.0841*WTMRZ/HTM 2

Females: FAT = (1.0 - (.3981*H2R + WTM + (HTM - 100) + .7414/WTM)*100

BD = Body Density
RZ = Resistance
H2R = HTM 2/RZ
HTM = Metric Height
WTM = Metric Weight

(RJL BIA-103, RJL Systems, Inc. Detroit, Michigan, 1986)

APPENDIX G

OVERALL DATA COLLECTION FORM

NAME: AGE: #

Date: Time:
Solution: tap water Temp.: C.
Volume: ml.

Dry Wt.(.25 lb)
Dry Wt. (kg)
Height (in) (in) (cm)
1.5% BW (kg) ml.

BIA: (BIA-B)
Resistance (R)
Conductance (XC)
% fat *
Fat mass (lb)
Lean mass (lb) (kg)
% H20
L H20
Lean/fat ratio

HW: (HW-B)
Residual volume #1 #2
Body Density EN2
% fat *
LBW (lb) UWW FN2
LBM (kg)

Ingst. time begin
Ingst. time end
Abs. time begin
Abs. time end

Dry Wt. (.25 lb) (lb) (kg)

BIA: (BIA-PS)
Resistance (R)
Conductance (XC)
% fat *
Fat mass (lb)
Lean mass (lb) (kg)
% H20
L H20
Lean/fat ratio

HW: (HW-PS)
Residual Volume #1 #2
Body density EN2
% fat *
LBW (lb) UWW FN2
LBM (kg)

COMMENTS:

APPENDIX H

RAW DATA

RAW DATA

Subject	Test	RV	% BF	LBM	R	XC	%H_2O	LH_2O
01	1	0.000	12.00	71.71	430	057	64.2	52.3
01	2	1.421	08.81	74.33	000	000	00.0	00.0
01	3	0.000	13.30	71.76	439	065	62.7	51.9
01	4	1.375	09.49	74.91	000	000	00.0	00.0
02	1	0.000	10.40	52.21	508	064	70.0	40.8
02	2	1.570	11.21	51.74	000	000	00.0	00.0
02	3	0.000	10.90	52.71	507	068	69.3	41.0
02	4	1.692	10.79	52.79	000	000	00.0	00.0
03	1	0.000	14.20	59.97	469	069	63.6	44.5
03	2	1.240	16.37	58.50	000	000	00.0	00.0
03	3	0.000	13.90	60.37	464	078	63.9	44.8
03	4	1.478	15.04	59.52	000	000	00.0	00.0
04	1	0.000	03.10	56.16	344	100	81.6	47.3
04	2	1.015	14.98	49.26	000	000	00.0	00.0
04	3	0.000	09.90	53.16	425	062	70.4	41.6
04	4	1.210	14.67	50.40	000	000	00.0	00.0
05	1	0.000	12.40	66.41	455	071	64.5	48.9
05	2	2.353	04.37	72.53	000	000	00.0	00.0
05	3	0.000	13.60	66.41	464	076	63.1	48.5
05	4	2.124	04.79	73.19	000	000	00.0	00.0
06	1	0.000	13.80	54.93	498	072	65.3	41.6
06	2	2.005	11.79	56.21	000	000	00.0	00.0
06	3	0.000	14.50	55.16	501	073	64.3	41.5
06	4	1.942	11.94	56.81	000	000	00.0	00.0
07	1	0.000	12.90	51.94	433	063	66.9	39.9
07	2	1.507	13.07	51.84	000	000	00.0	00.0
07	3	0.000	13.70	52.21	436	069	65.9	39.9
07	4	1.310	12.95	52.70	000	000	00.0	00.0
08	1	0.000	11.30	69.99	454	060	65.3	51.5
08	2	1.614	11.85	69.56	000	000	00.0	00.0
08	3	0.000	12.20	70.26	459	067	64.1	51.3
08	4	2.017	09.44	72.49	000	000	00.0	00.0
09	1	0.000	12.60	53.89	514	066	66.8	41.2
09	2	2.379	02.85	59.92	000	000	00.0	00.0
09	3	0.000	13.50	54.20	518	075	65.7	41.2
09	4	1.844	07.44	58.03	000	000	00.0	00.0
10	1	0.000	12.90	59.47	495	066	65.2	44.5
10	2	1.647	07.96	62.82	000	000	00.0	00.0
10	3	0.000	13.80	59.65	500	071	64.1	44.4
10	4	1.551	10.47	62.02	000	000	00.0	00.0

RAW DATA (cont'd)

Subject	Test	RV	% BF	LBM	R	XC	%H_2O	LH_2O
11	1	0.000	11.30	65.64	410	064	65.9	48.8
11	2	2.049	06.48	69.24	000	000	00.0	00.0
11	3	0.000	12.10	65.82	414	067	65.0	48.7
11	4	2.159	05.66	70.70	000	000	00.0	00.0
12	1	0.000	14.30	66.50	507	064	62.3	48.4
12	2	1.280	12.82	67.71	000	000	00.0	00.0
12	3	0.000	15.70	66.32	518	069	60.9	47.9
12	4	1.228	12.51	68.84	000	000	00.0	00.0
13	1	0.000	10.50	72.67	411	062	66.0	53.6
13	2	1.541	12.34	71.16	000	000	00.0	00.0
13	3	0.000	11.40	72.80	417	070	64.7	53.2
13	4	0.914	16.24	68.85	000	000	00.0	00.0
14	1	0.000	16.90	57.34	508	074	61.4	42.4
14	2	1.175	09.00	62.83	000	000	00.0	00.0
14	3	0.000	18.60	57.11	520	074	59.8	42.0
14	4	1.151	10.49	62.82	000	000	00.0	00.0
15	1	0.000	09.70	67.59	430	054	67.8	50.7
15	2	1.379	08.58	68.41	000	000	00.0	00.0
15	3	0.000	10.50	67.81	435	059	66.6	50.5
15	4	1.290	09.73	68.47	000	000	00.0	00.0
16	1	0.000	16.40	56.97	501	064	61.9	42.2
16	2	1.746	07.94	62.73	000	000	00.0	00.0
16	3	0.000	16.80	57.38	500	069	61.4	42.4
16	4	1.712	07.98	63.54	000	000	00.0	00.0
17	1	0.000	09.00	54.70	412	058	71.6	43.0
17	2	1.391	05.08	57.04	000	000	00.0	00.0
17	3	0.000	10.00	54.75	420	063	70.0	42.6
17	4	1.316	06.16	57.13	000	000	00.0	00.0
18	1	0.000	19.70	51.90	565	075	59.9	38.8
18	2	1.280	12.26	56.80	000	000	00.0	00.0
18	3	0.000	21.10	51.89	573	076	58.9	38.7
18	4	1.109	15.36	55.66	000	000	00.0	00.0
19	1	0.000	13.80	57.56	498	061	64.5	43.1
19	2	2.459	12.76	58.26	000	000	00.0	00.0
19	3	0.000	14.20	58.02	497	065	64.0	43.3
19	4	2.277	14.55	57.84	000	000	00.0	00.0
20	1	0.000	14.30	73.66	374	051	61.3	52.7
20	2	1.531	14.15	73.78	000	000	00.0	00.0
20	3	0.000	16.30	72.89	388	055	59.4	51.7
20	4	1.557	13.49	75.33	000	000	00.0	00.0

RAW DATA (cont'd)

Subject	Test	RV	% BF	LBM	R	XC	%H_2O	LH_2O
21	1	0.000	11.60	60.92	444	060	66.4	45.8
21	2	1.444	14.56	58.90	000	000	00.0	00.0
21	3	0.000	13.00	60.78	455	071	64.9	45.3
21	4	1.334	15.81	58.80	000	000	00.0	00.0
22	1	0.000	16.40	74.71	413	052	59.1	52.8
22	2	1.499	16.64	74.48	000	000	00.0	00.0
22	3	0.000	18.20	74.16	425	058	57.4	52.1
22	4	1.507	16.52	75.72	000	000	00.0	00.0
23	1	0.000	09.60	76.48	379	056	66.7	56.4
23	2	2.416	05.75	79.72	000	000	00.0	00.0
23	3	0.000	10.60	76.66	385	063	65.3	56.0
23	4	2.437	05.62	80.90	000	000	00.0	00.0
24	1	0.000	14.80	64.96	440	069	62.0	47.3
24	2	1.729	08.56	69.77	000	000	00.0	00.0
24	3	0.000	17.50	63.82	463	077	59.5	46.1
24	4	2.022	07.36	71.74	000	000	00.0	00.0
25	1	0.000	13.10	59.78	490	070	65.0	44.7
25	2	1.515	05.50	65.03	000	000	00.0	00.0
25	3	0.000	13.80	60.06	493	074	64.0	44.6
25	4	1.307	07.51	64.49	000	000	00.0	00.0
26	1	0.000	12.00	59.92	471	069	66.2	45.1
26	2	2.067	09.50	61.67	000	000	00.0	00.0
26	3	0.000	13.50	59.74	483	073	64.4	44.5
26	4	2.024	10.38	61.88	000	000	00.0	00.0
27	1	0.000	20.50	75.34	416	063	55.3	52.5
27	2	1.432	19.29	76.59	000	000	00.0	00.0
27	3	0.000	22.10	74.75	425	070	54.2	52.0
27	4	1.272	20.56	76.20	000	000	00.0	00.0
28	1	0.000	11.10	78.72	434	062	65.7	50.8
28	2	2.874	01.94	75.82	000	000	00.0	00.0
28	3	0.000	11.70	69.31	434	065	65.0	51.0
28	4	2.665	03.45	75.75	000	000	00.0	00.0
29	1	0.000	10.60	61.46	471	061	67.6	46.5
29	2	1.908	04.46	65.76	000	000	00.0	00.0
29	3	0.000	10.90	61.78	471	065	67.2	46.6
29	4	1.273	08.98	63.16	000	000	00.0	00.0
30	1	0.000	15.30	66.91	439	064	61.2	48.4
30	2	1.496	16.49	66.00	000	000	00.0	00.0
30	3	0.000	15.90	67.40	439	065	60.6	48.6
30	4	1.384	15.82	67.48	000	000	00.0	00.0

APPENDIX I

PRE- AND POST-HYDRATION DESCRIPTIVE DATA

Descriptive Characteristics of Subjects (N = 30).

Subject	Age(yrs)	Wt(kg)	Ht(in)
01	21	80.24	73.75
02	19	58.14	69.25
03	21	68.86	69.25
04	24	57.94	64.25
05	23	75.86	72.75
06	21	63.73	68.50
07	22	59.64	62.50
08	20	78.93	75.25
09	21	61.69	69.50
10	20	68.27	71.50
11	23	74.05	69.25
12	19	77.68	75.75
13	20	81.19	73.50
14	20	69.06	69.25
15	21	74.84	73.00
16	20	68.15	68.75
17	21	60.10	64.75
18	18	64.75	68.50
19	24	66.79	70.10
20	19	85.96	68.50
21	21	68.95	69.25
22	22	89.36	71.50
23	24	84.60	73.00
24	22	76.32	69.50
25	25	68.83	71.25
26	23	68.15	70.50
27	22	94.92	70.50
28	23	77.34	73.00
29	19	68.83	72.25
30	21	79.04	70.25
Mean	21.3	72.41	70.30
Range	18-25	58.14-94.92	62.50-75.75

DESCRIPTIVE DATA
PRE- AND POST-HYDRATION EFFECT ON DRY WEIGHT (KG)

Subject	Pre-	Post-	Δ
01	80.24	82.78	2.54
02	58.14	59.19	1.05
03	68.86	70.08	1.22
04	57.94	59.08	1.14
05	75.89	76.88	1.02
06	63.73	64.52	0.79
07	59.64	60.55	0.91
08	78.93	80.06	1.13
09	61.69	62.71	1.02
10	68.27	69.29	1.02
11	74.05	74.96	0.91
12	77.68	78.70	1.02
13	81.19	82.21	1.02
14	69.06	70.19	1.13
15	74.84	75.86	1.02
16	68.15	69.06	0.91
17	60.10	61.35	1.25
18	64.75	65.77	1.02
19	66.79	67.70	0.91
20	85.96	87.09	1.13
21	68.95	69.85	0.90
22	89.36	90.72	1.36
23	84.60	85.73	1.13
24	76.32	77.45	1.13
25	68.83	69.74	0.91
26	68.15	69.06	0.91
27	94.92	95.94	1.02
28	77.34	78.47	1.13
29	68.83	69.40	0.57
30	79.04	80.17	1.13
Mean =	72.41 kg	73.49kg	1.08
Range =	57.94-94.92	59.08-95.94	

APPENDIX J

INITIAL PERCENT BODY FAT AS DETERMINED BY HYDROSTATIC WEIGHING

INITIAL % BODY FAT (% BF) AS DETERMINED BY HYDROSTATIC WEIGHING (HW)

Subject	% BF (Pre-Hydration)
01	8.81
02	11.21
03	16.37
04	14.98
05	4.37
06	11.79
07	13.08
08	11.85
09	2.85
10	7.96
11	6.48
12	12.82
13	12.34
14	9.00
15	8.58
16	7.94
17	5.08
18	12.26
19	12.76
20	14.15
21	14.56
22	16.64
23	5.75
24	8.56
25	5.50
26	6.31
27	19.21
28	1.94
29	4.45
30	16.49

Mean % BF = 10.25%

APPENDIX K

BIOELECTRICAL IMPEDANCE ANALYSIS RESPONSE TO OVERHYDRATION FOR PERCENT BODY FAT

BIA RESPONSE TO OVERHYDRATION FOR % BF

	% Body Fat		
Subject	Pre-Hydration	Post-Hydration	Difference
01	12.0	13.3	+1.3
02	10.4	10.9	+0.5
03	14.2	13.9	-0.3
04	3.1	9.9	+6.8
05	12.4	13.6	+1.2
06	13.8	14.5	+0.7
07	12.9	13.7	+0.8
08	11.3	12.2	+0.9
09	12.6	13.5	+0.9
10	12.9	13.8	+0.9
11	11.3	12.1	+0.8
12	14.3	15.7	+1.4
13	10.5	11.4	+0.9
14	16.9	18.6	+1.7
15	9.7	10.5	+0.8
16	16.4	16.8	+0.4
17	9.0	10.0	+1.0
18	19.7	21.1	+1.4
19	13.8	14.2	+0.4
20	14.3	16.3	+2.0
21	11.6	13.0	+1.4
22	16.4	18.2	+1.8
23	9.6	10.6	+1.0
24	14.8	17.5	+2.7
25	13.1	13.8	+0.7
26	12.0	13.5	+1.5
27	20.5	22.1	+1.6
28	11.1	11.7	+0.6
29	10.6	10.9	+0.3
30	15.3	15.9	+0.6

Mean Difference = 1.24%

APPENDIX L

HYDROSTATIC WEIGHING RESPONSE TO OVERHYDRATION FOR PERCENT BODY FAT

HW RESPONSE TO OVERHYDRATION FOR % BF

Subject	% Body Fat		
	Pre-Hydration	Post-Hydration	Difference
01	8.81	9.49	+0.68
02	11.21	10.80	-0.41
03	16.37	15.04	-1.33
04	14.98	14.68	-0.30
05	4.37	4.79	+0.42
06	11.79	11.94	+0.15
07	13.08	12.95	-0.13
08	11.85	9.44	-2.41
09	2.85	7.44	+4.59
10	7.96	10.41	+2.45
11	6.48	5.66	-0.82
12	12.82	12.51	-0.31
13	12.34	16.24	+3.90
14	9.01	10.49	+1.48
15	8.58	9.60	+1.02
16	7.94	7.98	+0.04
17	5.08	6.16	+1.08
18	12.26	15.36	+3.10
19	12.76	14.55	+1.79
20	14.15	13.49	-0.66
21	14.56	15.81	+1.25
22	16.64	16.52	-0.12
23	5.75	5.62	-0.13
24	8.56	7.36	-1.20
25	5.50	7.51	+2.01
26	6.31	10.38	+4.01
27	19.29	20.56	+1.27
28	1.94	3.45	+1.51
29	4.45	8.98	+4.53
30	16.49	15.82	-0.67

Mean Difference = 1.43%

APPENDIX M

BIOELECTRICAL IMPEDANCE ANALYSIS RESPONSE TO OVERHYDRATION FOR LEAN BODY MASS

BIA RESPONSE TO OVERHYDRATION FOR LBM

Subject	Lean Body Mass Pre-Hydration	 Post-Hydration	 Difference
01	71.71	71.76	+0.05
02	52.21	52.71	+0.50
03	59.97	60.37	+0.40
04	56.16	53.16	-3.00
05	66.41	66.41	0.00
06	54.93	55.16	+0.23
07	51.94	52.21	+0.27
08	69.99	70.26	+0.27
09	53.89	54.20	+0.31
10	59.47	59.65	+0.18
11	65.64	65.82	+0.18
12	66.50	66.32	-0.18
13	72.67	72.80	+0.13
14	57.34	57.11	-0.23
15	67.59	67.81	+0.22
16	56.97	57.38	+0.41
17	54.70	54.75	+0.05
18	51.98	51.89	-0.09
19	57.56	58.02	+0.46
20	73.66	72.89	-0.77
21	60.92	60.78	-0.14
22	74.71	74.16	-0.55
23	76.48	76.66	+0.18
24	64.96	63.82	-0.86
25	59.78	60.06	+0.14
26	59.92	59.74	-0.18
27	75.34	74.75	-0.59
28	68.72	69.31	+0.59
29	61.46	61.78	+0.32
30	66.91	67.40	+0.49
Mean	63.02	62.97	±0.40