

Weight management & satiety effects of whey proteins

Introduction

Weight management is a high priority in western societies, considering the prevalence of obesity and its associated disease risks, which include cardiovascular disease, type 2 diabetes, cancer, inflammatory diseases and degenerative diseases of the joints. Obesity is a global epidemic that has proven daunting to prevent and treat. The prevalence of obesity (BMI>30) and morbid obesity (BMI>40) among American adults is about 30% and 5% respectively [1]. Obesity is so strongly associated with medical comorbidities (a disease or disorder that is not directly caused by another disorder but occurs at the same time) and mortality, that it has begun to overtake infectious diseases as the most significant contributor to ill health worldwide [2,3]. Overweight people are also frequently stigmatised and consequently suffer from low self esteem. The traditional paradigm of diet, exercise and medication generally achieves no more than a 5-10% reduction in body weight [4,5]. After such weight loss, over 90% of people return to their original body weight within 5 years [6,7]. This failure arises because a robust homeostatic system of body weight reduction compensates for weight loss with increased hunger and decreased energy expenditure [8,9].

Reduction of energy intake and enhancement of physical activity is thought to be the key to the prevention of obesity and the reduction of body mass in overweight people. Much attention is given nowadays to the importance of high protein diets in weight management [10-14].

The main reason for this is that dietary proteins cause short-term satiety and have a stronger satiation value than either carbohydrates or fats. This could help the body to comply better with energy reduction and maintain the lower body weight obtained after weight loss [15,16].

Satiety is defined as the post-ingestive or inter-meal satiety, which appears when the meal is ended, ie the feeling of fullness having eaten and the time lapse before the desire to eat again [10]. Any endogenous factor that causes a sensation of fullness and reduces consumption is called a satiety signal and several different gastrointestinal peptides are thought to have this effect.

The exact mechanistic explanation for the higher satiation value of proteins is unknown, but it is proposed that it could be related to the following three reasons:

1. Diet induced thermogenesis (generation or production of heat, by physiological processes)
2. Raised level of certain amino acids in the plasma in the postprandial (post feeding / eating a meal) state
3. Effects on gut hormone levels, playing a role in the satiation / satiety cascade.



Diet-induced thermogenesis

The thermic response (rise in internal temperature of the body) to ingestion of protein is 50 to 100% higher than that of carbohydrate. In a study by Crovetti et al [17], a high protein meal providing 2331 kJ, given to ten healthy women, produced a thermogenic effect of 261 kJ over 7 hours following consumption. This was almost 200% higher than that from either an iso-energetic high carbohydrate meal or a high fat meal. The thermogenic effect of the protein was associated with a significantly enhanced sensation of fullness, however, the research indicates that this thermogenic response is dependent on the type of protein consumed. 'Fast proteins', such as those found in whey, are rapidly absorbed and demonstrate a stronger thermogenic effect and satiety than 'slow proteins' [12]. Thus, correlation exists between the thermogenic response after a meal and satiety. It has been postulated that the increase in temperature on ingestion of food could be sensed by temperature-sensitive neurons in the hypothalamus in the brain, which, in turn, could control the brain's food intake circuitry [18]. Various studies on protein intake, thermogenesis and satiety support the thermostatic theory as one of the possible mechanisms that could explain how whey protein enhances short-term satiety.

Elevated level of plasma amino acids

It has been shown that a postprandial rise in concentration of plasma amino acids is associated with a loss of appetite [19,20]. Studies have shown that a high protein meal exceeds the body's anabolic (protein synthesis) and catabolic (protein utilisation) capacities, leading to a positive protein balance [21]. This results in elevation of plasma amino acids, loss of appetite and slower gastric emptying. This data would support the view that satiety by high protein intake is caused by raised levels of plasma amino acids. Other data shows that satiety by dietary proteins is probably mediated by satiety signals reaching the brain via opioid receptors in the gut, reducing stomach emptying or gut hormone receptors for cholecystokinin (CCK) and glucagon-like peptide 1 (GLP-1) [22].

In addition to the amount of protein consumed, the protein source is also a significant factor for satiety. Boirie et al [23] demonstrated that ingestion of 'fast' whey proteins in humans resulted in a dramatic but short increase in plasma amino acid levels. This is associated with a stronger stimulation of protein synthesis when compared to ingestion of the 'slow' casein, which resulted in a more prolonged plateau of moderately elevated plasma amino acid levels and a much slower rate of protein synthesis.

Gut hormones

Gut hormones secreted in the blood in response to a meal or fasting are known to influence the digestion process and to be involved in regulation of satiety / satiety. Effects include secretion of digestive enzymes, stimulation / inhibition of gastrointestinal motility, bile bladder contraction and nerve stimulation, as well as direct effects on the brain.

In the context of food intake, many gut hormones act on the hypothalamic and brain stem centres of appetite control. This provides a means by which the gut may signal energy status to the seat of satiety – the central nervous system.

GLP 1 and 2, CCK, ghrelin, peptide YY (PYY) and glucose-dependent insulinotropic polypeptide (GIP) are known to influence satiety / satiety.

Gastrointestinal peptides that influence food intake [24]

Reduce consumption	Increase consumption
CCK Glucagon Glucagon like Peptide 1 and 2 Apolipoprotein A-IV Amylin Somatostatin Enterostatin Peptide YY	Ghrelin (presence is stimulated by hunger and a desire to eat)

CCK

This was the first gut peptide to be implicated in the control of appetite. It is synthesised in a number of tissues in humans, including I-cells in the small intestine [25], from where it is rapidly released into circulation following a meal [26]. The concentration of CCK remains elevated for up to 5 hours after consumption and dietary fat and protein are more potent stimulators of CCK release than carbohydrate.

When CCK is secreted, it enters the blood and stimulates the pancreas and liver / gallbladder to secrete appropriate enzymes to facilitate the digestive process in the duodenum. Experiments where CCK is fed to rats before a meal resulted in a reduction in the amount consumed. Dozens of experiments have demonstrated the role of CCK in eliciting satiety. This is shown by the observation that the administration of specific CCK-1 receptor antagonists before a meal causes increased consumption in animals and humans and reduces the subjective feeling of satiety in humans [24,27].

Glycomacropeptide, the glycosylated portion of caseinomacropeptide, is present in sweet whey formed after cleavage of k-casein by rennin. This protein is absent from acid whey, which is formed when caseins are precipitated by lowering the pH to 4.6.

Glycomacropeptide is a powerful stimulator of CCK [28]. Thus, whey protein ingredients manufactured from sweet whey containing glycomacropeptide, or the individually fractionated GMP portion, could act as useful ingredients in food products to switch on CCK, which in turn will suppress appetite and lead to reduced intake of food. This, in association with a low calorie diet and regular exercise, can be used to reduce body weight in people who are overweight and obese.

Ghrelin

The endocrine peptide hormone, ghrelin, was identified in 1999 as the endogenous ligand (an effector – a molecule that binds to a site on a macromolecule's surface by intermolecular forces, thereby changing the chemical conformation of the macromolecule) for the type 1a growth hormone secretagogue receptor (GHS-R1a). Soon after its discovery, ghrelin was found to act as a powerful stimulant of food intake [29-33]. Ghrelin is thought to be the most powerful orexigenic



(stimulating effect on appetite) peptide known to date. In experiments, ghrelin has been shown to cause long-term increase in weight gain and adiposity (laying down of fat in adipose tissue). These long term effects are mediated through the peptide's stimulation of ingestive behaviours, as well as a reduction in energy expenditure and fat utilisation [33].

Ghrelin is a 28 amino acid peptide, of which several variants have been identified. Ghrelin clearly stimulates the brain centres associated with the regulation of appetite and body weight. Numerous studies have demonstrated that this peptide binds to and activates the receptors on the surface of NPY/AgRP neurons in the arcuate nucleus of the hypothalamus in the brain. Ghrelin's role as a factor in meal initiation (the desire to eat) is well supported. The peptide is primarily made in the stomach and upper intestine [34], placing ghrelin's biosynthesis in the organ best suited to sense the acute nutritional supply. Levels of circulating ghrelin in the blood rise during non-ingestive periods (fasting in between meals) and drop immediately after nutrient uptake [35]. Ghrelin triggers food intake in rodents and stimulates food intake in humans. Subjects treated with ghrelin ate on average 30% more at a self service buffet than the control subjects [10]. The resulting bout of feeding following such a ghrelin dose is immediate and quickly passes as one would expect from a meal initiator.

In fasting humans, plasma levels of ghrelin are around 140 ± 14 fmola/ml. Plasma ghrelin concentration peaks in humans at 2AM [36,37]. It increases two-fold before each meal and decreases back to baseline within one hour of eating – a pattern opposite to that of insulin. Plasma ghrelin concentrations are unchanged by water intake. Administration of ghrelin to healthy subjects induces hunger sensations. These results suggest that preprandial elevation of ghrelin is a signal to initiate food intake. Plasma levels are increased by a low protein meal and decreased by a high fat meal. It is profoundly decreased after gastric bypass surgery, suggesting that ingested nutrients are important in inducing ghrelin secretion from the stomach. In general, however, the mechanisms underlying the regulation of ghrelin secretion remain poorly understood [38].

Plasma ghrelin concentration is lower in obese humans and in Pima Indians who have obesity and type-2 diabetes mellitus [36,39,40]. On the contrary, plasma ghrelin is higher under fasting conditions [36,37,42] and in individuals with anorexia, bulimia nervosa, cachexia [41,43-45] (weight loss, wasting of muscle, loss of appetite, and general debility that can occur during a chronic disease) and Prader-Willi syndrome (PWS) – a rare genetic disorder characterised by a severe morbid obesity and unabated hyperphagia (abnormally increased appetite for and consumption of food, thought to be associated with a lesion or injury in the

hypothalamus) [47]. These results, except for the correlations with PWS, suggest that plasma ghrelin levels correlate negatively with body mass index (BMI)[36,40,41,43]. Increased plasma levels in PWS patients might be related to their insatiable appetite and obesity. Plasma ghrelin levels are increased in patients with anorexia nervosa but return to normal after weight gain [41,46].

Blood ghrelin levels do not only rise and fall with ingested meals, they also correspond to the organism's chronic feeding state. Extended periods of caloric restriction lead to an increase in blood ghrelin levels, which quickly return to normal upon return to the pre-diet calorie intake [39]. This observation may well play a role in the poor success of low-calorie diets, which tend to cause initial weight loss followed by quick regain of the pounds shed. The first demonstration of ghrelin's adipogenic properties originated from researchers at Eli Lilly. They showed that repeated injections cause weight gain and fat deposition in mice [42]. In humans, extended treatment has not yet been tested, but patients suffering from PWS displayed elevated ghrelin levels [47-49].

In the majority of obese subjects, ghrelin levels are, on average, lower than in normal weight subjects, but follow the typical prandial pattern [39]. Interestingly, after gastric bypass, the most effective form of obesity treatment to date, patients' blood ghrelin levels are extremely low and show no correlative pattern with meal times. It has been suggested that this reduction of circulating ghrelin contributes to the effectiveness of the procedure (low ghrelin levels, no desire to eat) [48]. In rodents it was recently shown that the effects of gastric bypass on weight and fat loss could be reversed by ghrelin replacement (administered exogenously), highlighting its contribution to the procedure's success [50].

The most conclusive evidence in support of ghrelin as a viable obesity target comes from studies aimed at shutting down the ghrelin pathway. All studies reported a decrease in food intake, and two of the three studies demonstrated a reduction in body weight [32,51,52]. These studies collectively argue in favour of ghrelin antagonism (interference with the physiological activity and switch off) as an approach for obesity treatment [47].



Activity in the pharmaceutical sector – targeting obesity with ghrelin

In summary, there are many lines of evidence that powerfully support the notion that targeting the ghrelin pathway will be beneficial in the treatment of morbid obesity.

It is not surprising then that biotechnology and pharmaceutical companies are actively pursuing the development of ghrelin or ghrelin receptor antagonists. Scientists at Abbot, Ispen are actively searching for antagonists for ghrelin binders [53].

Recently, Cytos of Switzerland announced the beginning of clinical trials for a vaccination targeting endogenous ghrelin. Their highly intriguing approach uses an immunisation technology that directs the immune response to the organism's own harmful molecules, thus mounting an immune reaction to the disease [54]. While the technology is very interesting, the safety of this approach will have to be carefully evaluated. To date, neither the details of how ghrelin is targeted or any in vivo results have been disclosed [47].

NOXXON Pharma AG has developed the compound NOX-B11 – a Spiegelmer (a mirror image nucleic acid generated from a RNA library) that specifically binds acylated ghrelin [55]. Spiegelmer NOX-B11 displays low nanomolar affinity for the ligand ghrelin and neutralises its stimulatory effects on GH release, food intake and neuronal activation in rats [55,56]. When administered sub chronically to diet-induced obese mice for 1 week or longer NOX-B11 decreases body weight and body fat [57]. Targeting ghrelin with a Spiegelmer may confer some advantages over receptor-targeting approaches: because the Spiegelmer does not cross the blood brain barrier, the risk of undesirable CNS (central nervous system) effects is greatly reduced [47,55].

Despite these compelling arguments for a causative role in obesity, the million dollar question of whether the interference with the ghrelin pathway is sufficient to effect a sustained down regulation of body weight remains. Conceptually, what has happened in humans is that the environmental factors, namely the access to a palatable, calorie-rich diet combined with lack of physical activity, are ultimately the trigger for the emergence of obesity as a widespread public health problem. In addition, some metabolic conditions promote weight gain and many subjects fail to lose



weight even when placed on very strict low calorie diets with daily physical activity routines [47].

With obesity becoming a world wide health threat, the pharmaceutical industry is eager to find the therapeutic means to treat this looming epidemic. Ghrelin entered the stage with a splash. Today, the existing body of data makes a compelling argument for ghrelin as a target in obesity. As the early drug candidates are becoming available, we will soon find out how effectively obesity can be treated through the antagonism of the ghrelin pathway. As triggering the need to eat is only one of the activities that ghrelin regulates in the human body, it will be interesting to see what side effects, if any, are evident in clinical studies where ghrelin activity is switched off. One would have to ask the question: instead of administering a pharmaceutical drug to antagonise ghrelin activity, could the natural appetite suppressant properties of CCK and ghrelin present in whey be incorporated into foods to ultimately control weight gain and / or promote weight loss?

Clinical applications of ghrelin in humans are already being tested. Ghrelin might be a useful tool for the diagnosis of growth hormone deficiency and treatment of chronic heart failure, cachexia and feeding disorders. In the future, it is possible that a ghrelin antagonist for the treatment of obesity may be developed. Further research on ghrelin will contribute to our understanding of physiological and pathophysiological feeding mechanisms and provide a novel therapeutic tool for patients with altered nutritional homeostasis.

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Appendix [58]

An investigation of the effect of consuming Carbery whey protein isolate and whey protein hydrolysate on a hormonal regulator of hunger and subjective feeling of appetite and fullness in humans.

In recent human trials, Carbery fed subjects drinks made from Isolac (WPI) and Optipep (WPH). Serum ghrelin (pg/ml) was measured and the results are presented in Figure 1. During the 3 hour postprandial period, the area under the curve (AUC) of serum ghrelin was 21% greater after ingestion of the Optipep.

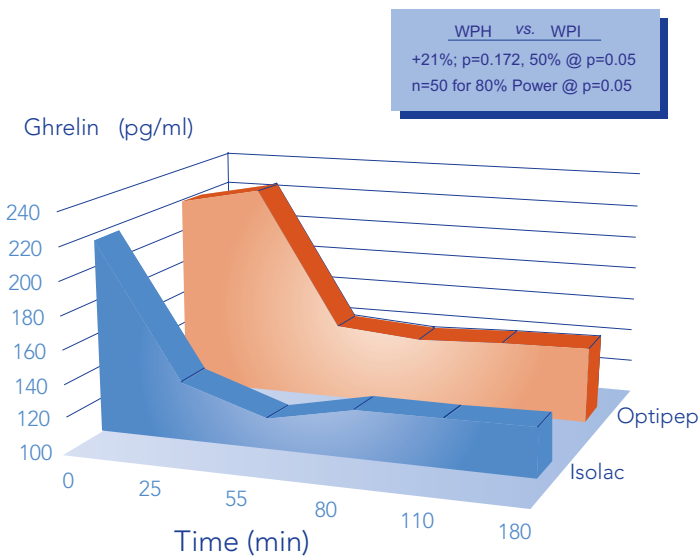


Figure 1 – Serum ghrelin concentration during the 3 hour postprandial period after ingestion of Isolac and Optipep.

Both protein sources resulted in a decrease in the ghrelin response as expected after the intake of a protein drink (ie ingestion of protein sources suppressed ghrelin levels in blood). The Isolac displayed a marginally faster decrease in serum ghrelin – 25 minutes vs 55 minutes. However, serum ghrelin remained suppressed for the entire 3 hours after ingestion of both Isolac and Optipep. This may imply that a person hoping to lose weight would feel satiated for a minimum of 3 hours following ingestion of the prescribed amount of Isolac or Optipep.

While there was large inter-subject variability in the serum ghrelin response, this preliminary data suggests that ingestion of both Isolac and Optipep may decrease serum ghrelin.

The results for these trials are not statistically significant and we have 50% power. To prove statistical significance, we would need to increase subject numbers in future investigations.

Subjective sensations of appetite and satiety (fullness) were rated by **Visual Analogue Scale** during the 3 hour postprandial period. Both whey proteins reduced subjects’ appetite, particularly in the first 2 hours after ingestion (Figure 2). Subjects had less of an appetite after ingestion of the Optipep, a decrease in subjective appetite rating of up to 18%.

Subjects reported feeling most full in the first hour after ingestion of both whey proteins (Figure 2). Over the entire 3 hour period subjects were fuller after ingestion of the Optipep, an increase in mean subjective fullness rating of up to 20%.

Taken together, these results suggest that Carbery Optipep can make you feel fuller and reduce your desire to eat.

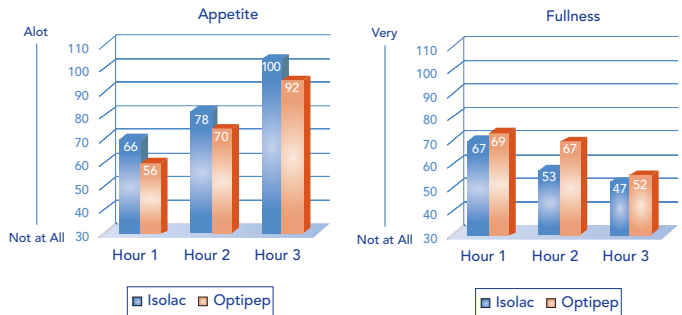


Figure 2 – Mean subjective appetite and fullness rating during the postprandial period for the Isolac and Optipep.

In conclusion, Carbery whey proteins can suppress one of the hormonal regulators of hunger (ghrelin) and can make you feel fuller and reduce your desire to eat. Potentially, when these ingredients are incorporated into food products such as protein beverages, bars etc., they could be used in low calorie, high protein diets. Not only would they serve as an excellent source of essential amino acids, but the properties reported above may make them fundamental to an effective weight management strategy.

Carbery whey proteins would increase satiety and feelings of fullness, which could potentially result in a reduced intake of food and subsequent reduction in body weight over a prolonged period of consumption.



For further information about Carbery's high quality whey proteins, please contact:
Carbery Food Ingredients, Ballineen, Co. Cork, Ireland
T: +353 (0)23 22200 F: +353 (0)23 47541 E: info@carbery.com or visit www.carbery.com