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# A single dose of a beetroot-based nutritional gel improves endothelial function in the elderly with cardiovascular risk factors

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## ABSTRACT

Beetroot consumption has been associated with increased NO bioconversion, which may promote beneficial effects on vascular health. The present study evaluated the effects of a beetroot-based nutritional gel (BG) on endothelial function, arterial stiffness and blood pressure in the elderly with cardiovascular risk factors. Twenty elderly individuals were submitted to BG and nitrate-depleted gel (PLA) interventions. Brachial flow-mediated dilation (FMD), reactive hyperaemia (RH), blood flow velocity (BFV), pulse wave velocity (PWV<sub>0</sub>), augmentation index (AI), stiffness parameter ( $\beta$ ), pressure-strain elasticity modulus (Ep) and arterial compliance (AC) were measured 120 min after interventions. Urinary nitrate, nitrite, systolic blood pressure (SBP), diastolic blood pressure (DBP) and heart rate (HR) were measured at baseline, 120 min and 180 min after interventions. BG intervention promoted increase ( $P < 0.05$ ) in FMD (77%), BFV (31%), RH (18%) and urinary nitrite (214%) and nitrate (283%) compared with PLA. A single dose of a new food matrix rich in dietary nitrate improves NO bioconversion and endothelial function in the elderly.

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## 1. Introduction

Cardiovascular diseases (CVD) remain the leading cause of morbidity and mortality in modern societies and the presence of risk factors for CVD is linked to the dysfunction of arteries (Mozaffarian et al., 2016). Aging is the primary risk factor for CVD, and therefore, most forms of CVD are diseases of aging (Mozaffarian et al., 2016; Najjar, Scuteri, & Lakatta, 2005). Aging causes numerous changes to arteries that increase the risk for CVD, but two key contributors are the stiffening of the large elastic arteries (aorta and carotid arteries) and the development

of vascular endothelial dysfunction (Seals, Jablonski, & Donato, 2011). Reduced bioavailability of nitric oxide (NO) has been associated as the possible mechanism mediating vascular endothelial dysfunction in individuals with the presence of risk factors for CVD, including dyslipidaemias, hypertension and aging (Seals et al., 2011; Taddei et al., 2001).

Recently, the consumption of beetroot, a food rich in nitrate, has gained popularity in scientific literature due to the possible effect of the nitrate present in this food on increasing the bioavailability of NO (Baião, Conte-Junior, Paschoalin, & Alvares, 2016). The dietary nitrate can be reduced to nitrite in the oral cavity by the enzymatic action of nitrate reductase, which is

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expressed by oral commensal bacteria. Once in contact with gastric acid from the stomach, nitrite may be decomposed to NO and other bioactive nitrogen oxides (Lundberg et al., 2009). The notion that dietary nitrate may stimulate bioconversion into NO has been attracting the interest of the scientific community for the nutritional and therapeutic approach to this dietary compound.

Foods in gel form tend to have higher concentrations of nutrients in a reduced volume, which provide cells with greater access to dietary nutrients (Morgado et al., 2016). Furthermore, foods in gel form have the technological advantages of being easy to store at room temperature, easy to carry, and easy to ingest and tolerate (Campbell, Prince, Braun, Applegate, & Casazza, 2008).

Recently we developed a beetroot-based nutritional gel rich in nitrate as a new nutritional strategy to increase dietary nitrate ingestion and we demonstrated increased bio-accessibility of antioxidants present in this food matrix as compared to beetroot juice (Morgado et al., 2016). Therefore, the beetroot-based nutritional gel represents an alternative and convenient source for consuming dietary nitrate and other nutrients as well as having positive acceptance and purchase intention (Morgado et al., 2016). However, no data in humans investigating the acute vascular effects of a beetroot-based gel, which is rich in dietary nitrate, have been published. Therefore, knowledge on the functional properties of beetroot-based nutritional gel is important to provide valuable information about the potential benefit of the nitrate present in this alternative food matrix on vascular health in humans. For this reason, the purpose of the present work was to evaluate whether the beetroot-based nutritional gel rich in nitrate promotes changes in endothelial function, arterial stiffness and arterial blood pressure.

We hypothesised that a single dose of the beetroot gel rich in dietary nitrate would result in increased NO bioconversion and would, consequently, promote changes in the endothelial function (increases in the flow-mediated dilation, FMD; reactive hyperaemia, RH; and blood flow velocity, FV), arterial stiffness (reductions in pulse wave velocity, PWV<sub>β</sub>; augmentation index, AI; stiffness parameter,  $\beta$  and increases in the pressure-strain elasticity modulus, Ep and arterial compliance, AC) and blood pressure (decreases in systolic and diastolic blood pressures) in the elderly with cardiovascular risk factors.

## 2. Methods

### 2.1. Participants

Twenty elderly participants (13 women and 7 men) who were taking between 1 and 3 anti-hypertensive medications for high blood pressure were recruited through announcements in flyers, newspapers, websites, and advertisements during community events. Interested participants were screened for inclusion and exclusion criteria. Participants who qualified after this initial screening were then invited to provide a fasting blood draw and anthropometric measurements to confirm the presence of cardiovascular risk factors. All participants were fully informed of the nature and purpose of the investigation and provided written consent to participate. Inclusion criteria were:

elevated triglycerides  $\geq 150$  mg/dL, reduced HDL-cholesterol  $< 50$  mg/dL for women and  $< 40$  mg/dL for men and elevated waist circumference by population definition (male:  $> 102$  cm and female:  $> 88$  cm). Exclusion criteria included elevated fasting glucose ( $\geq 100$  mg/dL), smoking, beetroot allergy, unwillingness to avoid beetroot products during the entire study, other chronic diseases (diabetes, liver disease, etc.), or acutely ill. All experimental procedures were performed in accordance with the ethical standards of the Declaration of Helsinki and approved by the institutional ethics committee.

### 2.2. Experimental design

The study was conducted in a randomised, double-blind, cross-over and placebo-controlled way. All subjects reported to the laboratory on three occasions, with at least 1-week interval between visits. The first visit was used to explain the experimental procedures and collect clinical and anthropometric data. In the second and third visits, arterial blood pressure measurement and urine samples were drawn at baseline (T0) after a 10-min period of quiet rest. Thereafter, subjects were randomly divided into either a beetroot-based nutritional gel (BG) or a control nitrate-depleted gel (PLA). Endothelial function and arterial stiffness measurements began 120 min after the nutritional intervention (T120) and lasted for approximately 30 min. Arterial blood pressure measurement and urine samples were drawn again at T120 and 180 min after nutritional intervention (T180) (Fig 1). The three visits were held between 07:00 and 11:00 a.m. The participants were instructed to fast for at least 8 hours before each visit. In addition, on the day of the study, the participants were instructed to restrict carbonated mineral water and did not use any mouthwash.

### 2.3. Nutritional intervention

The beetroot-based nutritional gel was prepared according to Morgado et al. (2016). In brief, beetroot juice (BJ) was centrifuged to remove solid particles, filtered and passed into a spray-dryer (Büchi 190, BüchiLaboratoriums – Technik AG, Flawil, Switzerland) to make a beetroot powder. The beetroot powder was mixed with BJ, carboxymethylcellulose (as a thickener), and artificial orange flavour, and the mixture was homogenised

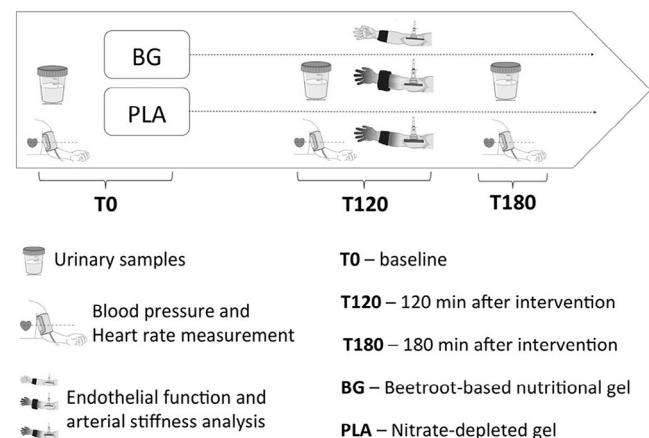


Fig. 1 – Experimental design of the study.

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