



## Randomized control trials

## Increasing the protein to carbohydrate ratio in yogurts consumed as a snack reduces post-consumption glycemia independent of insulin

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## SUMMARY

**Background & aims:** We aimed to compare the effects of protein to carbohydrate ratio and physical form in dairy on glucose homeostasis and food intake.

**Methods:** In a crossover design, 20 healthy males consumed 250 g of one of five treatments, plain yogurt, plain yogurt with honey, strawberry yogurt, skim milk or orange juice, as mid-morning snacks. Food intake was assessed 120 min later. Blood glucose, serum insulin and subjective satiety were measured pre- and post-meal.

**Results:** Pre-meal glucose responses were attenuated in a dose-dependent manner to the increasing protein and decreasing sugars in dairy. Protein to carbohydrate ratio correlated negatively with pre-meal glucose due to improved efficacy of insulin action rather than to increased insulin concentrations. Compared with a carbohydrate beverage (orange juice), cumulative blood glucose was lower after dairy snacks but the effect was not explained by their protein to carbohydrate ratio or physical form. Skim milk, with the lowest protein to carbohydrate ratio among dairy products, attenuated both pre-meal and post-meal glucose compared to orange juice without inducing higher insulin levels. There was no effect of treatments on appetite and food intake.

**Conclusions:** While pre-meal glycemia was attenuated dose-dependently to increased protein to carbohydrate ratio in dairy snacks, the contribution of dairy products to post-meal glucose control and to satiety and food intake was independent of their protein to carbohydrate ratio and physical form in healthy men.

This trial was registered at [clinicaltrials.gov](http://clinicaltrials.gov) as NCT01673321.

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## 1. Introduction

Metabolic syndrome, characterized by abdominal obesity, insulin resistance, hypertension, atherogenic dyslipidemia and a low-grade pro-inflammatory state, is a leading public health problem worldwide. Regulation of satiety and food intake as well as modulation of glucose homeostasis constitute a major cornerstone in the management of obesity and insulin resistance, common

denominators of metabolic syndrome. A well-balanced dietary approach is the most preferred strategy for the prevention and treatment of metabolic syndrome. Foods, food components and dietary practices that can contribute to reduced risk of metabolic syndrome-related co-morbidities need to be identified. Dairy consumption is inversely associated with the incidence of metabolic syndrome and type 2 diabetes in a number of epidemiological studies.<sup>1</sup> Observational studies similarly report an inverse relationship between dairy intake and body weight.<sup>2</sup> As a result, dairy products have been postulated to reduce the risk of components of the metabolic syndrome including obesity and glycemic dysregulation.

The role of dairy in the management of metabolic syndrome has focused on milk, but yogurt, the fermented version of milk, has been of more recent interest especially upon the introduction of Greek yogurt into the United States in 2008. Some epidemiological

**Abbreviations:** PCR, protein to carbohydrate ratio; BMI, body mass index; VAS, visual analog scale; ANOVA, analysis of variance; iAUC, incremental area under the curve.

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studies show yogurt to have cardio-protective properties<sup>3</sup> and to exert protective effects against type 2 diabetes<sup>1</sup> when compared to other dairy products. Beneficial effects of yogurt on body weight and body composition have also been described. In a study on 120,877 US adults from 3 large cohorts, increased yogurt intake, but not other dairy products, was inversely associated with weight gain.<sup>4</sup> On the other hand, experimental findings of the effects of yogurt on postprandial satiety and subsequent energy intake are contradictory. In obese females, the consumption of yogurt at meals or at least four times per week over 10 weeks failed to reduce energy intakes.<sup>5</sup> In contrast, short-term studies show that yogurt may be a preferred pre-meal snack. In a comparison of the effects of strawberry-flavored liquid yogurt and chocolate bars as afternoon snacks on satiety and subsequent energy intake at a dinner meal, participants experienced a lower level of motivation to eat when consuming the liquid yogurt.<sup>6</sup> Similarly, a 160 kCal-yogurt afternoon snack reduced hunger, increased fullness and delayed request for dinner versus no snacking<sup>7,8</sup> and delayed subsequent eating compared to crackers and chocolate<sup>9</sup> in healthy pre-menopausal women.

Satiating properties of yogurt have been attributed to milk proteins.<sup>10</sup> Consequently, Greek yogurt with elevated protein content, in the range of 20–24 g/serving which is almost five times the protein content of regular yogurt products, has been introduced and marketed as ideal snack options for satiety, food intake and body weight management. A threshold protein concentration in yogurt appeared to be needed to elicit beneficial impact on satiety regulation in healthy women. A yogurt snack containing 14 g of protein/160 kCal did not stimulate greater subjective satiety in comparison to a yogurt snack containing 5 g protein/160 kCal.<sup>8</sup> However, yogurt afternoon snacks with 24 g protein/160 kCal (250 mL) resulted in greater increases in satiety and a longer delay in request for dinner compared to yogurt products with lower protein content.<sup>7</sup> It is unclear if this unique property of proteins in yogurt as a result of fermentation accounts for these benefits or if it is similar to dairy with intact proteins. In one comparison, all dairy snacks, including milk, yogurt and cheese, lowered energy intake relative to water, but greater reductions in appetite were reported with the yogurt snack in overweight men.<sup>11</sup> Short-term studies have also shown that milk proteins, when consumed alone in beverage form or with carbohydrate, reduce glycemia.<sup>12,13</sup> The effect of consuming yogurt as a whole food on short-term glucose responses is not known.

Therefore, we hypothesized that yogurt consumption as a mid-morning snack will lead to greater control of glycemia and suppression of appetite and subsequent food intake than milk, responses that would reflect an increase in their protein to carbohydrate ratio (PCR). The objective of the current study was to compare the effects of milk and Greek yogurts of increasing protein content on postprandial glucose, insulin and subjective satiety responses and on food intake at the next meal in healthy adult men.

## 2. Materials and methods

### 2.1. Participants

Using a single-blinded within-subject repeated-measures randomized design, 20 healthy males, between the ages of 20–30 years and with a body mass index (BMI) ranging from 20 to 24.9 kg/m<sup>2</sup>, were recruited through advertisements in the University of Toronto campus. Breakfast skippers, smokers, dieters, individuals with lactose intolerance, allergies to milk and dairy products, gastrointestinal problems, diabetes or other metabolic diseases and those scoring  $\geq 11$  on an Eating Habit Questionnaire were excluded from the study. The sample size was determined

with a power analysis for a within-subject design from previous studies,<sup>12,13</sup> to detect a 150 kCal difference in food intake with a power of 0.80 and an alpha of  $<0.05$ . Procedures were approved by the University of Toronto Health Sciences Research Ethics Board.

### 2.2. Treatments

Treatments included: 1) Non-fat Yogurt-Plain (Athenos Greek Yogurt, Kraft Foods, Chicago, Ill) (PCR: 2.30); 2) Non-fat Yogurt with honey-Plain (Athenos Greek Yogurt, Kraft Foods, Chicago, Ill) (PCR: 1.24); 3) Non-fat Yogurt-Strawberry flavored (Athenos Greek Yogurt, Kraft Foods, Chicago, Ill) (PCR: 0.79); 4) Skim milk (Selection Skim Milk Powder, Canada) (PCR: 0.69); and 5) Orange juice (Minute Maid Frozen Juice Concentrate, Toronto, Canada) (PCR: 0.07). Plain yogurt product was prepared by Kraft Foods from skim milk through fermentation by live cultures, mainly composed of lactic acid bacteria. Milk was heated prior to culturing, in order to capture most of the whey proteins with caseins. Heat application denatures whey protein in skim milk, which facilitates the cross-linking of denatured whey with intact casein micelles within the gel network and improves various physical properties of yogurt including viscosity and water-holding capacity. Afterward, water and lactose were almost all separated from the proteinous phase through centrifugation. The ratio of whey to casein in the plain yogurt product was maintained similar to that of skim milk (20:80 whey:casein). A strawberry fruit blend (26% sucrose and 70% moisture) provided by Kraft Foods was mixed with the plain yogurt for the preparation of the strawberry-flavored yogurt product; whereas, honey (Billybee Pure Natural Honey, McCormick, Canada), having 82% sugar distributed as 36% glucose, 41% fructose, 3% galactose and 1% maltose, was mixed with the plain yogurt for the preparation of the plain yogurt product with honey. Orange juice was included as a protein-free control because sugar-containing beverages are frequently consumed between meals, and as well to allow a comparison of lactose and sucrose effects on glycemia. The nutritional composition of each treatment is outlined in Table 1. All products were isovolumetric (250 g), based on the commercially available serving size of dairy products. Treatments were served chilled.<sup>16</sup>

**Table 1**  
Nutritional information of treatments.

Nutritional information <sup>a</sup>	Treatments				
	Plain yogurt	Plain yogurt with honey	Strawberry yogurt	Skim milk	Orange juice
Weight (g)	250	250 <sup>b</sup>	250 <sup>c</sup>	250 <sup>d</sup>	250 <sup>e</sup>
Calories (kCal)	141	165	167	166	141
Protein (g)	23.1	22.2	18.3	16.6	2.4
Energy (%)	65.5	53.8	43.8	40.0	6.7
Total carbohydrate (g)	10.0	17.9	23.3	23.9	32.9
Energy (%)	28.4	43.4	55.8	57.6	93.3
Lactose (g)	10.0	9.6	8.0	23.9	0
Sucrose (g)	0	0	13	0	29.4
Mixture of sugars (g) <sup>f</sup>	0	8.3	2.3	0	3.5
Fat (g)	0.96	0.51	0.07	0.44	0
Energy (%)	6.1	2.8	0.4	2.4	0
Protein to carbohydrate ratio	2.30	1.24	0.79	0.69	0.07

<sup>a</sup> Nutritional information of each treatment as provided by the manufacturer.

<sup>b</sup> Prepared as 240 g of plain yogurt with 10 g of honey.

<sup>c</sup> Prepared as 120 g of plain yogurt with 30 g of strawberry fruit blend (strawberries, sugar, water, corn starch, cranberry juice concentrate (color), lemon juice concentrate, natural flavor and black carrot juice (color)).

<sup>d</sup> Prepared as 46 g of skim milk powder in 250 mL of water.

<sup>e</sup> Prepared as 74 mL of orange juice concentrate in 250 mL of water.

<sup>f</sup> Glucose, fructose, galactose and maltose for plain yogurt with honey; and fructose for strawberry yogurt and orange juice.

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