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Recommended Citation

Greer, B. K., Edsall, K. M., & Greer, A. E. (2015). Reliability of BOD POD measurements remain high following a short duration low-carbohydrate diet. *International Journal of Sport Nutrition & Exercise Metabolism*, 26(2), 145-149. doi:10.1123/ijsnem.2015-0184

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Note: This article will be published in a forthcoming issue of the *International Journal of Sport Nutrition and Exercise Metabolism.* This article appears here in its accepted, peer-reviewed form; it has not been copyedited, proofed, or formatted by the publisher.

Section: Original Research

Article Title: Reliability of BOD POD Measurements Remain High Following a Short Duration Low-Carbohydrate Diet

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Running Head: BOD POD Reliability

Journal: International Journal of Sport Nutrition and Exercise

Acceptance Date: September 16, 2015

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DOI: http://dx.doi.org/10.1123/ijsnem.2015-0184

ABSTRACT:

The purpose of the present study was to determine whether expected changes in body weight via a three day low-carbohydrate (CHO) diet will disrupt the reliability of air displacement plethysmography measurements via BOD POD. Twenty-four subjects recorded their typical diets for three days prior to BOD POD and seven-site skinfold analyses. Subjects were matched for lean body mass and divided into low-CHO (LC) and control (CON) groups. The LC group was given instruction intended to prevent over 50 grams/day of carbohydrate consumption for three consecutive days, while the CON group replicated their previously recorded diet. Body composition measurements were repeated post-dietary intervention. Test -retest reliability measures were significant (p < 0.01) and high for body fat percentage in both the LC and CON groups (r = 0.993, 0.965, respectively). Likewise, skinfold analysis for body fat percentage reliability was high in both groups (r = 0.996, 0.997, respectively). There were significant differences between first and second BOD POD measurements for body mass (72.9 ± 13.3 vs. 72.1 \pm 13.0 kg) and body volume (69.0 \pm 12.7 to 68.1 \pm 12.2 L) in the LC group (p < 0.05). However, there were no differences (p > 0.05) in BOD POD-determined body fat percentage, lean body mass, or fat mass between the first and second trial in either groups. Body composition measures via BOD POD and seven-site skinfolds remain reliable after three days of a LC diet despite significant decreases in body mass.

Keywords: plethysmography, body composition, ketogenic

INTRODUCTION

The use of low-carbohydrate (CHO = carbohydrate) diets experienced a popular resurgence starting in the mid-1990s among non-athletic populations, but they were also employed during investigations examining the ergogenic potential of "fat loading" (Burke & Hawley, 2002); interest in their chronic use for sport performance has recently reemerged (Noakes, Volek, & Phinney, 2014; Volek, Noakes, & Phinney, 2015).

Water losses account for a majority of weight loss observed in the initial weeks of a low-CHO diet (Freedman, King, & Kennedy, 2001). It is generally accepted that the majority of fluid losses in the first weeks of a low-CHO diet stem from the loss of muscle glycogen-bound water stores. Although such diets are typically not designed to be high protein diets, individuals often increase protein consumption in lieu of fat; Purkerson and Klahr report increased diuresis and electrolyte losses with higher protein diets (Purkerson & Klahr, 1984). In addition, dehydration is the most common, albeit acute, symptom of very low-CHO (ketogenic) diets in epileptics (Kang, Chung, Kim, & Kim, 2004).

Dehydration significantly lowers body fat percentage determinations made via hydrodensitometry and bioelectrical impedance (Thompson et al., 1991). A concern exists that the rapid loss of water weight observed with low-CHO diets (Golay et al., 1996), regardless of clinically relevant dehydration, will affect the reliability of air displacement plethysmography and consequently bias conclusions regarding diet efficacy as it relates to body composition. BOD POD (Life Measurement Instruments, Concord CA) is the sole commercially available product using air displacement plethysmography for adult densitometry determination. BOD

POD operates on a similar principle as hydrostatic weighing with the substitution of air displacement for water displacement (Dempster & Aitkens, 1995). Both the validity and reliability of air displacement plethysmography via BOD POD for determination of body composition have been established (Noreen & Lemon, 2006; Tseh, Caputo, & Keefer, 2010).

Pilot data from our laboratory (unpublished) showed large decreases in body fat percentage following a three-day very low-CHO (< 50 g/day) diet. Studies investigating low-CHO diet efficacy have relied on the BOD POD for outcome measurements (Hays et al., 2004; Johnstone, Horgan, Murison, Bremner, & Lobley, 2008), and consequently their conclusions may be called into question if BOD POD measures are shown to be unreliable with similar dietary patterns. Additionally, if densitometry methodology reliability is compromised with short-term use of low-CHO diets, less accurate methods such as skinfolding that are assumed to be unaffected by hydration status may be more appropriate for research or practitioner use.

To date, no study has investigated the acute influence of diet on BOD POD measurement reliability, and the renewed interest in low-CHO diets among athletic populations provides the rationale for such an investigation. The primary hypothesis of the present study was that a three-day low-CHO diet will disrupt BOD POD-derived body fat percentage reliability due to measurement alterations in lean body mass, whereas body fat percentage determined by skinfold analysis will be unaffected. Body composition measures are almost universally employed immediately before and after a dietary intervention; if the hypothesis were accepted, investigators of long term low-CHO diets may wish to employ additional air displacement plethysmography measures immediately after the brief period of rapid weight/fluid loss in

order to attenuate reliability disruptions caused by alterations in fluid rather than tissue volume.

METHODS

Subjects

A priori power analysis based upon pilot data indicated a minimum of six subjects per condition for a power > 0.8 in regards to body fat percentage and lean body mass measures via BOD POD. Consequently, 24 subjects (32 ± 12.5 yrs), or double the predicted minimum including a control group, were recruited. Both male and female subjects were recruited as no evidence suggests that gender affects BOD POD reliability; as the intervention was only three days, menstrual cycle status was not collected. Methods were approved by the Institutional Review Board at Sacred Heart University and all subjects granted informed consent.

Experimental Procedures

Subjects recorded their diet for the three days prior to initial body composition analysis (Phase 1); three day dietary records are a valid dietary assessment tool (Yang et al., 2010). Dietary data were analyzed using NutritionCalc Plus 2.0 (Salem, OR), a commercially available dietary software. Kilocalorie (kcals/day), macronutrients (g/day and % of total kcals), sodium (mg/day), and potassium (mg/day) intakes were recorded. Sodium and potassium were included due to their influences on fluid balance.

The day following the Phase 1 dietary recording period, subjects reported to the lab between 6 a.m. and 11:00 a.m. having abstained from food or drink for at least three hours. All BOD POD manufacturer instructions were followed in regards to proper attire and procedures.

Function residual capacity was measured (as opposed to estimated) using the BOD POD system, a valid and reliable method for determining lung volumes (Davis et al., 2007). Body fat percentage, fat free mass (kg), fat mass (kg), body mass (kg), and body volume (L) were recorded. After BOD POD measurements were taken, a 7-site skinfold analysis was performed following Jackson-Pollack procedures and equations for determination of body density and body fat percentage (Jackson, 1985). All skinfold measurements were made by the same tester who had 12 years of testing and teaching experience and had passed a practical examination of skinfolding skill included in a certification previously offered by the American College of Sports Medicine.

Subjects were divided into low-CHO (LC = low carbohydrate) and control (CON = control) groups. Groups were matched for fat free mass (determined by the BOD POD) as this variable experienced the largest change during pilot data collection. CON subjects were instructed to replicate their previously recorded diet for three days immediately following body composition testing (Phase 2). The LC group was provided instructions intended to keep carbohydrate intake under 50 grams/day for the three day period; this included exclusion of grains, legumes, tubers, fruit, and non-fermented dairy from the diet. A three day intervention was chosen as it is long enough to cause changes in fluid balance (Purkerson & Klahr, 1984) but short enough to not induce practically important changes in fat mass under ad libitum feeding conditions.

Body composition measurements were repeated the day immediately following the three-day Phase 2 dietary intervention. Pre-test skinfold data were not reviewed prior to posttesting in order to eliminate measurement bias. Again, subjects reported to the lab between 6

a.m. and 11:00 a.m. having abstained from food or drink for at least three hours. For these post-test procedures, subjects were also instructed to replicate any food or drink that was consumed on testing days prior to the required three hour fast.

Statistical Analysis

All data were tested for normal distribution via a Shapiro-Wilk test. Consequently, a paired-sample Wilcoxon signed rank test was used for these non-parametric analyses. Dietary intake that occurred on body composition testing days was excluded from analysis as the majority of subjects (22 of 24) remained fasted from the night prior. Pearson product-moment correlation coefficients (*r*) were calculated for an index of test-retest reliability for body composition measures. All body composition variables (which were distributed normally) were treated by a paired sample t-test in order to determine if any significant change occurred between measurements. The level of significance for both parametric and non-parametric tests was set at 0.05. Coefficients of variation (CV) for repeated measures were calculated as $(SD_x/Y) \times 100$, where SD_x represents the mean standard deviation of repeated trials, and Y represents the mean across repeated trials.

RESULTS

All 24 subjects recruited for the study completed all testing procedures and provided the required dietary recalls. There were no significant pre to post dietary changes within the CON group. As expected, CHO intake in grams/day as well as percentage of total kilocalories fell significantly (p < 0.05) in the LC group, and there was an increase in fat grams/day as well as fat as a percentage of total kilocalories (p < 0.05). Both total protein and protein as a

percentage of kilocalories were significantly increased in the LC group (p < 0.05), with no additional statistically significant changes occurring. Dietary variables can be seen in Table 1.

There were no pre to post differences in either group for percent body fat, fat-free mass, or fat mass via BOD POD (p > 0.05). Percent body fat via skinfolding also did not change in either group (p > 0.05). Both body mass and body volume decreased in the LC group (p < 0.05). Reliability for all body composition measurements remained very high and statically significant (p < 0.01) in either dietary condition. All body composition data can be seen in Table 2.

DISCUSSION

The primary finding of the present study is that reliability of anthropometric measurements via BOD POD or skinfolds remain very high ($r \ge 0.993$ for all measures) after three days of a low-CHO diet. BOD POD reliability in the CON group is similar to previously reported data without dietary intervention (Tucker, Lecheminant, & Bailey, 2014).

The dietary invention was marginally successful as the mean daily CHO intake for Phase 2 in the LC group was 54.6 \pm 26.3 grams with an initial goal of < 50 grams. Even though the mean CHO intake was approximately 5 grams above the intended maximum, three days was still long enough in duration to induce a significant loss of body mass and volume in the LC group. These results provide support for the study protocol.

Percent body fat is derived from body density, and body density is equivalent to body mass divided by body volume. As such, % body fat via BOD POD went unchanged in the LC group despite a statistically significant drop in body mass as there was a concurrent drop in

body volume (69.0 \pm 12.7 to 68.1 \pm 12.2 L). We hypothesize that the absence of fibrous foods (e.g., fruits, legumes, whole grains) during the LC trial reduced the volume of air in the gastrointestinal tract and consequently the degree to which the abdominal wall is pushed anteriorly. Plausibility of this contention is bolstered by evidence that the BOD POD can detect minor volume changes solely caused by changes in body position (Peeters, 2012). It should be noted that despite the significant drop in body mass within the LC group, pre to post body mass reliability remained significant and quite high, partially due to the consistent standard deviation.

Seven-site skinfold reliability (0.996 and 0.997 for the VLD and CON groups, respectively) is higher than previously reported in comparable populations (Rouwmaat, Everaert, Stappaerts, & Aufdemkampe, 1998) but similar to those made in military personnel of both genders (Aandstad, Holtberget, Hageberg, Holme, & Anderssen, 2014). Higher skinfold reliability than typically reported was expected due to the experience of the tester, the use of a single tester(Kispert & Merrifield, 1987), and use of Jackson-Pollack equations which were recently reported to have a smaller test-retest measurement error than alternative prediction equations (Aandstad et al., 2014).

Given that the LC group lost over 1% of their initial body mass over just three days, it may be prudent for long-term studies involving CHO manipulation for body weight loss to measure and report weight losses after 1-2 weeks in addition to post-test measures (typically at 6 or 12 months). This will help in distinguishing between weight reduction predominantly from water loss that could be rapidly regained via glycogen replenishment (Sherman et al., 1982) and from actual loss of non-CHO based stored energy.

There has been some concern regarding the potential cardiovascular risk with low-CHO diets (Fung et al., 2010; Lagiou et al., 2012; Ornish, 2004). Many of these concerns refer to the pro-atherosclerotic effect of saturated fats, a contention sometimes contested (Siri-Tarino, Sun, Hu, & Krauss, 2010). Although not included in the analysis as dietary fat type would not influence fluid balance, saturated fat intake only increased by 2.6 grams/day in the LC group, providing a most likely negligible increase in cardiovascular risk. This is not an abnormal finding as individuals often reduce the intake of certain types of fat when the carbohydrate carrier (e.g., pasta, bread) is eliminated from the diet (Ornish, 2004).

In conclusion, the primary hypothesis of the present study was rejected as body composition measures determined by both BOD POD and skinfold techniques had remarkably high reliability following a three day low-CHO diet. Given that body mass but not percent body fat was significantly reduced in the experimental group, practitioners may wish include anthropometric measurements beyond simply bodyweight for outcome variables related to low-CHO interventions.

Acknowledgements

None.

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	Kilocalories		Carbohyd	drates (g)	Carbohydrates (% of kilocalories)		
	Phase 1 Phase 2		Phase 1 Phase 2		Phase 1	Phase 2	
LC	2069 ± 880	1906 ± 686	244.5 ± 96.2	54.6 ± 26.3*	45.6 ± 6.3	12.6 ± 6.7*	
CON	2595 ± 1012	2441 ± 937	256.2 ± 149.7	266.5 ± 161.3	38.3 ± 14.7	41.8 ± 15.7	

	Fats (g)		Fats (% of	kilocalories)	Protein (g)		
	Phase 1	Phase 2	Phase 1	Phase 2	Phase 1	Phase 2	
LC	87.1 ± 47.7	119.8 ± 47.6*	35.3 ± 8.2	55.3 ± 7.7*	92.7 ± 30.8	148.2 ± 69.0*	
CON	101.1 ± 47.3	87.7 ± 42.6	33.8 ± 7.2	31.3 ± 8.0	165.3 ± 80.3	153.3 ± 78.6	

	Protein (% of kilocalories)		Sodiur	m (mg)	Potassium (mg)		
	Phase 1 Phase 2		Phase 1	Phase 2	Phase 1	Phase 2	
LC	18.3 ± 4.2	31.8 ± 6.8*	3851 ± 1691	4083 ± 1387	2976 ± 1353	2786 ± 886	
CON	27.1 ± 13.0	26.6 ± 13.3	4102 ± 2040	4146 ± 1766	3373 ± 1275	3256 ± 1393	

* Significantly different than Pre (p < 0.05)

LC = low-carbohydrate group, CON = control group

	Body Fat via BOD POD (%)				Fat Free Mass (kg)				Fat Mass (kg)			
	Pre	Post	r	CV	Pre	Post	r	CV	Pre	Post	r	CV
LC	19.7 ± 8.8	19.2 ± 7.9	0.993*	7.12%	58.7 ± 13.0	58.4 ± 12.7	0.997*	1.67%	14.3 ± 7.2	13.7 ± 6.8	0.995*	8.92%
CON	23.2 ± 6.4	22.7 ± 6.2	0.965*	7.26%	58.9 ± 17.8	59.3 ± 18.2	0.997*	2.36%	19.2 ± 11.1	18.8 ± 10.7	0.995*	6.16%
	Body Mass (kg)				Body Volume (L)				Body Fat via 7-site Skinfolds (%)			
	Pre	Post	r	CV	Pre	Post	r	CV	Pre	Post	r	CV
LC	72.9 ± 13.3	72.1 ± 13.0^	0.999*	1.01%	69.0 ± 12.7	68.1 ± 12.2^	0.998*	1.28%	14.9 ± 7.5	14.5 ± 7.6	0.996*	4.57%
CON	78.1 ± 28.0	78.1 ± 28.1	1.000*	0.85%	74.7 ± 27.6	74.6 ± 27.5	1.000*	0.84%	20.7 ± 7.4	20.6 ± 7.8	0.997*	3.46%

* Statistically significant (p < 0.01)

^ Significantly different than Pre (p < 0.01)

LC = low-carbohydrate group, CON = control group, *r* = Pearson's product-moment correlation coefficient, CV = coefficient of variation