Plasma amino acids response to ingestion of breakfast cereal with milk containing enhanced protein concentration and modified casein:whey ratio by healthy adults

Bonnie Kung*, Sylvie L. Turgeon§, Amanda J. Wright†, G. Harvey Anderson‡, H. Douglas Goff*

*Department of Food Science, University of Guelph, § STELA Dairy Research Centre, Institute of Nutrition and Functional Foods (INAF), Université Laval, Quebec City, Canada
† Department of Human Health & Nutritional Sciences, University of Guelph, ‡ Department of Nutritional Sciences, Faculty of Medicine, University of Toronto.

Introduction & Background

Dairy proteins have been associated with reduced insulin resistance by attenuation of blood glucose (BG) response.[1] In our previous study, healthy men and women (n=32) consumed 58 g of Honey Nut Cheerios breakfast cereal, along with 250 mL of a beverage treatment of low (3.1 wt%) or high (9.3 wt%) protein concentration and normal (80:20) or modified (40:60) casein to whey ratio. Both milk treatments attenuated 2 hr BG ($P<0.01$) compared to a dairy protein-free control, but there was no difference in area under the curve based on milk protein ratio ($P=0.91$).[2] Reduction of BG from enhanced protein concentration could be potentially related to the reported "fast" whey digestibility compared to "slow" casein[3]. Thus this study aims to elucidate the mechanisms behind BG reductions by investigating the impact of protein concentration and casein:whey ratio on gastric emptying (by indirect paracetamol method). It was hypothesized that increased plasma amino acids (AAs) from consumption of a higher whey breakfast would result in slower gastric emptying compared to casein consumption.

Methodology

- **Study design:** Randomized, double-blinded, repeated measure for 5 study visits
- **Inclusions:** Healthy young adults, 18-30 y, BMI 20-24.9 kg/m^2, normal fasting BG (3.5 – 5.5 mmol/L)
- **Participant baselines:** n=12 (6 M/6 F), 22.8 ± 3.0 y, BMI 23.2 ± 2.7 kg/m^2
- **Timepoints:** Pre-lunch (0, 30, 60 and 120) and post-lunch (140, 170 and 200 min)
- At 120 min, caloric intake from an *ad libitum* 5-inch cheese pizza was recorded[2]
- Venous blood sample was collected by indwelling catheter with an IV line
- Samples were centrifuged and frozen at -80°C until analyses, whereby
  - Plasma paracetamol was analyzed by ELISA at the University of Toronto, ON[4]
  - Plasma AAs by EZ-Faast (Megazyme Inc.) and GC-FID at Université Laval, QC

Preparation of breakfast

Available carbohydrates were standardized at 76.7 g

Table 1. Protein (g), amino acids (g) composition and caloric value (kcal) for the breakfast meal (milk 250 mL and cereal 58 g)

Treatment	Description	Total Protein / *BCAA*	Casein / *EAA*	Casein / *NEAA*	Whey / *TAA*	Calories (kcal)
		(g)				
Control	Water and Permeate	**4.6** *0.78*	*1.60*	-- *2.70*	**0.03** *4.30*	403.8
3.1 wt% MP (80:20)	Normal protein (MP) conc. milk with normal casein to whey ratio (80:20)	**12.4** *2.43*	*5.05*	**6.2** *6.97*	**1.6** *12.02*	433.1
3.1 wt% MP (40:60)	Normal protein (MP) conc. milk with modified casein to whey ratio (40:60)	**12.4** *2.50*	*5.18*	**3.1** *6.86*	**4.8** *12.04*	434.3
9.3 wt% MP (80:20)	Higher milk protein (MP) conc. (3x) milk with normal casein to whey ratio (80:20)	**27.9** 5.74	11.95	**0.95** 15.51	**0.24** 27.46	485.6
9.3 wt% MP (40:60)	Higher milk protein (MP) conc. (3x) milk with modif casein to whey ratio (40:60)	**27.9** *5.96*	*12.40*	**0.47** *15.22*	**0.71** *27.62*	499.1

Acknowledgements

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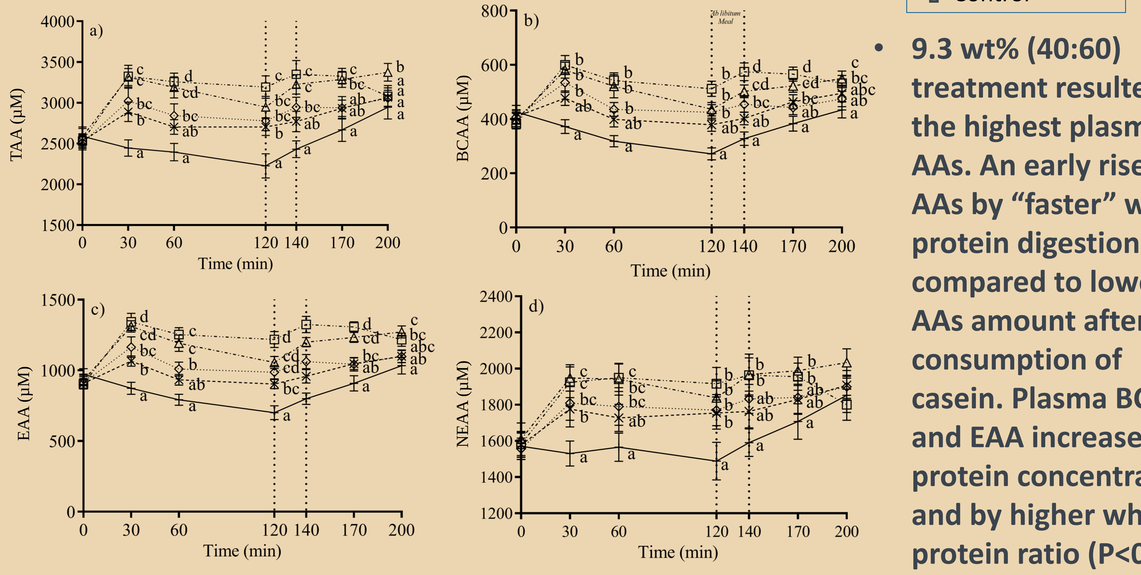
Data Analysis

- AAs were grouped as total (TAA), branched-chain (BCAA), essential (EAA), and non-essential (NEAA)
- Pre-lunch data was analyzed as iAUC and absolute values, and post-lunch was only analyzed as absolute values using an ANOVA followed by Tukey-Kramer's post hoc test for treatment effect ($P<0.05$)

Results

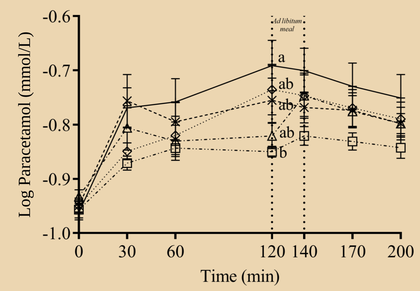
- **There were no differences in lunch caloric intake between milk treatment concentration or ratio ($P>0.05$) (data not shown).**

Figure 1. a) TAA, b) BCAA, c) EAA, d) NEAA from 0 – 200 min.



- **9.3 wt% (40:60) treatment resulted in the highest plasma AAs. An early rise of AAs by "faster" whey protein digestion compared to lower AAs amount after consumption of casein. Plasma BCAA and EAA increased by protein concentration and by higher whey protein ratio ($P<0.05$).**

Figure 2. Plasma Paracetamol



- **Also, 9.3 wt% (40:60) had the lowest paracetamol (iAUC and mean) compared to the control. High protein concentration lowered paracetamol AUC compared to low concentration, and lowered plasma paracetamol due to higher whey protein ratio was observed at 30 min. Thus, whey protein seems to have a greater impact on gastric emptying over casein coagulation.**

Discussion and Conclusion

- This work suggests higher whey protein content results in a higher rise of plasma AAs which is an observation of "fast" and "slow" protein digestion
- Attenuation of BG may be due to slower gastric emptying mechanism, elicited by the rise of AAs, which is influenced by high protein concentration and to a modest extent modified ratio. Thus, slower gastric emptying seems more affected by whey protein over casein
- Furthermore, consumption of the high protein treatments increased GLP-1 and CCK, but there was no insulin difference between treatments[5]

References

[1] Anderson et al. 2011. *Nestle Nutr Workshop Pediatr Prog,* 67:147-59 [2] Kung et al. 2018. *J of Dairy Science*. 101:8688-8701. [3] Hall et al. 2003. *Br. J. of Nutr.* 89:239-248 [4] Akhavan, et al. 2013. *J of Nutr Biochem.* 25(1):36-43. [5] El Khoury et al. 2019. *J of Dairy Science* (Manuscript Accepted).