Utilization Of B-Mode Ultrasound As A Body Fat Estimate In Collegiate Football Players

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The purpose of the present study was to validate a 7-site ultrasound imaging protocol to predict percent body fat (%BF) in a Division I football team. Body composition was estimated by ultrasound, seven site skinfolds (SKINFOLD), and the three compartment-water (3C-W) model of Siri (1961), using Bioimpedance spectroscopy (BIS) to estimate total body water (TBW) and air-displacement plethysmography (BODPOD®) to determine body density (Db). Pearson’s product-moment correlation analyses were run to determine between ΣUltrasound and the criterion 3C-W, and between the ΣSkinfold and ΣUltrasound. Strong positive correlations were observed between ΣSkinfold and ΣUltrasound (r=.984; p<.001). A Strong positive correlation was observed between ΣUltrasound and %BF from 3C-W (r=0.878, p<0.001). Based on the significant correlation analysis, a linear regression equation was developed to predict %BF from ΣUltrasound, (%BF= 6.194+(.096* ΣUltrasound); standard error of the estimate [SEE]=2.97%). Cross validation analyses were performed using an independent sample of 29 players. Mean observed %BF and mean predicted %BF were 18.32 ± 6.26% and 18.78 ± 6.22%, respectively. The constant error (CE), SEE and validity coefficient (r) were 0.004%, 2.64%, and 0.91, respectively. The total error (TEE) was 2.87%. Conclusion: The positive relationship between ultrasound measurements and the 3C-W model suggests the B-mode ultrasound may be a practical alternative of predicting %BF in Division I football players.

Key Words: 3-compartment model, validation, athlete, skinfold
UTILIZATION OF B-MODE ULTRASOUND AS A BODY FAT ESTIMATE IN COLLEGIATE FOOTBALL PLAYERS

by

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“If I have seen further than others, it by standing upon the shoulders of giants”

- Sir Isaac Newton
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INTRODUCTION

Body composition has been shown to be a significant predictor of performance in several tests for football athletes, including vertical jump and sprint performance (Miller, White, Kinley, Congleton, & Clark, 2002). Moreover, the ability of an individual to produce an isometric force is directly related to the muscle mass of that individual (Tonson, Ratel, Le Fur, Cozzone, & Bendahan, 2008). Previous research has shown that increased body mass as a result of increased fat free mass (FFM) has led to increases in performance of Division I football players (Noel, Vanheest, Zaneteas, & Rodgers, 2003a). Contrastingly, an increased body mass of football players as a result of increased fat mass and no increases in FFM will most likely result in performance decrements demonstrated by decreases in both speed and power, as well as increased risk of cardiovascular disease (CVD), stroke, and all-cause mortality (Lee, Blair, & Jackson, 1999; Miller et al., 2002; Noel, Vanheest, Zaneteas, & Rodgers, 2003a). Thus, the achievement and maintenance of an ideal body composition is important to the success and health of football athletes.

The estimation of body composition is a highly utilized practice by athletes and coaches alike (Moon et al., 2008). Body composition is often assessed using different laboratory measures. Dual energy x-ray absorptiometry (DEXA) uses a low dose x-ray radiation to determine the percent body fat (%BF) and bone mineral density (BMD) of the individual being tested. Although DEXA has been shown to be a valid measurement of body fat in athletes and nonathletes, its cost to purchase makes it impractical for the majority of athletes and coaches (Prior et al., 1997). Air Displacement Plethysmography (ADP) (most commonly via a BODPOD®) works by using the concepts of Boyle’s Law of displacement to estimate the body composition of the individual. While long considered the “gold standard”
in body composition, hydrostatic weighing (HW) may not be a feasible mode of body composition for as large of a team as commonly found in American football due to time requirements of the test. Consequently, the cost of equipment, potential size limitations, and time requirements for these tests make them inefficient for a large team such as American football.

Field measures for assessment of body composition are generally highly portable and more affordable. The use of field tests also allow for a more convenient method of taking repeated measurements of body composition over the course of an athletic season for both the athletes and investigators. Skinfold measurement is one viable option for field-testing body composition. When measured by a trained and experienced individual, skinfold thickness and the resulting estimation of body fat have a high degree of agreement with the associated multi-compartment criterion method (Evans, Rowe, Misic, & Prior, 2005). However, the use of skinfold measurements is much more difficult to obtain in overweight and obese subjects. This is primarily due to thicker adipose tissue making the proper isolation of a fold more difficult (Gray et al., 1990). Skinfold measurements are also limited by access to trained assessors, high inter-rater error, and an inability to palpate the adipose and muscular fascia border (Utter, McAnulty, Sarvazyan, Query, & Landram, 2010).

Ultrasound has been proposed as an alternative noninvasive technique to measure subcutaneous fat thickness. The utilization of a Brightness Mode (B-mode) ultrasound as a measure of body composition has been found to be a valid and reliable way to estimate body fat of an individual (Pineau, Filliard, & Bocquet, 2009; Wagner, 2013). Research has shown that in leaner, weight class athletes ultrasound imaging is a more accurate predictor of %BF than skinfold and bioelectrical impedance spectroscopy (BIS) when compared to
measurements obtained from DEXA (Pineau et al., 2009). B-mode ultrasound imaging
principally works by sending an acoustic wave from a transducer and interpreting the
reflection of the wave by a receiver, which is located within the transducer. These reflections
are interpreted by the machine and displayed as a 2-D image (Wagner, 2013). However,
previous research shows that a lack of cohesive standards for imaging sites exists (Wagner,
2013). Furthermore, the authors have noted a lack of predictive equations for athletic
populations to estimate the body composition of individuals using ultrasound. Nevertheless,
the ease of distinguishability of tissue planes and depth with on-screen calipers offer a
considerable advantage of skinfold measurements.

To the researchers knowledge, no studies have investigated the validity of a B-mode
ultrasound to estimate %BF in collegiate football players. Therefore, the purpose of this
study was to examine the relationship of a B-mode ultrasound, skinfold measurements, and a
criterion three compartment model of Siri (1956) and to develop a regression equation to
predict %BF from ultrasound measurements in collegiate football players.

METHODS

Experimental Approach to the Problem

A cross-sectional experimental design was applied to assess the body composition of a
Division I collegiate football team. A singular testing session included all of the body
composition measurements in the same order (BODPOD® (BP), BIS, skinfold, and
Ultrasound). BIS, skinfold, and ultrasound were each performed by the same technician to
eliminate inter-rater variability. The relationships between %BF from skinfold and %BF
from US, as well as the $\Sigma_{\text{Ultrasound}}$ and %BF 3C-W were assessed. A regression equation was then generated to predict %BF using $\Sigma_{\text{Ultrasound}}$.

Subjects
Fifty-eight collegiate Division I football players, including both African Americans [n=48 (Age (yr): 20.33 ± 1.24, weight (kg): 96.61 ± 19.14, height (cm): 179.71 ± 6.26)] and Caucasians [n=10 (age (yr): 20.10 ± 1.29, weight (kg): 100.76 ± 18.23 kg, height (cm): 182.63 ± 5.47)] (Table 1) volunteered to participate in this study approved by the Georgia Southern University Institutional Review Board. All participants provided written informed consent prior to participation. Participants were asked to arrive to the laboratory hydrated, in a fasted state (minimum of eight hours), refrain from caffeine consumption and to abstain from exercise 24 hours prior to testing. Water intake was allowed one hour prior to testing.

PROCEDURES

Total Body Water

BIS was used to estimate total body water (TBW) following the procedures recommended by the manufacturer (Bodystat Quadscan 4000: Bodystat LTD, Douglas, UK). This technique uses a range of frequencies (5KHz-200KHz), encompassing both low and high ranges that allow electrical current to pass around and through each cell. After resting in a supine position for 5 to 10 minutes, TBW estimates were taken while the participant lay in the supine position on a table with arms $\geq 30^\circ$ away from the torso and legs separated. Prior to each analysis, each participant’s height, weight, and gender were entered into the BIS device. Electrodes were placed at the wrist (dorsal surface at the ulnar styloid process) and ankle (dorsal surface between the malleoli) with additional electrodes being placed 5 centimeters

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distally from the wrist and ankle. Before electrode placement, excess body hair was removed, and the skin was cleaned with alcohol at each site. Multifrequency (5, 50, 100 and 200 kHz) currents were introduced from the positive leads and traveled throughout the body to the negative leads. Resistance values were used to calculate extracellular water (ECW) and intracellular water (ICW) and summed to equal TBW.

Air Displacement Plethysmography (BOD POD®)

Body density (Db) was estimated from air-displacement plethysmography using the BOD POD® (COSMED, Rome, Italy). Prior to each test, the BP was calibrated according to the manufacturer’s instructions using a two-point calibration. It was first calibrated with the chamber empty, and then with a cylinder of known volume (50.097 L). Prior to testing participants were instructed to wear tight fitting compression shorts and a swimming cap, and were asked to remove all metal, including jewelry and watches. Body mass was measured to the nearest 0.01 kg using the system’s calibrated scale. Participants were instructed to sit in the chamber, breath normally, but minimize any movement. A minimum of two trials were performed, and if measurements were not within 150 ml of each other, a third trial was conducted. Thoracic gas volume was estimated using the BP software, which uses standard prediction equations. It has previously been demonstrated that predicted lung volumes are not significantly different than measured volumes (McCrory, Molé, Gomez, Dewey, & Bernauer, 1998).
Three-Compartment Model (3C-W model)

The criterion %BF was estimated using the 3C-W model described by Siri (1956). The equation includes measurements of Db, TBW, and body mass (BM). The equation for percent body fat is listed below:

\[
\%BF = \left( \frac{2.118}{\text{Db}} - 0.78 \times \frac{\text{TBW}}{\text{BM (kg)}} - 1.354 \right) \times 100
\]

Skinfolds

Skinfold measurements were taken on the right side of the body with a calibrated Lange caliper at the following sites: chest, triceps, subscapular, midaxillary, abdomen, supraillium, and thigh. Skinfold measurements were made in duplicate at each site and recorded to the nearest 0.5mm, with a third measurement taken if the values differed by more than 2 mm (Brock, Nieman, Utter, Harris, & Rossi, 2009). All skinfold measurements were performed by a trained technician. Body density (Db) values were calculated using the generalized skinfold equation of Jackson et al (Chest, Midaxillary, Triceps, Subscapular, Suprailliac, Abdomen, Thigh) (1978). Percent body fat was calculated from Db using the formulas of Brozek and Schutte (Brožek, Grande, Anderson, & Keys, 1963; Schutte et al., 1984).

Ultrasound

Ultrasound measurements were taken using a Terason T3200 B-Mode device (Burlington, MA, USA) to measure subcutaneous fat thickness. B-mode ultrasound works by emitting ultrasound waves from the transducer head via piezoelectric crystals. As the ultrasound waves pass through the body tissue they are reflected back to the transducer head producing an image. If a tissue is denser with particles held closely together (i.e. fascia and bone) the image is reflected back white, if the particles are held loosely together (i.e. fluid)
and are less dense the image is reflected back black (Abu-Zidan, Hefny, & Corr, 2011). Measurements were taken on the right side of the body while the participant was standing using the seven-site skinfold locations according to Jackson and Pollock (1978). Measurements were made by applying transmission gel to the transducer and lightly placing the transducer parallel to the site. Care was taken to control the pressure of the transducer with minimal movement across the skin. The transducer was positioned so that a clear image was viewable on the monitor of the ultrasound. Once a clear image appeared, it was saved and labeled, and researchers progressed to the next site. At a later time point the researchers returned to the saved images to measure the thickness of the subcutaneous fat layer (Figure 1). Researchers calculated subcutaneous fat thickness using the electronic calipers associated with the T3200 software. Two measurements were taken for each site, with the average used for the final measurement. All seven values were summed.

Statistical Analysis

Twenty-nine football players were randomly selected from the pool of fifty-eight football players for the derivation of the prediction equation. Means for the groups can be found in Table 2. Pearson’s product-moment correlation analyses were run to determine the strength of the relationship between $\Sigma_{\text{Ultrasound}}$ and $\Sigma_{\text{Skinfold}}$, and the strength of relationship between $\Sigma_{\text{Ultrasound}}$ and the criterion %BF from 3C-W. A linear regression was used to generate a prediction equation for determining %BF using $\Sigma_{\text{Ultrasound}}$.

Cross-validation analysis of the new equation was conducted on the sample of 29 football players who were withheld from the derivation of the equation. Constant error (CE), total error (TE), correlation coefficient ($r$), and standard error of the estimate (SEE) were calculated. Correlation coefficients and bias $\pm$ 95% limits of agreement (as represented by
Bland-Altman plots) were used to assess the relationships between the criterion %BF and predicted %BF. SPSS Version 21 (IBM, New York, USA) was used for all statistical comparisons. The $\alpha$-level was set at $p \leq 0.05$ to determine statistical significance. All data are reported as mean ±SD.

RESULTS

A significant, positive relationship was observed between %BF from skinfold (Schutte) and %BF US (Schutte) ($r = .984; p<.05$) and %BF from skinfold (Brozek) and %BF from US (Brozek) ($r = .984; p<.001$). A statistically significant and positive correlation was observed between $\Sigma_{\text{Ultrasound}}$ and %BF from 3C-W ($r = 0.878, p<0.001$). Based on the significant correlation analysis, $\Sigma_{\text{Ultrasound}}$ was entered into a regression equation. The following equation was developed to predict %BF from $\Sigma_{\text{Ultrasound}}$:

$$\%BF = 0.096(\Sigma_{\text{Ultrasound}}) + 6.194; \text{ SEE} = 2.97\%$$

Based on the sample of 29 football players withheld from the derivation of the equation, the mean predicted %BF was 18.78% compared to the criterion %BF of 17.91% (Figure 2). The CE value of 0.004% was not significant ($p>.05$). The SEE and validity coefficient ($r$) were 2.64% and 0.91, respectively. The TE value was 2.87%. The associated Bland-Altman plot can be found in Figure 2.

DISCUSSION

The primary purpose of this study was to evaluate the use of B-mode ultrasound 7-site measurement as a predictor of %BF in collegiate football players. Our results indicate a significant positive correlation between %BF determined from skinfold and US and are in agreement with the findings of Fanelli and Kuczmarki (1984). Fanelli and Kuczmarki (1984) found that B-mode ultrasound produces %BF estimates similar to skinfold calipers in a non-
athletic, Caucasian population. This is in contrast to previous research which found that ultrasound was not a valid measure when compared to skinfold or 3C measurements (Loenneke et al., 2014; Smith-Ryan, Fultz, Melvin, Wingfield, & Woessner, 2014). However, these studies both used A mode ultrasound, which may explain the differences from our findings. Loenneke et al. (2014) obtained 1-site and 3-site measurements, while the current study utilized a 7-site measurement. The addition of the greater number of sites in the present study may have provided a more accurate representation of total body fat. While US and skinfold have been shown to have a high level of agreement, skinfold tends to overestimate %BF in individuals with higher levels of subcutaneous fat (Selkow, Pietrosimone, & Saliba, 2011). Furthermore, although skinfold measurements may be acceptable for tracking changes over time, higher degrees of inter-rater error reduce the likelihood that measures will be consistent when using skinfold unless the same person performs the measurements each time, as previously stated by Utter and Hager (2010). This may lead to inaccurate predictions about improvements by coaches or athletes.

The secondary purpose of this study was to develop a prediction equation that may be used to accurately predict %BF in collegiate football players via B-mode ultrasound use. The SEE from the produced regression equation was 2.97%, which indicates that the equation may be accurately used for prediction of %BF in this population. The findings of the present study are in agreement with those of Muller and colleagues (Müller et al., 2013), which found that B-mode ultrasound following the 10-site International Society of Advancement of Kinanthrometry (ISAK) standard was an accurate estimator of %BF in a healthy population. The results of our study support the use of a 7-site method as an alternative to the ISAK 10-site, in football athletes. In agreement to the present study, Fanelli and Kuczmarski (1984)
demonstrated that a nonsignificant difference existed between the criterion (hydrostatic weighing) results, and results of a predictive regression equation using ultrasound measurements, while the present study demonstrated a non-significant difference between criterion (3C-W) and the generated prediction equation. Smith-Ryan and colleagues (2014) found that ultrasound tended to underestimate %BF in overweight and obese men and women when compared to a 3-C W model. However, their study used A-mode ultrasound, which could potentially produce erroneous results in an overweight population due to changes in the pulse through the thicker adipose tissue (Smith-Ryan et al., 2014). Since %BF may be used as a predictor of performance in football players, the ability to consistently obtain accurate results is critical (Noel, Vanheest, Zaneteas, & Rodgers, 2003a).

Evaluation of the results of the cross validation analysis were established according to previous research, including the following criteria: (a) the mean values for observed and predicted %BF should be comparable; (b) the TE should be calculated because it reflects the true difference between the actual and predicted values for %BF, whereas the SEE only gives an indication about the error associated with the regression between the variables; (c) the TE and SEE should be similar because this reflects the relationship between the regression line for actual vs. predicted %BF and the line of identity; (d) a low SEE is preferred over correlation coefficients due to the SEE not being sensitive to differences in means and is affected by differences between samples in variability of %BF; and (e) there should be no relationship between the CE and %BF (Sinning et al., 1985). The SEE from the cross validation analysis of the current study was 2.64%, indicating minimal difference between observed %BF values and those predicted from the regression equation. The TE was 2.87%,
which indicates a strong relationship between observed and predicted values. The closeness of these values to each other and the non-significant CE value of 0.004% indicate that the associated regression equation is a valid measure for estimating %BF in collegiate football players.

Future research should aim to validate the developed equation in other populations such as high school football players and other collegiate athletes. Future studies should also aim to develop and validate standardized techniques for assessment of body composition using B-mode ultrasound since there is a current lack of research in this area.

PRACTICAL APPLICATIONS

The results of this study indicate that a 7-site B-mode ultrasound as a measure of %BF may produce results similar to skinfolds making it a cost effective, time efficient alternative to typical laboratory testing methods for coaches. Ultrasound offers other distinct advantages over skinfold measurements. The high degree of interrater error seen when using skinfold measurements may be reduced when using ultrasound imaging due to the ability to capture and save images (Utter et al., 2010). Additionally, since ultrasound does not require isolation of folds, it may be easier to measure the full thickness of adipose tissue.
References


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APPENDIX A

RESEARCH QUESTIONS, LIMITATIONS, DELIMITATIONS, ASSUMPTIONS, AND DEFINITIONS

Research Questions

1. Will a significant correlation exist between sum of ultrasound, and sum of skinfolds?

2. Will a significant correlation exist between 3C-W and sum of ultrasound?

3. Will a significant regression equation be able to be developed to predict 3C-W using the B-mode ultrasound measurements?

Limitations

1. A sample of convenience was utilized.

2. Generalizability to an athletic population, other than Division I football players, will difficult.

3. Urine specific gravity was not used to test hydration status

Delimitations

1. Exclusively male participants

2. Comparison of body composition measures is to a 3-CW model. It does not account for bone mineral density.

Assumptions

1. Each participant was fasted, and caffeine free for a period of at least eight hours.

2. Each participant was adequately and normally hydrated.

3. Each participant had not exercised for a period of at least 24-hours prior to testing.
APPENDIX B

Introduction:

Body composition testing provides an estimate of the approximate proportional constituents of the human body (i.e. adipose, muscle, bone). The following literature review will serve to provide a thematic synopsis of body composition testing in athletic populations, as well as to expound upon the general need for further research to examine the validity of ultrasonic body composition assessment practices.

Effects of Body Composition on Performance and Health

Estimations of body composition of an athletic population can be a useful tool for a myriad of reasons including: tracking progress across a competitive season, prediction of success in a game, and future health related outcomes. Miller and colleagues (2002) sought to evaluate the overall impact of body composition and body mass on anaerobic performance (power clean, bench press, squat, vertical jump, 40-yd dash, 20-yd shuttle). In general, significant performance decrements in regards to vertical jump, and power clean were observed when athletes had increased fat mass compared to fat free mass. Running performance of offensive linemen was inversely related to bodyweight and body composition. Tonson and colleagues (2008) evaluated young boys (n=14), adolescents (n=16) and adult men (n=16) to determine whether muscular volume was related to isometric force production capabilities. It was found that muscular volume was proportionally related to the ability of the individual to produce an isometric force. Thus, a greater muscle volume results in a greater ability to produce an isometric force. In a game-type scenario it is understood
that football players will have to elicit a high degree of isometric force in various situations (i.e. blocking, tackling, holding onto the ball).

Body composition measurements may also serve to elucidate previously unrecognized health risks, which is important not only for career longevity but also long term health of football players. Noel, Vanheest, Zaneteas, and Rodgers (2003a) evaluated Division I college football players (n=69) to determine if football players in the modern era are significantly bigger than their predecessors from 1980-1990. In line with their original hypothesis, Noel and colleagues (2003a) found that body mass, skinfold thickness and body fat percentage were significantly greater than players from previous decades. Furthermore, it was found offensive lineman, defensive lineman and tight ends had a mean body fat percentage of 25%. The researchers proposed that this increase in near-obesity level body fat percentages, and the region of adiposity might lead to long-term negative health consequences for the players. The findings of Miller et al. (2002) are in agreement with this higher body fat percentage of the “non skill-positions,” which exemplifies the importance of body composition testing of football players to provide not only performance data, but also more importantly disease risk stratification.

**Modes of Body Composition Assessment**

Body composition assessment strategies can be subdivided into three major categorical factions: 1) direct analysis, 2) clinical assessment methods, and 3) field assessment methods. Direct analysis of body composition can only be completed posthumously and requires painstaking precision. Due to the inability to complete direct analysis of body composition, estimative means of clinical and field assessment are required.
Clinical Body Composition Estimations

Clinical estimation of body composition is usually completed via dual energy x-ray absorptiometry (DEXA), air displacement plethysmography (ADP), hydrostatic weighing (HW) or multi-compartment model. Use of DEXA to estimate body composition, in a varied young adult population similar to our sample, is significantly correlated with a criterion method four-compartment model (4-C) (Prior et al., 1997). However, use of DEXA can be both cost-prohibitive and due to time requirements, inconvenient to test larger samples of individuals.

Alternative to the DEXA, clinical researchers can estimate body composition in collegiate football players using ADP (Collins et al., 1999). BodPod (CosMed, Rome, Italy) is the most common instrument utilized to estimate body composition via ADP techniques. Collins et al. (1999) compared BodPod to the consensus “gold-standard” HW and found that body density (Db) estimated via BodPod was consistently higher. Higher reported Db values, as assessed via BodPod, resulted in significantly lower %BF estimates as compared to HW, three compartment model (3C) and DEXA (Collins et al., 1999). Comparison of the Prior et al. (1997) and Collins et al (1999) studies demonstrate that DEXA may be the best choice for assessment of individual football players, completed in an independent, case study manner. However, as previously mentioned, DEXA tests have a longer time to completion than the counterpart BodPod. Due to the high degree of reliability associated with BodPod and timing constraints with sampling larger groups, such as a Division I football team, researchers may benefit from use of BodPod rather than DEXA.

Traditional body composition assessment techniques divided the body into two compartments (2C): fat mass (FM) and fat free mass (FFM). Closer inspection of the 2C
model reveals that anything not measured as FM would then be categorized as FFM, including but not limited to: muscle, skin, fluid, bones, and organs. The lack of inclusion of a direct measurement of the aforementioned variables results in an increased probability of incorrect estimation of body composition. This error is expected in result of an increased assumption of a uniform Db of a varied population. Siri (1956) first developed the three-compartment water (3CW) model of body fat estimation. This model further divided the FFM into fluid and non-fluid FFM, which allowed for a more accurate estimate of body density. Modern era is host to several different 3C models, of which 3CW and three-compartment bone mineral density (3CBMD) (1986) are the most prevalent. Andreoli and colleagues (2004) assessed the differences between 3CW and 3CBMD measurements in male water polo players (n=10). They found that while 3CW and 3CBMD were both significantly correlated with the criterion 4C model, 3CBMD had greater associated variability (Andreoli et al., 2004). Andreoli and colleagues (2004) suggest that when available, use of a 3CW model may provide a truer representation of body composition compared to 3CBMD (Andreoli et al., 2004). The findings of Andreoli (2004) and the use of 3C models are a superior estimation technique when compared to a 2C DEXA or other 2C measures.

Clinical methods to estimate body composition, while more accurate, are more invasive for the participant to complete. Additionally, clinical methods of assessment are relatively immovable and thusly must remain in a research or medical facility.
Field-based body composition tests provide researchers, clinicians and applied professionals the ability to quickly and accurately estimate body composition of large sample sizes. Furthermore, field tests are mobile and can be utilized in a setting removed from a research lab or medical setting. Body composition measures that can be categorized as field-based tests include but are not limited to the following: skinfold thickness assessment, bioelectrical impedance spectroscopy, and portable ultrasound.

Skinfold Assessment

Skinfold measures are assessed by: palpation of the adipose tissue, pinching of the subcutaneous adipose and subsequent measurement of the fold via calibrated calipers. Jackson & Pollack (1978) developed a series of skinfold measurement and associated equation that could be used to accurately estimate Db. Determined Db is then used, in conjunction with the estimation equation developed by Siri (Siri, 1961), to estimate %BF of the individual. Oppliger and colleagues (1992) found that skinfold measurements resulted in minor, non-significant, difference from HW when used with a collegiate football team. In agreement with the findings of Oppliger and colleagues (1992), Evans et al (2005) employed a similar skinfold methodology and found no significant differences between a 4C model and the skinfold mode. However, Evans et al (2005) sought to develop a newer, cross-validated, predictive equation specific to athletes. Similar to the current study, Evans and colleagues (2005) sampled collegiate athletes (n=132), and developed a novel predictive equation via cross-validation procedures to predict %BF without the need for the Siri (1961) equation. While it is not possible to directly compare the results of Evans (2005) to
Oppliger (1992), it is possible to extrapolate the accurate predictive capability of skinfold measure. Contrastingly, Gray et al. (1990) found that skinfold measurement underestimated body composition of participants, and was rendered useless in obese participants due to an inadequate size of calipers. As referenced earlier, Noel and colleagues (2003b) found that offensive lineman, defensive lineman and tight ends are near obesity level. Increased degrees of %BF in these, and other, positions may result in errant results as evidenced by Gray et al. (1990).

Bioelectrical Impedance Spectroscopy/ Bioelectrical Impedance Analysis

Bioelectrical impedance, another common field method for body composition estimation, works by sending an electrical impulse from one lead to another. This signal is then analyzed for time of travel, and strength of signal. Further differences exist between Bioelectrical Impedence Spectroscopy (BIS), and Bioelectrical Impedence Analysis (BIA). Those differences however are beyond the scope of this literature review. Gray and colleagues (1990) compared %BF estimations from BIA, skinfold and HW. It was found that BIA and skinfold both significantly underestimated the %BF of the overweight or obese individuals. Contrastingly, Oppliger et al. (1992) found that in a collegiate football specific population BIA overpredicted %BF of the sample as compared to criterion HW. In direct objection to the studies of both Oppliger (1992) and Gray (1990), Rutherford and colleagues (2011) found that BIA body fat estimations are not statistically dissimilar from those determined via HW.

In the current study BIS will be employed as a method for assessment of total body water, not an estimator of %BF. Use of BIS to assess TBW has been substantiated in the
body of published literature when compared to the criterion deuterium dilution method (Matias et al., 2013). Ellis et al (Ellis, Shypailo, & Wong, 1999) demonstrated that BIS has the ability to produce similar results for assessment of TBW as compared to a more traditional DEXA and total body potassium (TBK) assessment technique. These findings were later confirmed by Matias and colleagues (2013). The 2013 study conducted by Matias and colleagues (Matias et al., 2013) assessed TBW, extracellular water (ECW), and intracellular water (ICW) in elite athletes (n=62) during preseason training of the various sports. It was found that the results of BIS and deuterium dilution were not significantly different from one another.

**Ultrasound Imaging**

The central premise of the use of ultrasonic imaging (US) to assess %BF of an individual is not dissimilar from that of skinfolds. As described by Wagner et al. (2013), US emits an ultrasonic acoustic wave from piezoelectric crystals located within the head of the transducer. These waves then penetrate deep in the body. When the waveform is reflected back towards the transducer (a high density region), a white gradient is cast onto an external monitor. When the waveform is absorbed (a lower density region), a black gradient is reported. It is through varying degrees of reflection and absorption that a 2-dimensional image is able to be projected to the external monitor. Therefore, US should provide researchers and clinicians the ability to reduce inter-rater error observed as a result of compressive forces associated with skinfold measurements.

Pineau, Filliard & Bocquet (2009) compared the results of B-mode ultrasound imaging to DEXA in an athletic population (n=93). It was found that US and DEXA %BF
estimates were significantly correlated ($r=0.98$). In a cross-validation manner, Pineau and colleagues (2009) developed a predictive equation using the US measurements and other anthropometric data to accurately predict %BF of the individual as compared to DEXA. As documented previously in the Clinical Body Composition Estimation Section, DEXA has been found to be an accurate estimator of body composition when compared to other 2C models and a 4C model (1997) but when compared to a 3CW model, it was found to be less accurate (Andreoli et al., 2004). In agreement with the validity of US measurement of %BF, Fanelli and Kuczmarki (1984) found that US estimations in white males (n=124) demonstrated non-significant differences in estimation of Db from HW and adipose tissue thickness as assessed by skinfold measurement. Constrasting findings from Loenneke, Barnes & Waggenaer (2014) demonstrate that 3 site US measurements are not an accurate predictor of adiposity, or %BF of an individual. Loenneke and colleagues (2014) evaluated eleven male participants and utilized the Bodymetrix Pro BX2000 (Intelametrix, Livermore, California), an A-mode US, and the Bodymetrix proprietary %BF estimation equations. Both the findings and methodological analysis is in direct contrast to the study by Pineau et al (2009), who developed a sample specific predictive regression equation, rather than utilize the Bodymetrix proprietary equation. Furthermore, Loenekke and colleagues (2014) employed a 3-site model, as opposed to the 7-site model used by Pineau (2009). Smith-Ryan et al (2014) also found that US as an estimator of %BF was invalid for an overweight, or obese, population. While not a valid measurement of assessment for %BF, Smith-Ryan (2014) found that A-mode US was a reliable method for tracking changes in adiposity and thus postulated that it may be adequate for tracking changes across an intervention or period of time.
Conclusions/Recommendations:

In general, there exists a plethora of literature evaluating the multiple modes of body composition assessment. While clinical, or gold standard testing tends to be held to the highest regard, it is often unaffordable or too cumbersome to perform with a large sample size or athletic team. It is therefore necessary to further advance and evaluate the validity of field-based body composition estimation techniques. A paucity of published literature exists examining the validity of B-mode ultrasound as a body composition measure. The novel nature of this study will help to further the scientific body of literature focused on body composition evaluation techniques.
APPENDIX C

FIGURES AND TABLES

Figure 1. Measurement of subcutaneous adipose tissue
Figure 2. Bland-Altman plot of the difference between %BF measured by 3C-W and ultrasound. The light solid line indicates the line of best fit, the heavy solid line indicates the mean difference, and the dotted lines (mean difference ± 2SD) indicate upper and lower 95% limits of agreement.
Figure 3. Comparison of %BF determined by 3C-W and ultrasound in collegiate football players. Validity coefficient=0.91, SEE=2.64%.

<table>
<thead>
<tr>
<th>Variable</th>
<th>African American (n=48)</th>
<th>Caucasian (n=10)</th>
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<tbody>
<tr>
<td>Age (yr)</td>
<td>20.3 ± 1.2</td>
<td>20.1 ± 1.3</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>179.7 ± 6.3</td>
<td>182.6 ± 5.5</td>
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<tr>
<td>Weight (kg)</td>
<td>96.6 ± 19.1</td>
<td>100.8 ± 18.2</td>
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</tbody>
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Table 2. Descriptive characteristics of development and cross-validation groups (mean ± SD)

<table>
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<tr>
<th>Variable</th>
<th>Development Group (n=29)</th>
<th>Cross-Validation Group (n=29)</th>
<th>Combined (n=58)</th>
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</thead>
<tbody>
<tr>
<td>Age (yr)</td>
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<td>20.3 ± 1.2</td>
<td>20.3 ± 1.2</td>
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<tr>
<td>Height (cm)</td>
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<td>181.4 ± 5.0</td>
<td>180.2 ± 6.2</td>
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<tr>
<td>Weight (kg)</td>
<td>96.1 ± 19.62</td>
<td>98.6 ± 18.4</td>
<td>97.3 ± 18.9</td>
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