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Impact of Beetroot Juice Supplementation Behaviour on Strength and Physiological

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Abstract – Beta vulgaris L. (beetroot) contains high amounts of biologically active substances including betalains and inorganic nitrate. We determined the amounts of various compounds (minerals, betalains, oxalic acid, phenolic acids, and sugars) in juice prepared from seven different beetroot varieties cultivated in Upper Austria. Large differences were found between the varieties for some substances (such as nitrate), whereas others showed only minor variation (certain minerals and sugars). The total betalain content was found to range between 0.8 and 1.3 g/L fresh juice (about 60% betacyanins and 40% betaxanthins) that accounted for 70–100% of the total phenolic content. Other detected phenolic were hydroxycinnamic acids, which accounted for up to 2.6% of total phenolic. Nitrate content varied 10- fold between single varieties. Sugar composition was similar in all varieties with an average total content of about 7.7%, consisting of 95% sucrose. Only minor differences in the concentration of oxalic acid (0.3–0.5 g/L fresh juice) were found between the varieties. In addition, 16 commercial juices and four powders were analyzed for their nitrate contents, as its metabolic product nitric oxide has been reported to provide cardiovascular benefits. Large variations of the nitrate levels, ranging from 0.01 to 2.4 g/L, were found. Dietary supplementation with beetroot juice (BR) containing ~5-8 mmol of inorganic nitrate (NO_3^-) increases plasma nitrite concentration ([NO_2^-]), reduces blood pressure, and may positively influence the physiological responses to exercise. However, the dose-response relationship between the volume of BR ingested and the physiological effects invoked has not been investigated. Following acute BR ingestion, plasma [NO_2^-] increased in a dose-dependent manner, with the peak changes occurring at ~2-3 h. Compared to PL, 70 ml BR did not alter the physiological responses to exercise.

Keywords: Beetroot Juice, Supplementation, Behaviour, Strength, Physiological, Physiological, Responses, Exercise, etc.

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INTRODUCTION

Beetroot (or red beet) is a cultivated form of Beta vulgaris subsp. vulgaris (conditiva) and describes a number of varieties of edible taproots that are grown throughout the Americas, Europe, and Asia. In contrast to their fellow subspecies Beta vulgaris subsp. vulgaris (altissima), known as sugar beet, the sugar content in the conditiva beetroot subspecies is about 2 times lower (U.S. Department of Agriculture, 2013). Therefore, beetroot is grown for food uses (pickles, salad, and juice) rather than for sugar production. In contrast to other fruits, the main sugar in beetroot is sucrose with only small amounts of glucose and fructose (Bavec et al., 2010). Because fructose reduces human exercise capacity, a low fructose and high sucrose content is preferable, for example, in sports drinks. The intense red color of beetroots derives from high concentrations of betalains, a group of phenolic secondary plant metabolites. Betalains are used as natural colorants by the food industry, but have also received increasing attention due to possible health benefits in humans, especially their antioxidant and anti-inflammatory activities. Other benefits include the inhibition of lipid peroxidation, increased resistance to the oxidation of low-density lipoproteins and chemo-preventive effects (Zhang et al., 2013). The betalains that are mainly found in beetroot are betacyanins and betaxanthins (Gandia-Herrero et al., 2010). Apart from betalains, small amounts of hydroxycinnamic acids such as gallic, syringic, and caffeic acids and flavonoids have been identified (Kazimierczak et al., 2014). Athletes, especially those in endurance sports, are the main targets of various beetroot products currently on the market. These commercial products, both juices and powders, are advertised as performance-enhancing legal nutrition supplements. The active ingredient is the inorganic nitrate (NO3_), which is reduced by bacteria in the salvia into nitric oxide (NO). Clinical studies suggest positive effects of increased NO levels on muscle efficiency and fatigue resistance and improvements in time-trial endurance tests of hobby athletes. Furthermore, nitrate ingestion reduced resting blood pressure suggesting it being a nutritional agent for the prevention and treatment of hypertension and cardiovascular diseases (Lundberg et al., 2011). However, there are reports that high levels of nitric oxide are correlated with depressive states, which has to be taken into consideration when consuming excessive amounts of beetroot products. Furthermore, recent evidence suggests that ingested nitrate and nitrite results in an increased endogenous nitrosation, which may lead to formation of carcinogenic metabolites (Habermeyer et al., 2015). In addition to the health beneficial compounds, however, beetroots also contain significant quantities of oxalic acid. Oxalic acid is a strong metal ion chelator interfering with iron and calcium metabolism and can lead to the formation of nephroliths. In the work described here the biochemical composition of juice prepared from seven different popular beetroot varieties grown in Upper Austria was analyzed, focusing on minerals, betalains, sugars, oxalic acid, and phenolic acids. In addition, the respective antioxidant potential of each juice was determined. Furthermore, the content of selected anions found in fresh juice was compared to the one detected in more than 20 commercial beetroot juices, concentrates, and powders. The results of this study highlight the large variation in certain biologically active substances found in popular beetroot varieties. This information will be of special interest as it is hoped to facilitate the development of functional beetroot products with a strong focus on health beneficial effects.

REVIEW OF LITERATURE:

Several studies over the last four years have evaluated the ergogenic effects of acute nitrate supplementation in cyclists, skiers, runners, rowers, kayakers, and team-sport athletes. Surprisingly, 5 out of 10 studies were performed in cyclists. Lansley et al. (2011) was the first to examine the effects acute nitrate supplementation on exercise performance in athletes highlighting its potential use as an ergogenic aid (33). Nine competitive cyclists (VO2max: 56 ml/kg/min) significantly improved performance in a 4 km and a 16.1-km time-trials following ingestion of beetroot juice (6.2 mmol of nitrate) compared with placebo. Beetroot consumption increased also the mean power output and the power output to VO2 ratio, supporting previous findings of increased contractile and mitochondrial efficiency following nitrate supplementation. In contrast to results obtained in untrained individuals, no reduction in VO2 was observed across both distances. Bescos et al. (2012) had competitive cyclists (VO2max: 65 ml/kg/min) cycle for 4 bouts of 6 min at submaximal intensities (35%-65% of peak power) and then perform a maximal incremental test. Sodium nitrate ingestion, 3 h prior to trials, had no significant effect on submaximal O2 cost, on time to exhaustion, on maximal power, and on blood lactate; VO2max, however, was significantly reduced by 3.7%. It should be noted that despite the substantial increase in plasma nitrate (86%) in this study, nitrite, the regulator of NO-induced hypoxic signaling, was increased only by 16%. Cermak et al. (2012) also failed to document an ergogenic effect of a single bolus of nitrate administration (8.7 mmol of nitrate) in well-trained cyclists (VO2max: 60 ml/kg/min). Performance parameters (time, average power and cycling cadence), as well as body fuel selection and metabolites (glucose, free fatty acids, and lactate) were not different after ingestion of nitrate-rich and nitrate-depleted supplements.

As noted, nitrates are natural inorganic components of plant foods. Hord and others (2009) note that approximately 80 percent of human dietary nitrate intake is derived from vegetable consumption, but also note that the total dietary nitrate intake is determined by the type of vegetables consumed, the levels of nitrate in the vegetables, and the amount of vegetables consumed. Table 1 provides a classification of vegetables based on nitrate content, given in milligrams per 100 grams (3.5 ounces) food weight. Other sources of nitrate in the human diet include sodium nitrate as a preservative in processed meats and varying amounts in drinking water.

Nitrate Content*	Vegetables
Very low (< 20 mg/100 g)	Artichoke; asparagus; garlic; onion; mushroom; pea; pepper; potato; sweet potato; tomato
Low (20-50 mg/100 g)	Broccoli; carrot; cauliflower; cucumber; pumpkin; chicory
Middle (50-100 mg/100g)	Cabbage; dill; turnip; Savoy cabbage
High (100-250 mg/100 g)	Celeriac (celery root); Chinese cabbage; endive; fennel; kohlrabi; leek; parsley
Very High (> 250 mg/100 g)	Celery; cress; chervil; lettuce; red beetroot; spinach; rucola (arugula)

*Nitrate content in milligrams per 100 grams of fresh weight

Table 1- Classification of vegetables according to nitrate content

Beetroot Juice and Exercise Performance: In the world of athletic competition, margins of victory are becoming smaller, and in some cases may literally come down to a fraction of a second or the ability to contract a single motor unit one more time. Thus, athletes are constantly in pursuit of any advantage to improve athletic performance. Some athletes may turn toward nutritional supplements, from both natural and organic sources, to provide this edge. Not surprisingly, during the period from 1999 to 2009, the US market for organic and natural foods experienced an increase in annual growth rate from 22.5% to 31.1%, whereas the supplement market had a decline in annual growth rate from 34.5% to 24.8%. In addition, the forecast for "Estimated Compound Annual Sales Growth" from 2010 to 2017 is projected to be 5% for supplements compared to 8% for natural and organic foods. Given this trend for organic and natural food products, it is particularly relevant to understand whether there is an added performance benefit due to the ingredients within these food products acting additively, synergistically, or even negatively compared to a concentrated dose of the isolated bioactive ingredient from the whole food or product.

Beetroot, NO_3^-, and NO-: Beetroot has a high NO_3^- content (>250 mg/100 g of fresh weight), among the highest assessed, and other foods high in NO_3^- include spinach, celery, lettuce, and carrot juice. NO_3^- can be reduced to nitrite via bacteria in the oral cavity and by specific enzymes (eg, xanthine oxidase) within tissues. There are several pathways to metabolize nitrite to NO and other biologically active nitrogen oxides. NO is a signaling molecule formed in the endothelium by the enzyme endothelium NO synthase, which triggers the vasculature to relax (vasodilatation) by interacting with vascular smooth muscle leading to increased blood flow. NO facilitates increased blood flow at rest and during exercise. Given these properties, NO has gained a lot of attention for possible exercise improvements including increased O_2, glucose, and other nutrient uptake to better fuel working muscles. Bradley et al(1999) and Balon and Nadler(1997) reported NO production contributed significantly to exercise-induced skeletal muscle glucose uptake, independent of skeletal muscle blood flow. Currently there is no means to provide NO supplementation through the diet, as it is a gas, thus BRJ and its high NO_3^- concentration is used as a means to generate NO endogenously. In fact, up until this point, much of the support for NO use to improve exercise performance has relied heavily on "borrowed science" using amino acids such as L-arginine. Much more impressive is the growing body of scientific data in support of whole food sources of inorganic NO_3^-, such as that found in BRJ, and improved athletic performance.

Effect of Beetroot Juice on Exercise Performance in Normoxic and Moderate Hypoxia: At altitude, decreases in the barometric pressure and the alveolar partial pressure of oxygen result in less oxygen reaching the tissues. The drop in available oxygen detrimentally affects aerobic capacity. For example, O2max decreases by ~6%/1000 m between 300 m and 2800 m above sea level. These factors combine to reduce overall training quality, particularly with acute hypoxic exposure in absence of acclimatization. As athletes often train and compete at moderate altitude (i.e., 1500-2500 m), the ability to maintain high exercise intensity is important for performance.

Beetroot juice (BR), a supplement with a naturally high concentration of nitrate (NO3-) has gained popularity with endurance athletes aiming to improve performance. There is evidence to suggest that dietary NO3- improves oxygen delivery, lowers blood pressure during exercise recovery, and increases mitochondrial efficiency through an improved oxidative phosphorylation ratio. When oxygen is less available from the environment (e.g. at altitude), BR may have even greater physiological effects; however, the efficacy of acute BR supplementation for exercise performance in moderate hypoxia requires further research. As hypoxia limits aerobic capacity, the potential benefits of BR are particularly relevant to aerobic exercise.

Acute Dose of Beet Root Juice Does Not Improve Endurance Performance: Supplements thought to increase nitric oxide (NO) have received considerable attention as an ergogenic aid in both the scientific community and among athletes. NO is a signaling molecule which has a number of physiological roles in the body, including the regulation of vascular tone, immune function, neurotransmission, muscle contractibility, and energy metabolism. Nitrite (NO,) is a bioactive source of NO and levels in the plasma can be increased by consuming foods that are high in nitrate (NO,), such as leafy green vegetables and beetroot. Improvements in whole body exercise efficiency, tolerance, and performance have been reported in response to NO, -rich BRJ supplementation. More specifically, the volume of oxygen ($V0_2$) consumed at any given rate of work is reduced; or alternately, the power output (PO) is increased for any sustainable metabolic rate of oxygen utilization. Research has suggested that the increase in exercise efficiency may be due to the reduction in VO,; which may be the result of the decreased energy cost of muscle force production and/or the increased adenosine triphosphate production per unit oxygen consumed. Exercise tolerance has also been reported to be improved with NO.' supplementation. In fact, exercise tolerance during intense cycling has been reported to increase by 3 to 25% during
time-to-exhaustion tests. Performance, as determined by a cycling time trial (TT) test was also shown to improve by 1.2 to 2.8% in moderately trained subjects.

CONCLUSION:

BR supplementation has become popular with athletes; although the few studies investigating highly trained individuals in normoxia indicate that its efficacy may be limited in this population. Aerobic capacity is decreased in hypoxia and highly trained athletes experience greater decrements in endurance performance than untrained individuals in hypoxia. Previous research has indicated that even if athletes do not benefit from BR in normoxia, the supplements may be ergogenic at altitude. The proposed mechanisms of action behind BR supplementation may be particularly effective in hypoxia, where the NOS system and aerobic performance are both limited.

This investigation showed that the original, malt dextrin and gum Arabic based reconstituted beetroot juices behaved like Newtonian fluid. The Newtonian viscosity (η) ranges from 4.47 to 86.99, 4.76 to 176.15 and 5.60 to 1561.77 mPa s for original, malt dextrin (MD) and gum Arabic (GA) based juices respectively, depending upon the solid content and temperature used. The results indicated that the Newtonian viscosity increased significantly ($p<0.05$) with increase in solid content, whereas it decreased significantly ($p<0.05$) with increase in temperature. Among all the carrier materials, gum Arabic based juice showed maximum viscosity followed by maltodextrin and original juice at same solid content and temperature studied. The Arrhenius equation was able to describe the temperature dependency of Newtonian viscosity of beetroot juice. The flow activation energy (Ea) was markedly affected by type of carrier material and is increased significantly ($p<0.05$) with increase in solid content. The Newtonian viscosity of reconstituted spray dried beetroot juice increased with solid content and a significant ($p<0.05$) change was observed with different temperatures used and also markedly affected by type of carrier material. A combined single equation relating Newtonian viscosity (η) to solid content and temperature of different carrier material reconstituted beetroot juice was established. The results showed that the Newtonian viscosity of reconstituted beetroot juice was dependent on solid content, temperature and type of carrier material.

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