



Effects of a low-carbohydrate ketogenic diet on health parameters in resistance-trained women

Salvador Vargas-Molina^{1,2} · Leandro Carbone³ · Ramón Romance⁴ · Jorge L. Petro⁵ · Brad J. Schoenfeld⁶ · Richard B. Kreider⁷ · Diego A. Bonilla^{8,9} · Javier Benítez-Porres¹

Received: 7 June 2020 / Accepted: 29 April 2021 / Published online: 18 May 2021
© The Author(s), under exclusive licence to Springer-Verlag GmbH Germany, part of Springer Nature 2021

Abstract

Purpose The aim of this study was to evaluate the effect of a ketogenic diet on blood pressure, visceral adipose tissue (VAT), bone mineral content (BMC), and bone mineral density (BMD) in trained women.

Methods Twenty-one resistance-trained women performed an 8-week resistance training program after a 3-week familiarization phase. Participants were randomly assigned to a non-ketogenic diet ($n = 11$, NKD) or ketogenic diet ($n = 10$, KD) group. Health parameters were measured before and after the nutritional intervention. Blood pressure was measured using a digital automatic monitor, while VAT, BMC, and BMD changes were measured by dual-energy X-ray absorptiometry.

Results There was a significant reduction in systolic blood pressure in KD (mean \pm SD [IC 95%], P value, Hedges' g ; -6.3 ± 6.0 [$-10.5, -2.0$] mmHg, $P = 0.009$, $g = -0.81$) but not in NKD (-0.4 ± 8.9 [$-6.8, 6.0$] mmHg, $P = 0.890$, $g = -0.04$). The results on VAT showed no changes in both groups. The KD showed a small favorable effect on BMD (0.02 ± 0.02 [$0.01, 0.03$] $\text{g}\cdot\text{cm}^{-2}$, $P = 0.014$, $g = 0.19$) while NKD did not show significant changes (0.00 ± 0.02 [$-0.02, 0.02$] $\text{g}\cdot\text{cm}^{-2}$, $P = 0.886$, $g = 0.01$). No differences in *group* or in the *time* \times *group* interaction were found in any of the variables.

Conclusions Consuming a low-carbohydrate high-fat KD in conjunction with a resistance training program might help to promote the improvement of health-related markers in resistance-trained women. Long-term studies are required to evaluate the superiority of a KD in comparison to a traditional diet.

Keywords Bone mineral density · Blood pressure · Carbohydrate-restricted diet · Cardiovascular health · Female · Visceral fat

Abbreviations

KD Ketogenic diet
NDK Non-ketogenic diet
KB Ketone bodies

VAT Visceral adipose tissue
BP Blood pressure
HDL High-density lipoprotein
LDL Low-density lipoprotein
FM Fat mass
BMC Bone mineral content

Communicated by Kirsty Elliott sale.

✉ Salvador Vargas-Molina
salvadorvargasmolina@gmail.com

- ¹ Physical Education and Sports, Faculty of Medicine, University of Málaga, Málaga, Spain
- ² EADE-University of Wales Trinity Saint David, Málaga, Spain
- ³ University of Salvador, Buenos Aires, Argentina
- ⁴ Body Composition and Biodynamic Laboratory, Faculty of Education Sciences, University of Málaga, Málaga, Spain
- ⁵ Research Group in Physical Activity, Sports and Health Sciences (GICAFS), Universidad de Córdoba, Montería, Colombia

- ⁶ Department of Health Sciences, CUNY Lehman College, New York, NY, USA
- ⁷ Exercise and Sport Nutrition Lab, Human Clinical Research Facility, Texas A&M University, Texas, USA
- ⁸ Research Division, Dynamical Business and Science Society, DBSS INTERNATIONAL SAS, Bogotá, Colombia
- ⁹ Research Group in Biochemistry and Molecular Biology, Universidad Distrital Francisco José de Caldas, Bogotá, Colombia

| | |
|-----|----------------------------------|
| BMD | Bone mineral density |
| RT | Resistance training |
| FFM | Fat free mass |
| BMI | Body mass index |
| DXA | Dual-energy X-ray absorptiometry |
| LBM | Lean body mass |
| GLM | General linear model |
| ES | Effect size |
| SBP | Systolic blood pressure |
| DBP | Diastolic blood pressure |

Introduction

Research shows that dietary manipulation in conjunction with regimented physical exercise is an effective tool for improving body composition, health, and athletic performance (Burke 2015; Bartlett et al. 2015). While different strategies have been promoted for manipulating nutritional variables, the ketogenic diet (KD), a subtype of diets low in carbohydrates and high in lipids, has emerged as a popular option both in clinical and sports settings due to its capacity for improving body composition (Vargas et al. 2018; Gregory 2017; Zinn et al. 2017), variables related to athletic performance (Egan and D'Agostino 2016; Zajac et al. 2014; Phinney 2004), and markers of cardiovascular and metabolic health (Kosinski and Jornayvaz 2017; Francois et al. 2017).

Ketogenic diets are usually characterized by provision of less than 20% of daily caloric intake in the form of carbohydrates, more than 50% from lipid and a moderate but variable amount of protein. (Zajac et al. 2014; McArtney et al. 2017; Phinney 2004). This type of distribution of macronutrients presents physiological changes related to the use of energetic substrates, theoretically preserving the use of glycogen and lean tissue proteins, increasing the oxidation of fatty acids, and generating a marked elevation of plasma ketone bodies (KB), such as acetate, acetone, and β -hydroxybutyrate (Stubbs et al. 2017; Chang et al. 2017).

Under standard dietary conditions, the concentration of circulating KB, especially β -hydroxybutyrate, is approximately 3 mM. However, in the context of nutritional ketosis (Puchalska and Crawford 2017; Veech 2004), prolonged fasting (Cahill 1970, 2006), or strenuous exercise (Koeslag et al. 1980), ketonemia reaches 7–8 mM, which does not generate pathological changes, such as those changes observed on blood pH under ketoacidosis conditions, which are associated with diabetic patients (where ketonemia can reach 20 mM) (Puchalska and Crawford 2017). While their main function is to serve as an energy substrate, KB are purported to possess potential metabolic benefits (Puchalska and Crawford 2017).

In addition, KD has been implemented as a dietary intervention to achieve greater long-term reductions in

body weight, in comparison to low-fat diets (Bueno et al. 2013). Emerging evidence has suggested KDs may have potential clinical uses in neurodegenerative diseases (Włodarek 2019; Veech 2004), as a therapeutic aid in some types of cancer (Allen et al. 2014), and for the treatment of risk factors to prevent cardiovascular diseases, such as blood pressure (BP), high-density lipoprotein (HDL), low-density lipoprotein (LDL), and blood triglycerides (Cotter et al. 2013; Kosinski and Jornayvaz 2017; Hu et al. 2015). In particular, a KD has been demonstrated as an effective strategy to reduce systolic blood pressure (SBP) (Cicero et al. 2015; Castellana et al. 2020) and visceral adipose tissue (VAT) (Valenzano et al. 2019) in overweight and obese subjects; however, less data are available in trained subjects regarding these cardiometabolic risk factors. We previously demonstrated that a KD might be an alternative dietary approach to decrease fat mass (FM) and visceral adipose tissue (VAT) while preserving fat-free mass in resistance-trained men (Vargas et al. 2018); however, further research is needed in trained women since between-sex differences in energy substrate metabolism has been reported at given exercise intensities during a KD (Durkalec-Michalski et al. 2019).

On the other hand, although some randomized controlled studies have shown that a short-term KD does not cause negative changes in bone mineral content (BMC) in healthy obese adults (Colica et al. 2017; Perissiou et al. 2020), it is important to note that the KD might impair skeletal health parameters in growth children treated with this low-carbohydrate diet intervention (Simm et al. 2017; Draaisma et al. 2019). In fact, there have been reports that KD cause deleterious effects on sports performance and athletes' health parameters, which have been attributed to fatigue, dehydration, hypoglycaemia, increased risk of kidney stones, hyperlipidaemia, anemia, leukopenia, and deficiencies in certain vitamins and other trace elements such as zinc, selenium, and magnesium (Hartman and Vining 2007). Recent research indicates that a KD might impair markers of bone modeling/remodeling (Heikura et al. 2019), training adaptations (McKay et al. 2019; Heikura et al. 2019; Wroble et al. 2019), or have an unfavorable effect on muscle fatigue and perceived exertion during daily life activities in women (Sjödén et al. 2020). Thus, further research is warranted to draw better insights into the potential benefits and detriments of the short-term KD from a health-related standpoint.

The present study aimed to evaluate the effects of an eight-week low-carbohydrate KD intervention in combination with resistance training (RT) on cardiometabolic risk factors and bone health parameters in healthy young resistance-trained women. We hypothesized that a KD in conjunction with RT would positively improve BP and VAT without adversely affecting bone health-related parameters after the intervention.

Methods

Participants

A convenience sample of 21 women (age = 27.6 ± 4.0 years; height = 162.1 ± 6.6 cm; body mass = 62.3 ± 7.8 kg; BMI = 23.7 ± 2.9 kg·m⁻²) with over two years of continuous RT experience volunteered to participate in the study. Participants gave their written informed consent after being informed of the possible risks of the experiment. The study was designed in accordance with the ethical guidelines of the Declaration of Helsinki (WMA 2013) and approved by the local university ethics committee (code: 38-2019-H). Individuals admitting to administration of doping agents (e.g., anabolic–androgenic steroids) during the last two years or to taking any dietary supplement during the study period were excluded from participation. Moreover, women with oligomenorrhea, polycystic ovarian syndrome, or those outside the required age range of 18–35 years were also excluded. Participants were instructed to avoid performing any structured exercise during the study period other than that prescribed for the intervention. Figure 1 displays a flowchart of the data collection process as per the Consolidated Standards of Reporting Trials (CONSORT).

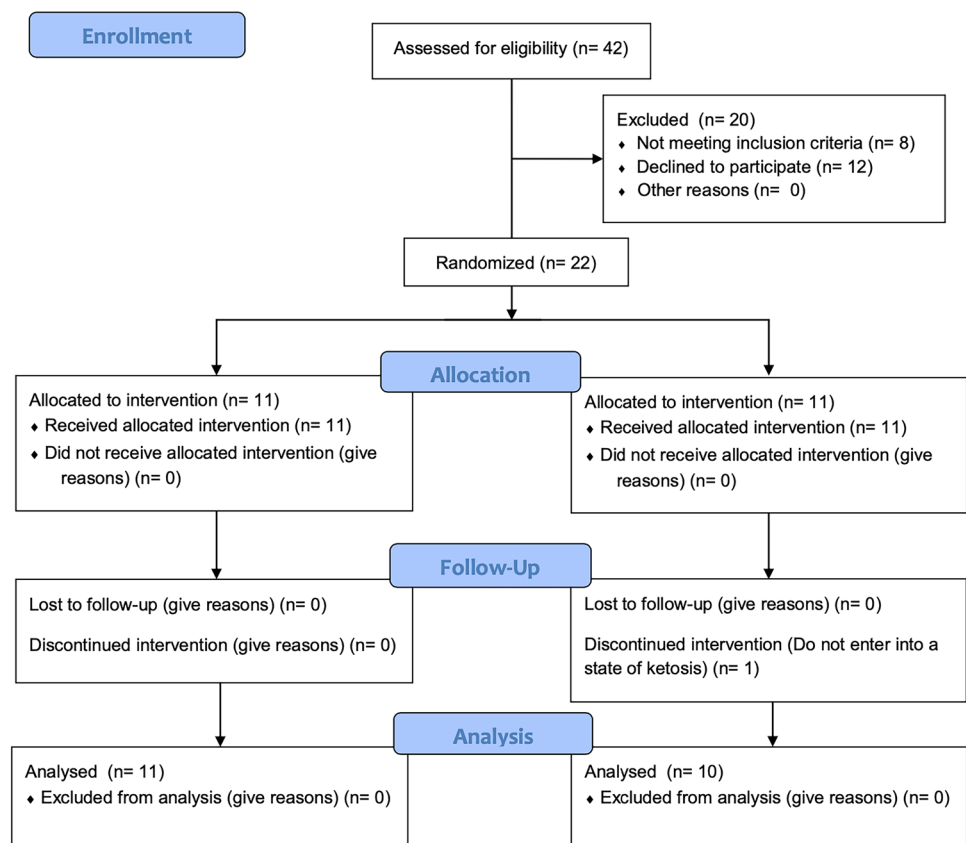
Procedures

Participants were randomly assigned to either the KD group ($n = 10$) or the NKD group ($n = 11$) and, after a three-week familiarization phase, they engaged in an eight-week training and diet intervention. We recorded all loads for both the familiarization phase and the intervention. The KD group initially had 11 participants but one subject was expelled from the study after the first week due to a negative test on the ketosis strips.

Dietary intervention

Diets for each respective group were designed so that participants maintained a non-negative energy balance. The KD group was instructed to consume a low-carbohydrate high-fat diet with moderate protein intake, while the NKD was prescribed a traditional diet. Both diets were similar in terms of energy intake and dietary protein, as far as possible. Nutrition plans were individually customized based on the participants' FFM (g·kg-FFM⁻¹·d⁻¹) as obtained via dual-energy X-ray absorptiometry (DXA), taking into account the fact that they had RT experience and possessed relatively low levels of body fat. To avoid low energy availability and consequent changes in their menstrual cycles, the participants' energy intake was set at ≈ 40 – 45 kcal·kg-FFM⁻¹·d⁻¹,

Fig. 1 CONSORT diagram



which is higher than in some previous studies (30 kcal·kg⁻¹ LBM) (Loucks 2003; Reed et al. 2015).

The KD diet comprised > 1.7 g·kg⁻¹·d⁻¹ of protein, between 30 and 40 g·d⁻¹ of carbohydrates, and the remaining calories were obtained in the form of dietary fats (see details in Table 1). Participants were advised to eat three to six meals per day to facilitate adherence to the program. To determine compliance with the KD, participants self-tested for the presence of acetoacetate and acetone in urine using the nitroprusside reaction on diagnostic strips (Ketostix, Bayer Vital GmbH, Leverkusen, Germany). Participants were instructed to wash their hands, immerse the absorptive end of the strip through the urine stream, and wait for 15 s after the reaction to report the color. This qualitative assessment of ketosis state was performed weekly in the early morning throughout the study in the KD group, according to previous recommendations (Urbain and Bertz 2016). The NKD group was instructed to consume > 1.7 g·kg⁻¹·d⁻¹ of protein as a function of the menstrual cycle in the luteal phase (Wooding et al. 2017), 1 g·kg⁻¹·d⁻¹ of fat, with the remaining energy intake obtained from carbohydrates (see Table 1).

To monitor dietary intake, participants recorded their daily macronutrient intake via a smartphone app (MyFitnessPal, LLC, CA, USA), which has been validated as viable tool for energy and macronutrient assessment (Teixeira et al. 2018). A sports nutritionist with experience in RT instructed participants on proper use of the app and managed participants' dietary consumption over the course of the study.

Exercise protocol

Participants completed a three-week familiarization phase designed to determine initial loads for each exercise, and

then engaged in an eight-week training program. Both groups performed a total body routine that included the following exercises: bench press, barbell row, military press, lat pulldown, incline chest press, biceps curl, triceps pushdown, back squat, lunge, leg press, hip thrust, leg extension, lying leg curl, and standing calf raise. Repetitions were controlled by a metronome at the tempo prescribed for the given training micro-cycle (Metronome M1, JSplash Apps).

After familiarization, participants performed an upper/lower split routine that encompassed four weekly training sessions (divided into two four-week cycles) for eight weeks. A 72-h recovery period was afforded between sessions for the same muscle complex. The programs followed a nonlinear periodized model that included three targeted phases as follows: strength, hypertrophy, and muscular endurance. To facilitate recovery, a deload week of reduced volume was employed in the last week of each cycle. In total, participants performed two, four-week cycles with variables manipulated as detailed in Fig. 2.

Loads were modified in both the hypertrophy and muscular endurance phases for each set based on participants' perceived exertion and the number of repetitions achieved to ensure concentric failure within the target repetition range. Alternatively, participants stopped 1–2 repetitions short of failure during the strength micro-cycle. Participants attempted to increase the loads during the first 3 weeks of each cycle provided form was not compromised. Training sessions were personally supervised by the research staff, who adjusted loads as needed. The lifted loads and perceived exertion ratings were charted for each exercise using a paper tracking form.

Table 1 Energy and nutrient intake throughout the study

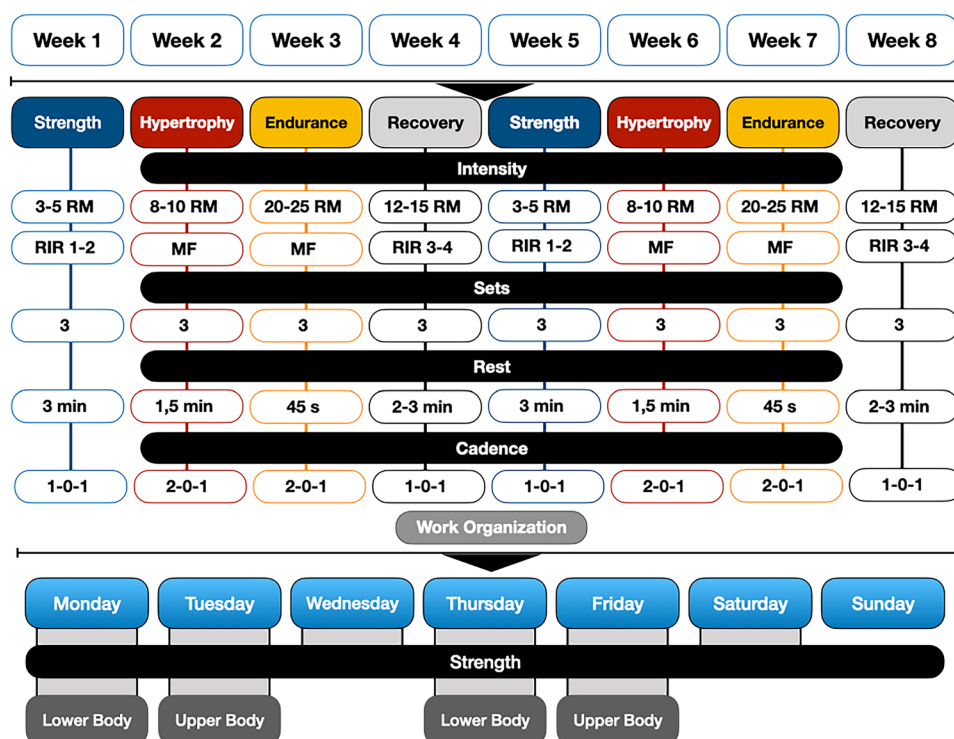
| Parameter | Units | KD | NKD | P value |
|---------------|--|---------------------------|---------------------------|---------|
| Energy | kcal·kg-FFM ⁻¹ ·d ⁻¹ | 40.1 ± 2.7 | 45.5 ± 1.6 | < 0.05 |
| | kcal·d ⁻¹ | 1710.4 ± 160.0 | 1979.6 ± 140.0 | 0.001 |
| Protein | g·d ⁻¹ | 115.1 ± 17.7 | 97.3 ± 7.6 | 0.012 |
| | % kcal Total | 26.8 ± 2.3 | 19.7 ± 1.4 | < 0.05 |
| | g·kg-FFM ⁻¹ ·d ⁻¹ | 2.7 ± 0.3 | 2.2 ± 0.1 | 0.002 |
| Carbohydrates | g·d ⁻¹ | 38.6 ± 4.5 | 282.1 ± 25.1 ^a | < 0.05 |
| | % kcal Total | 9.1 ± 1.3 | 57.0 ± 1.9 | < 0.05 |
| | g·kg-FFM ⁻¹ ·d ⁻¹ | 0.9 ± 0.1 | 6.5 ± 0.4 | < 0.05 |
| Fat | g·d ⁻¹ | 121.7 ± 11.8 ^b | 51.3 ± 4.6 | < 0.05 |
| | % kcal Total | 64.1 ± 2.3 | 23.3 ± 1.6 | < 0.05 |
| | g·kg-FFM ⁻¹ ·d ⁻¹ | 2.9 ± 0.2 | 1.2 ± 0.1 | < 0.05 |

The values are expressed as average ± standard deviation

KD ketogenic diet group, NKD non-ketogenic diet group, FM fat mass, FFM fat-free mass

^aCarbohydrate distribution in NKD group; ≈ 60% starch, ≈ 25% simple, and 15% fiber

^bFat distribution in KD group; ≈ 31% saturated, ≈ 40% monounsaturated, and ≈ 29% polyunsaturated fats

Fig. 2 Organization of the strength training protocol

Outcomes

The considered primary outcomes were SBP and diastolic blood pressure (DBP) and VAT as cardiometabolic factors. Secondary outcomes included bone health-related parameters, such as BMC, bone mineral content per area (BMA) and bone mineral density (BMD). All variables were measured at baseline and after the eight-week intervention.

Primary outcome measures

The SBP and DBP (in mmHg) were measured using an OMRON M3 Intellisense digital automatic monitor (Omron Healthcare, Kyoto, Japan). Testing was carried out with participants seated in a quiet room, with BP measurements obtained on the left arm after a 5–10-min rest period. The mean of two measurements, less than 2 mmHg apart, was used as the final value.

To evaluate VAT, body composition was measured seven days after menstruation in both the pre- and postintervention periods, to avoid the potential increase in body mass due to water retention caused by hormonal fluctuations (Rosenfeld et al. 2008; Stachenfeld 2008). Total and regional body composition measurements were estimated using DXA (Hologic QDR 4500, Bedford, MA). Each subject was scanned by a certified technician, and the machine's computer algorithm (software version APEX 3.0) distinguished bone and soft tissue, edge detection, and regional demarcations. After measuring the total fat mass

within the abdominal region, all area denoted by reference markers as “subcutaneous fat” is then subtracted from this number, to generate VAT. The participants wore sport clothes and removed all materials that could attenuate the X-ray beam including jewelry items and underwear containing wire. Calibration of the densitometer was checked daily against a standard calibration block supplied by the manufacturer. (Phantom 21965 Lumbar Spine with anthropomorphic characteristics of 4 hydroxyapatite vertebrae included in resin. C.V 0.415%). The abdominal region was delineated by an upper horizontal border located half the distance between the acromia and external ends of the iliac crests, a lower border determined by the external end of the iliac crests and the lateral borders extending to the edge of the abdominal soft tissue. All trunk tissue within this standardized region was selected for analysis. To determine intertester reliability, two separate observers manually selected the area for each participant.

Secondary outcome measures

The secondary outcomes were BMC (in g) and BMD ($\text{g}\cdot\text{cm}^{-2}$). They were measured for each individual with whole-body scans taken as mentioned previously. Z scores were calculated using age- and sex-specific normative data provided by Hologic. The same certified technician performed all analyses using the same technique for all measurements.

Table 2 Characteristics of the participants at baseline

| | KD (<i>n</i> = 10) | NKD (<i>n</i> = 11) | Total (<i>n</i> = 21) | <i>P</i> |
|-------------|------------------------|-------------------------|---------------------------|----------|
| Age (y) | 26.8 ± 3.9 | 28.3 ± 4.1 | 27.6 ± 4.0 | 0.41 |
| Height (cm) | 161.6 ± 7.4 | 162.6 ± 6.2 | 162.1 ± 6.6 | 0.73 |
| BM (kg) | 61.9 ± 9.8 | 62.6 ± 5.9 | 62.3 ± 7.8 | 0.51 |
| BMI | 23.8 ± 3.6 | 23.7 ± 2.2 | 23.7 ± 2.9 | 0.96 |
| FM (kg) | 18.4 ± 6.4 | 18.3 ± 4.3 | 18.4 ± 5.3 | 0.98 |
| FFM (kg) | 42.8 ± 5.4 | 43.5 ± 2.8 | 43.2 ± 4.1 | 0.70 |

Statistical analysis

The results for the variables are expressed as the mean and standard deviation. An independent samples *t* test was employed to compare between-group variables at baseline and their change over the study period (Δ = post-test–pre-test). The comparison between means (pre-test and post-test) was performed with the paired *t* test, and the effect size (ES) was determined by the Hedges' *g* test. Repeated measures ANOVA was used to establish the within-subject (*time*: pre-test vs. post-test), between-subject (*group*: NKD vs. KD) and interaction effects between these factors (*time* × *group*). In this general linear model (GLM), the Greenhouse–Geisser correction was used to establish the principal effect of *time* and *time* × *group*, considering the partial eta square value (η^2p) as a measure of ES. The normality of the data was determined with the Shapiro–Wilk test, and the homogeneity of the variance was determined with Levene's test. The significance level assumed for all tests was 0.05. Statistical analysis was performed with SPSS version 25 (SPSS Inc., Chicago, USA), and the graphics were processed with the application Estimation Stats (www.estimationstats.com).

Results

There were positive outcomes for compliance with the KD in every weekly test for all those completing the study (no data available since only dietary compliance was assessed, besides the dietary control during the intervention). Table 2 shows the characteristics of the participants at baseline, with no statistically significant differences observed between groups (*P* > 0.05).

Furthermore, there were no differences between the KD and NKD groups in terms of upper-body, lower-body, and total training load from weeks 1–4 and 5–8 (Table 3).

Blood pressure

The significant reduction in SBP was observed for the KD group with a large associated ES (-6.3 ± 6.0 mmHg,

Table 3 Results for the study variables after the intervention with the KD and NKD programmes

| | NKD | | KD | | | | | | P | ES | Time × Group P (η ² p) | Group P (η ² p) | Time P (η ² p) | |
|---------------------------|-----------------|-----------------|--------------------------------|-------|-------|----------------|-----------------|-----------------------------------|-------|-------|--------------------------------------|-------------------------------|------------------------------|------------------|
| | Pre-test | Post-test | Δ | P | ES | Pre-test | Post-test | Δ | | | | | | P |
| SBP (mmHg) | 114.0 ± 11.8 | 113.6 ± 9.8 | -0.4 ± 8.9 (-6.8-6.0) | 0.890 | -0.04 | 114.2 ± 8.5 | 107.9 ± 6.0 | -6.3 ± 6.0 (-10.5 to -2.0) | 0.009 | -0.81 | 0.102 | -0.71 | 0.066 (0.176) | 0.480 (0.028) |
| DBP (mmHg) | 73.8 ± 6.8 | 72.8 ± 6.0 | -1.1 ± 5.0 (-4.6-2.5) | 0.521 | -0.16 | 72.0 ± 3.1 | 70.0 ± 5.4 | -2.0 ± 5.2 (-5.7-1.7) | 0.254 | -0.43 | 0.681 | -0.18 | 0.196 (0.091) | 0.310 (0.057) |
| BMC (g) | 2015.7 ± 235.6 | 2019.7 ± 203.3 | 4.0 ± 91.1 (-61.2-69.2) | 0.893 | 0.02 | 1978.3 ± 365.8 | 1936.1 ± 325.0 | -42.2 ± 72.5 (-94.1-9.6) | 0.099 | -0.12 | 0.225 | -0.53 | 0.313 (0.057) | 0.643 (0.081) |
| BMD (g·cm ⁻²) | 1.11 ± 0.09 | 1.11 ± 0.08 | 0.00 ± 0.02 (-0.02-0.02) | 0.886 | 0.01 | 1.08 ± 0.10 | 1.10 ± 0.10 | 0.02 ± 0.02 (0.01-0.03) | 0.014 | 0.19 | 0.076 | 0.80 | 0.049 (0.197) | 0.657 (0.011) |
| VAT (g) | 6486.8 ± 1424.7 | 6509.1 ± 1482.8 | 22.3 ± 350.2 (-228.2-272.8) | 0.845 | 0.01 | 7360.9 ± 323.3 | 6917.5 ± 3079.7 | -443.4 ± 939.4 (-1115.4-228.6) | 0.170 | -0.13 | 0.159 | -0.74 | 0.201 (0.089) | 0.563 (0.019) |

$P=0.009$, $ES=-0.81$) but no improvements were found for the NKD group (-0.4 ± 8.9 mmHg, $P=0.890$, $ES=-0.04$). Analysis of the effect of the factors showed no differences in *time* ($P=0.066$, $\eta^2p=0.176$), in *group* ($P=0.480$, $\eta^2p=0.028$) nor in the *time* \times *group* interaction. However, a medium between-group ES difference was noted (-0.71). No significant changes were found in the two groups for DPB, and the GLM showed no difference in *time*, in *group* or in the *time* \times *group* interaction.

Visceral adipose tissue

We found no changes in VAT in either the NKD group (22.3 ± 350.2 g, $P=0.845$, $ES=0.01$) or the KD group (-443.4 ± 939.4 g, $P=0.170$, $ES=-0.13$). Similarly, no difference was found in the GLM for *time* ($P=0.201$, $\eta^2p=0.089$), *Group* ($P=0.563$, $\eta^2p=0.019$) or *Time* \times *Group* ($P=0.159$, $\eta^2p=0.107$), and the ES for the change between the groups was moderate (-0.74).

Bone health

The results showed no significant changes in BMC in both groups (NKD: 4.0 ± 91.1 g, $P=0.893$, $ES=0.02$; KD: -42.2 ± 72.5 g, $P=0.099$, $ES=-0.12$). The factor analysis showed no significant differences (*Time*: $P=0.313$, $\eta^2p=0.057$; *Group*: $P=0.643$, $\eta^2p=0.012$; *Time* \times *Group*: $P=0.225$, $\eta^2p=0.081$). We found a significant increase in BMD in the KD group (0.02 ± 0.02 g·cm $^{-2}$, $P=0.014$; $ES=0.19$), but not in the NKD group (0.00 ± 0.02 g·cm $^{-2}$, $P=0.886$, $ES=0.01$). Similarly, a difference was found for *time* ($P=0.049$; $\eta^2p=0.197$), but not for *group* ($P=0.657$; $\eta^2p=0.011$) or *time* \times *group* ($P=0.076$; $\eta^2p=0.165$), with a large between-group ES noted (0.80). These results are displayed in Table 4 and Fig. 3.

Discussion

We sought to evaluate the efficacy of a KD combined with a RT programme in resistance-trained women on cardio-metabolic (BP and VAT) and bone health parameters (BMC and BMD). An elevated BP is associated with an increased risk of cardiovascular disease (Vasan et al. 2001). Some evidence indicates that a substantial reduction in carbohydrates leads to a reduction of BP in adults with prehypertension or stage 1 hypertension (Appel et al. 2005). Yancy et al. (Yancy et al. 2010) showed that a low-carbohydrate KD decreased systolic BP by 5.9 mmHg, in comparison to an increase in a low-fat diet group ($+1.5$ mmHg) in overweight and obese individuals (25% female). Furthermore, two meta-analysis of randomized clinical trials concluded that KD not only induces a long-term more significant improvement in BP when compared to low-fat diets (Bueno et al. 2013), but also is effective and safe for improving cardiometabolic markers (e.g., SBP and DBP) in overweight and obese individuals (Castellana et al. 2020). Although RT has been associated with improvements in cardiovascular disease risk factors, including the reduction of SBP (Cornelissen et al. 2011), to our knowledge, this is the first time that a reduction effect on BP after KD plus RT is reported in trained women, which is in agreement with previous randomized trials in other populations. Our results showed that consumption of a KD in combination with RT led to a significant reduction in SBP compared to a NKD after an eight-week intervention period. Moreover, the corresponding ES for the KD group was large compared to a trivial value in the NKD group (see Table 4). However, it should be noted that our subjects had baseline BP within normal limits; therefore, further research is warranted to determine the cause of this hypotensive effect (e.g., decreased sympathetic activity, diminished systolic volume, reduced peripheral vascular resistance) and whether this might contribute to the chronic adaptation to exercise (Brito et al. 2018) or negatively affect physical performance.

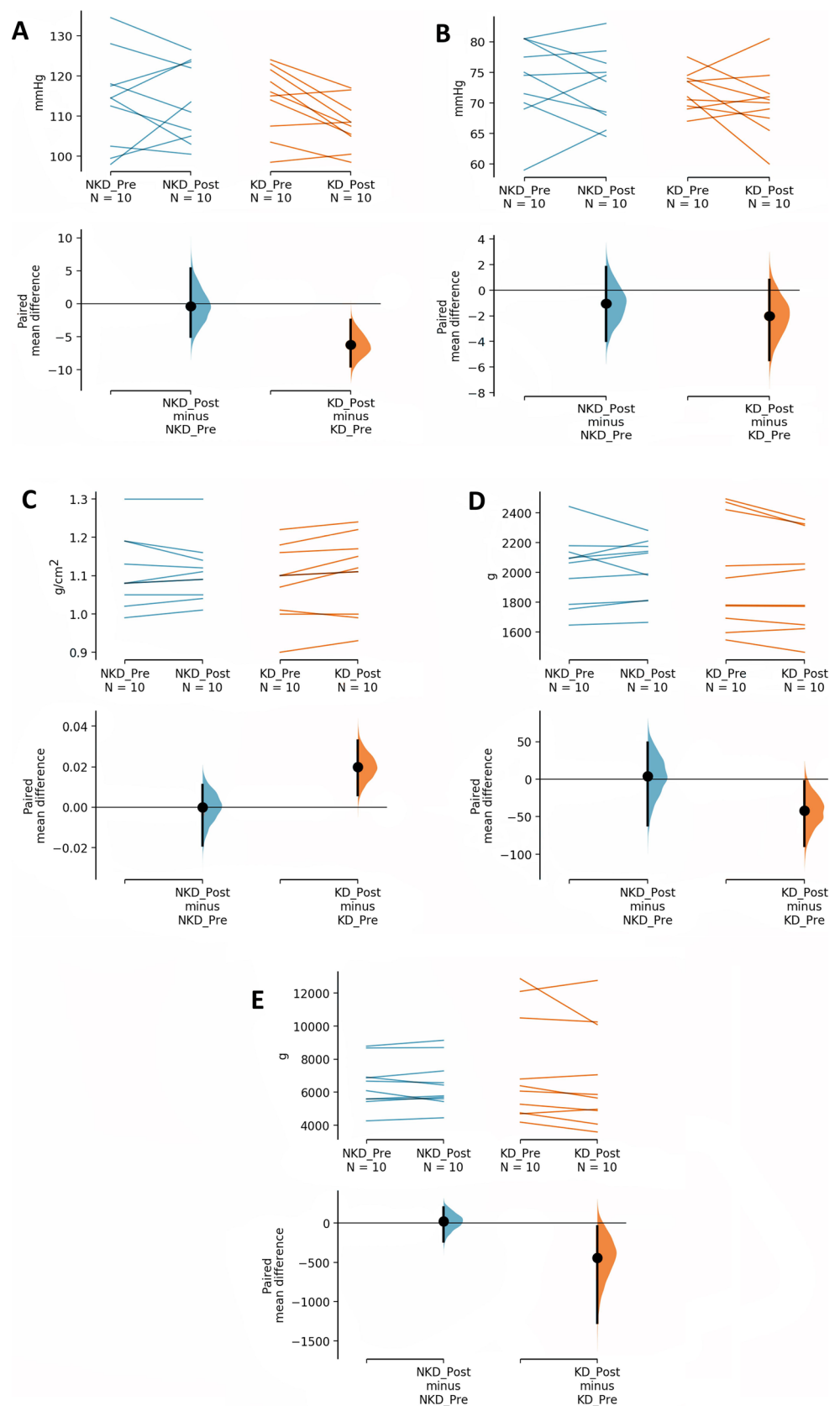
The VAT is another measure associated with cardio-metabolic risk, particularly because of its negative effects on insulin sensitivity (Zhang et al. 2015). Our study found

Table 4 Between-group comparison of the training load

| Training | Week | KD X \pm SD | NKD X \pm SD | Δ (95% CI) | Sig. |
|------------|------|----------------------|----------------------|--------------------------|-------|
| Upper-body | 1–4 | 3920.8 \pm 734.9 | 4406.6 \pm 1015.2 | - 485.8 (- 1352.8–381.1) | 0.253 |
| Lower-body | | 7934.1 \pm 747.5 | 8042.0 \pm 576.1 | - 107.9 (- 749.9–534.2) | 0.727 |
| Total load | | 11854.9 \pm 1149.6 | 12448.6 \pm 1352.6 | - 593.7 (- 1816.2–628.9) | 0.320 |
| Upper-body | 5–8 | 4422.4 \pm 1042.5 | 4821.4 \pm 1042.0 | - 399.0 (- 1409.3–611.4) | 0.416 |
| Lower-body | | 8839.4 \pm 665.7 | 8896.8 \pm 604.8 | - 57.4 (- 672.1–557.4) | 0.846 |
| Total Load | | 13261.9 \pm 1224.5 | 13718.2 \pm 1334.7 | - 456.4 (- 1701.1–788.4) | 0.450 |

Data are mean \pm SD

Fig. 3 The difference in paired means for **a.** Systolic blood pressure, **b.** diastolic blood pressure, **c.** bone mineral density and **d.** bone mineral content and **e.** visceral adipose tissue



no significant reductions in VAT in either the KD group or the NKD group. These data are contrary to previous results reported by our research group in trained men (Vargas et al. 2018). Notably, the nutritional prescription for the KD study in men was based on 39 kcal·kg⁻¹ FFM, while in the present was on \approx 40 kcal·kg⁻¹ FFM. Further research is needed to establish sex differences in this regard, given that a potential influence of female hormones on the response to the KD in energy substrates metabolism has been reported at given exercise intensities (Durkalec-Michalski et al. 2019). Overall, it seems that a KD in combination with RT has the potential to improve cardiometabolic health, given that previous randomized controlled studies have demonstrated that it might reduce body fat without significantly changing FFM in overweight (Jabekk et al. 2010) or resistance-trained women (Vargas-Molina et al. 2020). Researchers and practitioners must be cautious about the reported increase in LDL cholesterol after a 12-week KD in recreational cross-trainees (Kephart et al. 2018), which warrants further research.

Concerning our secondary outcome, negative effects on bone health have been reported in several studies of the longer term-side effects associated with the KD in a pediatric population (Bergqvist et al. 2008; Bergqvist 2012; Simm et al. 2017; Draaisma et al. 2019). Additionally, Heikura et al. assessed serum markers of bone modeling/remodeling and found deleterious effects in these outcomes in elite endurance athletes after a short-term low-carbohydrate KD (Heikura et al. 2019). In spite of the above findings, some clinical research concluded that short-term KD does not lead to bone impairment in healthy obese adults (Colica et al. 2017; Perissiou et al. 2020). Interestingly, a case series suggested that maintaining a KD for more than five years does not have major negative effects on bone health in women diagnosed with SLC2A1 deficiency syndrome (Bertoli et al. 2014). Our study actually found favorable BMD changes in the KD group but not in the NKD group. Although an explanation for discrepancies in findings is not clear, it should be noted that the previous investigations were not performed on strength-trained participants, which may have helped promote a positive effect on BMD in our study (Villalon et al. 2011). In addition, workload can contribute positively to the increase in BMD, for this reason, we report the total loads of the upper and lower limb of the first 4 weeks compared to the last 4. Despite the fact that both groups breastfed, there were no differences between groups.

Given that bone changes take time to develop, it is important to consider that more research is needed to evaluate the long-term effects of a low-carbohydrate KD, besides the proper medical supervision (Rondanelli et al. 2020).

Our study has several limitations that should be considered when drawing practical inferences. First, the use of urine ketone strips may be less sensitive than blood-sample methods. Notably, as opposed to blood collection, this test

does not allow the detection of the 3-hydroxybutyrate, which is the predominant metabolite in a ketosis state. Second, although we regularly assessed appetite and encouraged adherence to the nutritional protocol, several participants in the KD group showed difficulty in consuming the required calories, which in turn may have unduly influenced results. Third, the pre-test was performed in a condition where glycogen stores were full, and the post-test was performed when glycogen stores were depleted, which may have influenced the total FFM due to alterations in intracellular water balance. Finally, a relatively small sample size, a short intervention time (8 weeks), and the absence of variables related to cardiometabolic health (e.g., LDL-c, HDL-c, and triacylglycerol concentrations) limit the ability to draw strong inferences on studied outcomes.

Conclusions

We conclude that a non-energy-restricted KD diet in conjunction with a RT program may help to improve health-related cardiovascular and bone parameters in resistance-trained women. However, more studies are required to determine the potential advantages of a KD diet compared to a NKD diet. Meanwhile prudent considerations of the long-term recommendations of KD should be taken into account.

Acknowledgements Supported by University of Málaga (Campus of International Excellence Andalucía Tech).

Author contributions SV served as study coordinator. SV and JBP conceived and designed the experiments. RR and JBP served as lab coordinator and project manager for the study coordination, respectively. SV and RR assisted in data collection. SV designed the nutritional protocols. SV oversight nutrition and training. JLP analyzed the data. SV, JLP, BJS, RBK, JBP, LC, and DAB assisted in analysis, and manuscript review. SV, JLP, DAB, and LC wrote the first draft. SV, RBK, BJS, LC, DAB, JLP, and JBP assisted in the statistics advice, discussion analysis, and manuscript preparation. All authors read and approved the final manuscript.

Declarations

Conflict of interest No conflict of interest is declared by the authors.

References

- Allen BG, Bhatia SK, Anderson CM, Eichenberger-Gilmore JM, Sibellaller ZA, Mapuskar KA, Schoenfeld JD, Buatti JM, Spitz DR, Fath MA (2014) Ketogenic diets as an adjuvant cancer therapy: history and potential mechanism. *Redox Biol* 2:963–970. <https://doi.org/10.1016/j.redox.2014.08.002>
- Appel LJ, Sacks FM, Carey VJ, Obarzanek E, Swain JF, Miller ER 3rd, Conlin PR, Erlinger TP, Rosner BA, Laranjo NM, Charleston J, McCarron P, Bishop LM (2005) Effects of protein, mono-unsaturated fat, and carbohydrate intake on blood pressure and

- serum lipids: results of the OmniHeart randomized trial. *JAMA* 294(19):2455–2464. <https://doi.org/10.1001/jama.294.19.2455>
- Bartlett JD, Hawley JA, Morton JP (2015) Carbohydrate availability and exercise training adaptation: too much of a good thing? *Eur J Sport Sci* 15(1):3–12. <https://doi.org/10.1080/17461391.2014.920926>
- Bergqvist AG (2012) Long-term monitoring of the ketogenic diet: Do's and Don'ts. *Epilepsy Res* 100(3):261–266. <https://doi.org/10.1016/j.epilepsyres.2011.05.020>
- Bergqvist AG, Schall JI, Stallings VA, Zemel BS (2008) Progressive bone mineral content loss in children with intractable epilepsy treated with the ketogenic diet. *Am J Clin Nutr* 88(6):1678–1684. <https://doi.org/10.3945/ajcn.2008.26099>
- Bertoli S, Trentani C, Ferraris C, De Giorgis V, Veggiotti P, Tagliabue A (2014) Long-term effects of a ketogenic diet on body composition and bone mineralization in GLUT-1 deficiency syndrome: a case series. *Nutrition* 30(6):726–728. <https://doi.org/10.1016/j.nut.2014.01.005>
- Brito LC, Fecchio RY, Peçanha T, Andrade-Lima A, Halliwill JR, Forjaz CLM (2018) Postexercise hypotension as a clinical tool: a “single brick” in the wall. *J Am Soc Hypertens* 12(12):e59–e64. <https://doi.org/10.1016/j.jash.2018.10.006>
- Bueno NB, de Melo ISV, de Oliveira SL, da Ataíde Rocha T (2013) Very-low-carbohydrate ketogenic diet v low-fat diet for long-term weight loss: a meta-analysis of randomised controlled trials. *Br J Nutr* 110(7):1178–1187. <https://doi.org/10.1017/S0007114513000548>
- Burke LM (2015) Re-examining high-fat diets for sports performance: did we call the ‘nail in the coffin’ too soon? *Sports Med* 45(1):33–49. <https://doi.org/10.1007/s40279-015-0393-9>
- Cahill GF (1970) Starvation in man. *N Engl J Med* 282(12):668–675. <https://doi.org/10.1056/NEJM197003192821209>
- Cahill GF (2006) Fuel metabolism in starvation. *Annu Rev Nutr* 26:1–22. <https://doi.org/10.1146/annurev.nutr.26.061505.111258>
- Castellana M, Conte E, Cignarelli A, Perrini S, Giustina A, Giovanella L, Giorgino F, Trimboli P (2020) Efficacy and safety of very low calorie ketogenic diet (VLCKD) in patients with overweight and obesity: a systematic review and meta-analysis. *Rev Endocr Metab Disord* 21(1):5–16. <https://doi.org/10.1007/s1154-019-09514-y>
- Chang CK, Borer K, Lin PJ (2017) Low-carbohydrate-high-fat diet: can it help exercise performance? *J Hum Kinet* 56:81–92. <https://doi.org/10.1515/hukin-2017-0025>
- Cicero AF, Benelli M, Brancaloni M, Dainelli G, Merlini D, Negri R (2015) Middle and long-term impact of a very low-carbohydrate ketogenic diet on cardiometabolic factors: a multi-center, cross-sectional, clinical study. *High Blood Press Cardiovasc Prev* 22(4):389–394. <https://doi.org/10.1007/s40292-015-0096-1>
- Colica C, Merra G, Gasbarrini A, De Lorenzo A, Cioccoloni G, Gualtieri P, Perrone MA, Bernardini S, Bernardo V, Di Renzo L, Marchetti M (2017) Efficacy and safety of very-low-calorie ketogenic diet: a double blind randomized crossover study. *Eur Rev Med Pharmacol Sci* 21(9):2274–2289
- Cornelissen VA, Fagard RH, Coeckelberghs E, Vanhees L (2011) Impact of resistance training on blood pressure and other cardiovascular risk factors: a meta-analysis of randomized, controlled trials. *Hypertension* 58(5):950–958. <https://doi.org/10.1161/HYPERTENSIONAHA.111.177071>
- Cotter DG, Schugar RC, Crawford PA (2013) Ketone body metabolism and cardiovascular disease. *Am J Physiol Heart Circ Physiol* 304(8):H1060–H1076. <https://doi.org/10.1152/ajpheart.00646.2012>
- Draaisma JMT, Hampsink BM, Janssen M, van Houdt NBM, Linders E, Willemsen MA (2019) The ketogenic diet and its effect on bone mineral density: a retrospective observational cohort study. *Neuropediatrics* 50(6):353–358. <https://doi.org/10.1055/s-0039-1693059>
- Durkalec-Michalski K, Nowaczyk PM, Siedzik K (2019) Effect of a four-week ketogenic diet on exercise metabolism in CrossFit-trained athletes. *J Int Soc Sports Nutr* 16(1):16. <https://doi.org/10.1186/s12970-019-0284-9>
- Egan B, D'Agostino DP (2016) Fueling performance: ketones enter the mix. *Cell Metab* 24(3):373–375. <https://doi.org/10.1016/j.cmet.2016.08.021>
- Francois ME, Gillen JB, Little JP (2017) Carbohydrate-restriction with high-intensity interval training: an optimal combination for treating metabolic diseases? *Front Nutr* 4:49. <https://doi.org/10.3389/fnut.2017.00049>
- Gregory RM (2017) A low-carbohydrate ketogenic diet combined with 6-weeks of crossfit training improves body composition and performance. *Int J Sports Exerc Med*. <https://doi.org/10.23937/2469-5718/1510054>
- Hartman AL, Vining EPG (2007) Clinical aspects of the ketogenic diet. *Epilepsia* 48(1):31–42. <https://doi.org/10.1111/j.1528-1167.2007.00914.x>
- Heikura IA, Burke LM, Hawley JA, Ross ML, Garvican-Lewis L, Sharma AP, McKay AKA, Leckey JJ, Welvaert M, McCall L, Ackerman KE (2019) A short-term ketogenic diet impairs markers of bone health in response to exercise. *Front Endocrinol* 10:880. <https://doi.org/10.3389/fendo.2019.00880>
- Hu T, Yao L, Reynolds K, Whelton PK, Niu T, Li S, He J, Bazzano LA (2015) The effects of a low-carbohydrate diet vs a low-fat diet on novel cardiovascular risk factors: a randomized controlled trial. *Nutrients* 7(9):7978–7994. <https://doi.org/10.3390/nu7095377>
- Jabekk PT, Moe IA, Meen HD, Tomten SE, Hostmark AT (2010) Resistance training in overweight women on a ketogenic diet conserved lean body mass while reducing body fat. *Nutr Metab (Lond)* 7:17. <https://doi.org/10.1186/1743-7075-7-17>
- Kephart WC, Pledge CD, Roberson PA, Mumford PW, Romero MA, Mobley CB, Martin JS, Young KC, Lowery RP, Wilson JM, Huggins KW, Roberts MD (2018) The three-month effects of a ketogenic diet on body composition, blood parameters, and performance metrics in crossfit trainees: a pilot study. *Sports (basel)*. <https://doi.org/10.3390/sports6010001>
- Koeslag JH, Noakes TD, Sloan AW (1980) Post-exercise ketosis. *J Physiol* 301(1):79–90. <https://doi.org/10.1113/jphysiol.1980.sp013190>
- Kosinski C, Jornayvaz FR (2017) Effects of Ketogenic Diets on Cardiovascular Risk Factors: Evidence from Animal and Human Studies. *Nutrients*. <https://doi.org/10.3390/nu9050517>
- Loucks AB (2003) Energy availability, not body fatness, regulates reproductive function in women. *Exerc Sport Sci Rev* 31(3):144–148
- McArtney R, Bailey A, Champion H (2017) What is a ketogenic diet and how does it affect the use of medicines? *Arch Dis Child Educ Pract Ed* 102(4):194–199. <https://doi.org/10.1136/archdischi-2014-307000>
- McKay AKA, Peeling P, Pyne DB, Welvaert M, Tee N, Leckey JJ, Sharma AP, Ross MLR, Garvican-Lewis LA, Swinkels DW, Laarakkers CM, Burke LM (2019) Chronic adherence to a ketogenic diet modifies iron metabolism in elite athletes. *Med Sci Sports Exerc* 51(3):548–555. <https://doi.org/10.1249/MSS.0000000000001816>
- Perissiou M, Borkoles E, Kobayashi K, Polman R (2020) The effect of an 8 week prescribed exercise and low carbohydrate diet on cardiorespiratory fitness, body composition and cardiometabolic risk factors in obese individuals: a randomised controlled trial. *Nutrients*. <https://doi.org/10.3390/nu12020482>
- Phinney SD (2004) Ketogenic diets and physical performance. *Nutr Metab (Lond)* 1(1):2. <https://doi.org/10.1186/1743-7075-1-2>
- Puchalska P, Crawford PA (2017) Multi-dimensional roles of ketone bodies in fuel metabolism, signaling, and therapeutics. *Cell Metab* 25(2):262–284. <https://doi.org/10.1016/j.cmet.2016.12.022>

- Reed JL, De Souza MJ, Mallinson RJ, Scheid JL, Williams NI (2015) Energy availability discriminates clinical menstrual status in exercising women. *J Int Soc Sports Nutr* 12:11. <https://doi.org/10.1186/s12970-015-0072-0>
- Rondanelli M, Faliva MA, Gasparri C, Peroni G, Spadaccini D, Maugeri R, Nichetti M, Infantino V, Perna S (2020) Current opinion on dietary advice in order to preserve fat-free mass during a low-calorie diet. *Nutrition*. <https://doi.org/10.1016/j.nut.2019.110667>
- Rosenfeld R, Livne D, Nevo O, Dayan L, Milloul V, Lavi S, Jacob G (2008) Hormonal and volume dysregulation in women with premenstrual syndrome. *Hypertension* 51(4):1225–1230. <https://doi.org/10.1161/HYPERTENSIONAHA.107.107136>
- Simm PJ, Bicknell-Royle J, Lawrie J, Nation J, Draffin K, Stewart KG, Cameron FJ, Scheffer IE, Mackay MT (2017) The effect of the ketogenic diet on the developing skeleton. *Epilepsy Res* 136:62–66. <https://doi.org/10.1016/j.epilepsyres.2017.07.014>
- Sjödin A, Hellström F, Sehlstedt E, Svensson M, Burén J (2020) Effects of a ketogenic diet on muscle fatigue in healthy, young, normal-weight women: a randomized controlled feeding trial. *Nutrients*. <https://doi.org/10.3390/nu12040955>
- Stachenfeld NS (2008) Sex hormone effects on body fluid regulation. *Exerc Sport Sci Rev* 36(3):152–159. <https://doi.org/10.1097/JES.0b013e31817be928>
- Stubbs BJ, Cox PJ, Evans RD, Santer P, Miller JJ, Faulk OK, Magor-Elliott S, Hiyama S, Stirling M, Clarke K (2017) On the metabolism of exogenous ketones in humans. *Front Physiol* 8:848. <https://doi.org/10.3389/fphys.2017.00848>
- Teixeira V, Voci SM, Mendes-Netto RS, da Silva DG (2018) The relative validity of a food record using the smartphone application MyFitnessPal. *Nutr Diet* 75(2):219–225. <https://doi.org/10.1111/1747-0080.12401>
- Urbain P, Bertz H (2016) Monitoring for compliance with a ketogenic diet: what is the best time of day to test for urinary ketosis? *Nutr Metab* 13:77. <https://doi.org/10.1186/s12986-016-0136-4>
- Valenzano A, Polito R, Trimigno V, Di Palma A, Moscatelli F, Corso G, Sessa F, Salerno M, Montana A, Di Nunno N, Astuto M, Daniele A, Carotenuto M, Messina G, Cibelli G, Monda V (2019) Effects of very low calorie ketogenic diet on the orexinergic system, visceral adipose tissue, and ros production. *Antioxidants* (basel). <https://doi.org/10.3390/antiox8120643>
- Vargas S, Romance R, Petro JL, Bonilla DA, Galancho I, Espinar S, Kreider RB, Benitez-Porres J (2018) Efficacy of ketogenic diet on body composition during resistance training in trained men: a randomized controlled trial. *J Int Soc Sports Nutr* 15(1):31. <https://doi.org/10.1186/s12970-018-0236-9>
- Vargas-Molina S, Petro JL, Romance R, Kreider RB, Schoenfeld BJ, Bonilla DA, Benitez-Porres J (2020) Effects of a ketogenic diet on body composition and strength in trained women. *J Int Soc Sports Nutr* 17(1):19. <https://doi.org/10.1186/s12970-020-00348-7>
- Vasan RS, Larson MG, Leip EP, Evans JC, O'Donnell CJ, Kannel WB, Levy D (2001) Impact of high-normal blood pressure on the risk of cardiovascular disease. *N Engl J Med* 345(18):1291–1297. <https://doi.org/10.1056/NEJMoa003417>
- Veech RL (2004) The therapeutic implications of ketone bodies: the effects of ketone bodies in pathological conditions: ketosis, ketogenic diet, redox states, insulin resistance, and mitochondrial metabolism. *Prostaglandins Leukot Essent Fatty Acids* 70(3):309–319. <https://doi.org/10.1016/j.plefa.2003.09.007>
- Villalon KL, Gozansky WS, Van Pelt RE, Wolfe P, Jankowski CM, Schwartz RS, Kohrt WM (2011) A losing battle: weight regain does not restore weight loss-induced bone loss in postmenopausal women. *Obesity* 19(12):2345–2350. <https://doi.org/10.1038/oby.2011.263>
- Włodarek D (2019) Role of ketogenic diets in neurodegenerative diseases (Alzheimer's disease and Parkinson's disease). *Nutrients* 11(1):169
- Wooding DJ, Packer JE, Kato H, West DWD, Courtney-Martin G, Pencharz PB, Moore DR (2017) Increased protein requirements in female athletes after variable-intensity exercise. *Med Sci Sports Exerc* 49(11):2297–2304. <https://doi.org/10.1249/MSS.0000000000001366>
- Wroble KA, Trott MN, Schweitzer GG, Rahman RS, Kelly PV, Weiss EP (2019) Low-carbohydrate, ketogenic diet impairs anaerobic exercise performance in exercise-trained women and men: a randomized-sequence crossover trial. *J Sports Med Phys Fitness* 59(4):600–607. <https://doi.org/10.23736/S0022-4707.18.08318-4>
- Yancy WS Jr, Westman EC, McDuffie JR, Grambow SC, Jeffreys AS, Bolton J, Chalecki A, Oddone EZ (2010) A randomized trial of a low-carbohydrate diet vs orlistat plus a low-fat diet for weight loss. *Arch Intern Med* 170(2):136–145. <https://doi.org/10.1001/archinternmed.2009.492>
- Zajac A, Poprzecki S, Maszczyk A, Czuba M, Michalczyk M, Zydek G (2014) The effects of a ketogenic diet on exercise metabolism and physical performance in off-road cyclists. *Nutrients* 6(7):2493–2508. <https://doi.org/10.3390/nu6072493>
- Zhang M, Hu T, Zhang S, Zhou L (2015) Associations of Different Adipose Tissue Depots with Insulin Resistance: a systematic review and meta-analysis of observational studies. *Sci Rep* 5:18495. <https://doi.org/10.1038/srep18495>
- Zinn C, Wood M, Williden M, Chatterton S, Maunders E (2017) Ketogenic diet benefits body composition and well-being but not performance in a pilot case study of New Zealand endurance athletes. *J Int Soc Sports Nutr* 14:22. <https://doi.org/10.1186/s12970-017-0180-0>

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.