

The effect of meal replacements high in glycomacropeptide on weight loss and markers of cardiovascular disease risk¹⁻³

Jennifer B Keogh and Peter Clifton

ABSTRACT

Background: Glycomacropeptide (GMP) is a peptide that has been shown to stimulate release of cholecystokinin, which may promote satiety.

Objective: The aim of this one-year study was to examine whether greater weight loss could be achieved and sustained with a GMP-enriched whey powder supplement compared with a skim milk powder supplement.

Design: In a double-blind, randomized, parallel-design study using meal replacements, weight, body composition (determined by dual-energy X-ray absorptiometry), blood pressure, fasting lipids, glucose, and insulin were measured at baseline, 6, and 12 mo. Meal replacements contained 15 g protein from GMP-enriched whey protein isolate (GMP-WPI) or skim milk powder (SMP) and 900 kJ/sachet. Volunteers consumed 2 sachets/d instead of 2 meals for 6 mo and 1 sachet/d for a further 6 mo. Of the 127 participants (95 women, 32 men, 95.5 ± 15.4 kg, body mass index 33.4 ± 3.4 kg/m², 50.0 ± 12.4 y), 82 completed the 6-mo study and 72 of those completed the 12-mo study.

Results: At 6 mo, weight loss was 9.5 ± 5.8 kg compared with 11.0 ± 6.0, GMP-WPI and SMP, respectively, and 9.9 ± 8.8 kg compared with 10.8 ± 7.4 GMP-WPI and SMP, respectively, at 12 mo ($P < 0.001$ compared with baseline, at both timepoints) with no differences between treatments. Total and LDL cholesterol, triacylglycerols, glucose, insulin, and systolic and diastolic blood pressure decreased at 6 and 12 mo (all $P < 0.01$ compared with baseline with no difference between treatments). HDL cholesterol increased at 12 mo ($P < 0.001$ compared with baseline).

Conclusions: Meal replacements containing GMP had no additional effect on the overall sustained 12-mo weight loss of 10 kg. There were improvements in cardiovascular disease risk markers. *Am J Clin Nutr* 2008;87:1602-5.

INTRODUCTION

Glycomacropeptide (GMP) is a C-terminal fragment of kappa casein (residues 106-169) released by the endopeptidase chymosin (rennin) and is in whey at a level of 600 mg/L (1, 2). Oral GMP stimulates cholecystokinin (CCK), the leading candidate satiety hormone, which may make this protein a useful component of a weight-loss diet because CCK slows gastric emptying, which may in turn promote satiety (3-6). GMP has also been detected in the blood of volunteers after milk or yogurt ingestion, suggesting that GMP can be formed in the gut and can be absorbed intact into intestinal cells (7). We have previously shown that GMP was associated with reduced fat mass in Wistar rats fed

ad libitum for 7 wk with diets differing in protein type amount (8). To our knowledge the effects of GMP on weight loss or fat mass loss have not been studied in humans.

On an energy basis, protein appears more satiating than the other macronutrients, and 2 long-term studies have shown that protein per se leads to long-term weight loss (9, 10). We have previously observed a difference of 3.4 kg between reported higher and lower protein intakes, which was significant at $P < 0.05$ (10). A higher protein intake has also been shown to limit weight regain after weight loss (11). We have also shown a greater reduction in total and abdominal fat in women with raised triacylglycerol on a higher protein weight-loss diet (12). Very little attention has been paid to protein type during weight loss, and there are no studies investigating the effects of additional GMP during weight loss in human volunteers.

Meal replacements are an effective strategy for weight loss and weight-loss maintenance (13-15). In a study with twice-daily partial meal replacements (Slimfast; Unilever, Sydney, Australia), we observed a 9-kg weight loss at 6 mo (with a 70% retention rate) (14).

The objectives of the present study were to investigate, in a chronic human feeding study, whether greater sustained weight loss over a 12-mo period would be achieved with a GMP-enriched whey powder supplement compared with a predominantly whole milk powder supplement. Our second objective was to investigate if greater weight loss was due to greater fat loss and was associated with greater falls in cholesterol, triacylglycerol, insulin, and glucose.

SUBJECTS AND METHODS

Overweight and obese men and women (body mass index >27 and <40 kg/m²) aged 20-70 y were recruited by a newspaper advertisement. There were 280 responses of whom 133 were eligible and 127 commenced the study. Volunteers were included if they had no abnormality of clinical significance on medical history including liver and renal disease, unstable cardiac disease, metabolic disease, cancer within the last 5 y (except

¹ From the Commonwealth Scientific and Industrial Research Organization, Human Nutrition, Adelaide, South Australia, Australia.

² Supported in part by Murray Goulburn Nutritionals, Victoria, Australia.

³ Address reprint requests and correspondence to J Keogh, Research Scientist, CSIRO Human Nutrition, PO Box 10041, Adelaide BC, South Australia, 5000, Australia. E-mail: jennifer.keogh@csiro.au.

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nonmelanoma skin cancer), and respiratory disease that caused shortness of breath at rest; if female, they were not pregnant or breastfeeding. Subjects taking lipid-lowering or antihypertensive medication were allowed to enter the study provided the medication doses were not altered during the study. Exclusion criteria included type 1 or 2 diabetes as assessed from their medical questionnaire (no screening was performed); history of heavy alcohol consumption (>5 drinks/d (50 g alcohol)); frequent dining out (>2/wk and unable to cease); inability to meet diet requirements; inability to comprehend or cope with study requirements; hypersensitivity to the test food, current attempt to lose weight or weight loss >5 kg within the previous 6 mo; participation in another research study within 30 days preceding the commencement of this study; a history or presence of gastrointestinal, renal, or hepatic disease of any cause; diagnosis of an eating disorder in the past (as determined by self report); or taking any medication that was likely to affect the study outcomes. The study was approved by the Commonwealth Scientific and Industrial Research Organization (CSIRO) Human Nutrition Human Research Ethics Committee, and volunteers gave informed written consent.

Study design and methodology

This was a double-blind, placebo-controlled, randomized, parallel-design study of 1 y of weight loss using meal replacements containing either GMP-enriched whey protein isolate (GMP-WPI) containing 90% GMP (NatraPep; MG Nutritionals, Victoria, Australia) or skim milk powder (SMP; NatraPro, MG Nutritionals). Both contained 15 g of protein and 900 kJ per pack.

Subjects were blocked matched for age, sex, and body mass index and randomized using Clinstat computer program (Martin Bland, public domain) to receive the GMP-WPI or SMP meal replacements. Volunteers attended for measurement of height, weight, body composition by dual-energy X-ray absorptiometry, systolic (SBP) and diastolic blood pressure (DBP), fasting lipids (on 2 d), and glucose and insulin at baseline, 24 wk, and 1 y (Table 1). Volunteers had a consultation with a dietitian at baseline, 3, and 24 wk and attended the CSIRO clinic every 4 wk for measurement of weight and to collect meal replacements. Compliance with taking the supplements was assessed by a daily checklist and by the volunteer returning the empty sachets. Compliance was not different between treatments (data not shown).

Dietary methodology

Coded packages of protein supplements (Murray Goulburn Nutritionals, Victoria, Australia) were made up to 200 mL with water by the volunteer. Volunteers were advised to consume 2 sachets per day to replace 2 meals and to consume one energy-restricted meal containing 120 g raw weight meat/fish/chicken. They were also advised to eat 2 servings (300 g) of fruit/d, 2 cups cooked and 1 serving raw (salad) vegetables/d, 250 mL reduced-fat milk, and 30 g high-fiber cereal during the day. Bran or psyllium supplements were recommended to relieve constipation. At 6 mo participants were advised to reduce the supplements to one per day and have 2 meals/d and were provided with sample meal plan options.

Blood pressure

Resting blood pressure (mm Hg) was measured (mean of 3) by an automated sphygmomanometer (DYNAMAP 8100; Criticon, Tampa, FL) with subjects seated.

Weight, height, and body composition

Body weight (model AMZ14; Mercury Digital Scales, Tokyo, Japan) was recorded in light clothing without shoes. Height was measured to the nearest 0.1 cm using a stadiometer (SECA, Hamburg, Germany) without shoes.

Total fat mass and total lean mass were assessed by whole-body dual-energy X-ray absorptiometry (Norland densitometer XR36; Norland Medical Systems, Fort Atkinson, WI; CV of $2.3 \pm 0.7\%$ for total body fat mass and $2.1 \pm 0.4\%$ for lean mass) at the Endocrinology Department, Royal Adelaide Hospital, by qualified radiographers.

Biochemical analysis

Fasting blood samples were collected into tubes containing no additives for measurement of lipids and insulin and sodium fluoride/EDTA for glucose measurements. Plasma or serum was isolated by centrifugation at 2000 g for 10 min at 5 °C (Beckman GS-6R centrifuge; Beckman, Irvine, CA) and stored at -80 °C until the end of the study. Serum total cholesterol, HDL cholesterol, triacylglycerol (TAG), and glucose were measured in one run on a Roche Hitachi 902 auto-analyser (Roche Diagnostics Co, Indianapolis, IN) using standard Roche enzymatic kits (Roche Diagnostics, Basel, Switzerland) and control sera. LDL cholesterol was calculated according to the method described by Friedewald (16). Plasma insulin concentrations were determined using a commercial enzyme immunoassay kit (Mercodia ELISA; ALPCO Diagnostics, Uppsala, Sweden).

Statistical analysis

Statistical analyses were carried out using SPSS 14.0 for WINDOWS (SPSS Inc, Chicago, IL). Differences in baseline characteristics between groups were compared using independent *t* tests for continuous variables. Repeated measures analysis of variance with treatment as the between-subjects factor and time (3 timepoints of 0, 6, and 12) as the within-subject factor was

TABLE 1
Weight fat, percent fat, and lean mass at baseline and 12 mo¹

| Treatment ² | 0 mo | 12 mo ³ | <i>n</i> |
|------------------------|-------------|--------------------|----------|
| Weight (kg) | | | |
| GMP-WPI | 98.6 ± 17.4 | 88.6 ± 18.0 | 34 |
| SMP | 94.8 ± 14.7 | 84.0 ± 15.5 | 38 |
| Fat mass (kg) | | | |
| GMP-WPI | 40.4 ± 8.1 | 33.8 ± 7.9 | 34 |
| SMP | 41.4 ± 8.5 | 33.8 ± 9.2 | 38 |
| Percent fat (%) | | | |
| GMP-WPI | 43.6 ± 9.1 | 40.8 ± 8.6 | 34 |
| SMP | 45.8 ± 8.0 | 41.9 ± 9.0 | 38 |
| Midriff fat (kg) | | | |
| GMP-WPI | 3.9 ± 1.2 | 3.0 ± 1.2 | 34 |
| SMP | 3.4 ± 1.1 | 2.7 ± 1.1 | 38 |
| Lean mass (kg) | | | |
| GMP-WPI | 52.2 ± 15.0 | 50.4 ± 15.0 | 34 |
| SMP | 49.8 ± 12.2 | 47.3 ± 12.2 | 38 |

¹ Data are $\bar{x} \pm$ SD. There were no differences between the groups at baseline. Data were analyzed using repeated measures ANOVA with treatment as the between subjects factor.

² GMP-WPI, GMP-enriched whey protein isolate; SMP, skim milk powder.

³ $P < 0.001$, main effect of time (no treatment interaction).

used to assess the effects of the treatment. Statistical significance was set at $P \leq 0.05$. All data are presented as means \pm SDs, unless otherwise stated.

We had 80% power, $P < 0.05$ using a one-tailed t test, to detect a greater loss of 4 kg on GMP compared with control.

RESULTS

One hundred twenty-seven participants (GMP-WPI, $n = 63$, 47 women, 16 men; SMP $n = 64$, 48 women, 16 men) commenced the study. Subjects were not different at baseline (49.6 ± 12.3 compared with 50.3 ± 12.4 y, BMI 34.4 ± 3.7 compared with 34.4 ± 3.7 kg/m², 96.3 ± 15.7 compared with 96.7 ± 16.6 kg GMP-WPI and SMP, respectively). Eighty-two (GMP-WPI, $n = 42$, 28 women, 14 men; SMP, $n = 42$, 31 women, 11 men) completed 6 mo, and 72 (GMP-WPI, $n = 34$, 22 women, 12 men; SMP, $n = 38$, 29 women, 9 men), 12 mo.

Weight loss and body composition

Weight loss at 24 wk was 10.3 ± 5.8 and 11.0 ± 5.8 kg (9.8 ± 5.2 and $11.3 \pm 5.8\%$), and at 1 y was 9.9 ± 8.8 and 10.8 ± 7.4 kg (10.1 ± 7.8 and $11.4 \pm 7.8\%$), GMP-WPI and SMP, respectively ($P < 0.001$, with no difference between the groups; Table 1). The time course for weight loss is presented in **Figure 1**.

Total fat mass decreased by 7.7 ± 4.4 and 8.3 ± 5.2 kg at 24 wk and by 6.6 ± 6.1 and 7.7 ± 6.3 kg at 1 y, GMP-WPI and SMP, respectively ($P < 0.001$, with no difference between treatments). Lean mass also decreased by 2.1 ± 2.7 and 1.8 ± 2.4 at 24 wk and by 2.5 ± 2.3 and 2.5 ± 2.0 kg at 1 y, GMP-WPI and SMP, respectively ($P < 0.001$, with no differences between treatments; Table 1).

Lipids, glucose, and insulin

At the end of the study total and LDL cholesterol were decreased by 0.3 ± 0.5 and 0.4 ± 0.7 mmol/L, GMP-WPI and SMP, respectively, ($P < 0.001$, no difference between treatments; **Table 2**).

TAG also decreased at 12 mo, 0.3 ± 0.5 and 0.3 ± 0.5 mmol/L, GMP-WPI and SMP respectively ($P < 0.001$, no difference between treatments; **Table 2**).

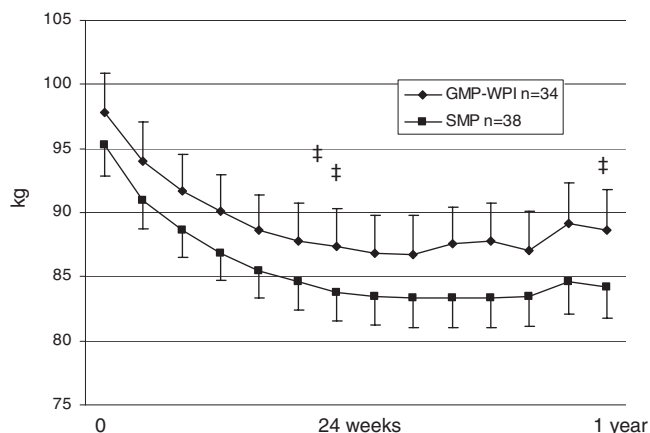


FIGURE 1. Weight loss in participants who completed the 12-mo study. There were no differences between the groups at baseline. Data were analyzed using repeated measures ANOVA with treatment as the between-subjects factor. $^{\ddagger}P < 0.001$, main effect of time (no treatment interaction). Data are mean \pm SEM.

TABLE 2

Lipids, glucose, and insulin at baseline and 12 mo¹

| Treatment ² | 0 mo | 12 mo ³ | n |
|------------------------|----------------|--------------------|----|
| TC (mmol/L) | | | |
| GMP-WPI | 5.2 \pm 0.7 | 4.9 \pm 0.8 | 34 |
| SMP | 5.6 \pm 1.1 | 5.1 \pm 1.0 | 38 |
| LDL-C (mmol/L) | | | |
| GMP-WPI | 3.2 \pm 0.7 | 2.9 \pm 0.7 | 34 |
| SMP | 3.4 \pm 0.9 | 3.0 \pm 0.9 | 38 |
| HDL-C (mmol/L) | | | |
| GMP-WPI | 1.3 \pm 0.3 | 1.4 \pm 0.3 | 34 |
| SMP | 1.5 \pm 0.4 | 1.6 \pm 0.4 | 38 |
| TAG (mmol/L) | | | |
| GMP-WPI | 1.6 \pm 0.5 | 1.3 \pm 0.5 | 34 |
| SMP | 1.5 \pm 0.7 | 1.2 \pm 0.6 | 38 |
| Glucose (mmol/L) | | | |
| GMP-WPI | 5.6 \pm 0.4 | 5.4 \pm 0.5 | 34 |
| SMP | 5.3 \pm 0.4 | 5.2 \pm 0.4 | 38 |
| Insulin (mIU/L) | | | |
| GMP-WPI | 10.0 \pm 4.1 | 7.2 \pm 3.5 | |
| SMP | 10.2 \pm 5.2 | 7.8 \pm 5.3 | |

¹ Data are $\bar{x} \pm$ SD. There were no differences between the groups at baseline. Data were analyzed using repeated measures ANOVA with treatment as the between subjects factor.

² GMP-WPI, GMP-enriched whey protein isolate; SMP, skim milk powder; TC, total cholesterol; LDL-C, LDL cholesterol; HDL-C, HDL cholesterol; TAG, triacylglycerol.

³ $P < 0.001$, main effect of time (no treatment interaction).

HDL cholesterol increased at 12 mo, 1.3 ± 0.3 to 1.4 ± 0.4 mmol/L and 1.5 ± 0.4 to 1.6 ± 0.4 mmol/L, GMP-WPI and SMP, respectively ($P < 0.001$ compared with baseline; **Table 2**).

Glucose decreased by $2.4 \pm 5.7\%$ and $2.8 \pm 4.8\%$, GMP-WPI compared with SMP, respectively at 12 mo ($P < 0.001$, with no difference between treatments; **Table 2**).

Insulin decreased by $26.3 \pm 21.4\%$ and 24.7 ± 27.6 , GMP-WPI compared with SMP, respectively ($P < 0.001$, with no difference between treatments) at 12 mo (**Table 2**).

Blood pressure

At baseline SBP was 132 ± 17 and 132 ± 15 mm Hg, and DBP was 77 ± 10 and 73 ± 9 mm Hg, GMP-WPI and SMP, respectively (NS). Blood pressure was decreased at 12 mo, SBP by 8 ± 17 and 10 ± 11 mm Hg, and DBP by 5 ± 12 and 3 ± 10 mm Hg, GMP-WPI and SMP, respectively (both $P < 0.01$, no treatment effect).

DISCUSSION

The main finding of this study was that whereas participants achieved substantial weight loss of 10 kg after 6 mo that was sustained at 12 mo, using protein-enriched meal replacements containing GMP had no additional effect. Overall, there were health benefits of sustained weight loss in all participants, irrespective of type of meal replacement, as glucose, insulin, LDL cholesterol, and TAG remained reduced compared with baseline, and HDL cholesterol was significantly increased.

Evidence supporting the efficacy of dairy peptides in weight loss is limited. Hall et al (17) showed that a 48-g whey preload reduced food intake at a buffet meal 90 min later compared with a similar amount of casein and that this was associated with

increases in CCK and gastrin-inhibiting peptide as well as total amino acids. It is possible that increased content of GMP in whey could have played a role in this finding. We have previously shown that GMP used in this study was associated with reduced fat mass in Wistar rats fed ad libitum for 7 wk with diets differing in protein type amount (8), whereas Gustafson et al (18), in an acute study in human volunteers of the effect of a preload drink containing caseinomacropeptide on satiety and satiation, found it had no effect on energy intake or on subjective indicators of satiety.

GMP is not a well-defined ingredient and may vary dramatically in degree of glycosylation so that, although this study is negative, it does not exclude the possibility that alternative forms of GMP may be more active in releasing CCK, enhancing satiety, and improving weight loss.

In this study the dose of GMP was 27 g/d for 6 mo, which was reduced to 13.5g/d after 6 mo. However, there is limited literature in the area of weight loss and dairy peptides, and it was not possible at this time to estimate the effective dose of GMP. In our previous study in rats (8), the dose of GMP used was much greater than in the present study. Thus, it is possible that insufficient GMP was provided in this study.

In conclusion meal replacements containing GMP had no additional effect on the biologically significant weight loss achieved in this study. Similarly to other studies, improvements in cardiovascular disease risk markers were observed.

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The authors' responsibilities were as follows—JK and PC: designed the study; JK: performed statistical analysis and wrote the manuscript; and PC supervised the study, the statistical analysis, and the manuscript. The authors had no conflict of interest in relation to work reported in this manuscript.

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Erratum

Heymisfield SB, Gallagher D, Mayer L, Beetsch J, Pietrobelli A. Scaling of human body composition to stature: new insights into body mass index. *Am J Clin Nutr* 2007;86:82–91.

Figures 1 and 2 are reversed. The charts shown in Figure 1 actually depict data for women, not men. Similarly, the charts in Figure 2 actually depict data for men, not women. The title for Figure 1 should therefore read as follows: “Weight and body composition versus height of women in the NY-1 database group.” The title for Figure 2 should read as follows: “Weight and body composition versus height of men in the NY-1 database group.” In addition, on page 84, the third sentence of the subsection “Allometric analyses” should read as follows: “The associations of weight and each component with height, fitted with univariate regression models, are presented in **Figure 1** and **Figure 2** for NY-1 women and men, respectively.”

Erratum

Keogh JB, Clifton P. The effect of meal replacements high in glycomacropeptide on weight loss and markers of cardiovascular disease risk. *Am J Clin Nutr* 2008;87:1602–5.

On page 1602, the second sentence of the “Design” section of the abstract should read as follows: “Meal replacements contained 33 g protein from GMP-enriched whey protein isolate (GMP-WPI) or skim milk powder (SMP) and 954 kJ/sachet.” A similar error occurred in the second sentence of the first full paragraph in the “Study design and methodology” subsection of the “Subjects and Methods” section (page 1603). That sentence should read as follows: “Both contained 33 g of protein and 954 kJ per pack.”