Whey Protein

Introduction

In recent years, milk constituents have become recognized as functional foods, suggesting their use has a direct and measurable effect on health outcomes.[1] Whey, a by-product of cheese and curd manufacturing, was once considered a waste product. The recognition of whey as a functional food with nutritional applications has elevated whey to a co-product in the manufacturing of cheese.[2] The two primary sources of protein in milk are the caseins and whey. After processing occurs, the caseins are the proteins responsible for making curds, while whey remains in an aqueous environment. The components of whey include beta-lactoglobulin, alpha-lactalbumin, bovine serum albumin, lactoferrin, immunoglobulins, lactoperoxidase enzymes, glycomacropeptides, lactose, and minerals.[2] Today, whey is a popular dietary protein supplement purported to provide antimicrobial activity, immune modulation, improved muscle strength and body composition, and prevention of cardiovascular disease and osteoporosis.

Whey Protein Constituents

Whey proteins contain all the essential amino acids in higher concentrations than vegetable protein sources.[2] The amino acids in whey are efficiently absorbed and utilized, relative to free amino acid solutions.[3] Whey proteins have a high concentration of branched-chain amino acids (BCAAs) – leucine, isoleucine, and valine – important factors in tissue growth and repair. Leucine is a key amino acid in protein metabolism.[4] Whey proteins are also rich in the sulfur-containing amino acids cysteine and methionine, which enhance immune function through intracellular conversion to glutathione. The primary whey components and their benefits are listed in Table 1.

Pharmacokinetics

Whey proteins do not coagulate under acidic conditions and withstand the action of chymosin in the stomach. They are considered to be "fast proteins," as they reach the jejunum quickly,[5] but after reaching the small intestine, hydrolysis is slower than that of casein, allowing for greater absorption over the length of the small intestine. Whey's rapid absorption patterns are superior for postprandial protein utilization and overall nitrogen balance in elderly women.[6] A randomized, single-blind study examining protein satiety found whey protein exhibits a higher postprandial level of plasma amino acids than casein.[7] A study by Troost et al revealed the lactoferrin component of whey survives stomach acid.[8]

Mechanisms of Action

Antioxidant Effects

Whey has potent antioxidant activity, likely by contributing cysteine-rich proteins that aid in the synthesis of glutathione (GSH), a potent intracellular antioxidant.[2] As an antioxidant, glutathione is most effective in its reduced form. As a result of whey's glutathione/antioxidant component, it is being investigated as an anti-aging agent.[9]

Detoxification

Practitioners use whey protein as a source of cysteine to increase intracellular glutathione levels.[10,11] Glutathione peroxidase (GSHPx) activity in cow's milk, and presumably whey, is the same as in human milk.[12] As a detoxifying agent, GSHPx, which is derived from selenium and cysteine, is an endogenous antioxidant enzyme that

Table 1. Primary Components of Whey Protein

Whey Component	% of Whey Protein	Benefits
beta-Lactoglobulin	50-55%	Excellent source of essential and branched chain amino acids – spares muscle and glycogen during exercise Binds fat soluble vitamins, increasing bioavailability
alpha-Lactalbumin	20-25%	Primary protein found in human breast milk Excellent source of essential and branched chain amino acids High in the essential amino acid tryptophan, which helps regulate sleep, mood, stress
Immunoglobulins	10-15%	IgA, IgD, IgE, IgG, IgM – primarily IgG Primary protein found in colostrum Immune enhancing benefits to all ages, particularly infants
Lactoferrin	1-2%	Antioxidant found in breast milk, tears, saliva, blood Antiviral, antibacterial, antifungal Promotes growth of beneficial bacteria Regulates iron absorption and bioavailability
Lactoperoxidase	0.5%	Inhibits growth of bacteria
Bovine Serum Albumin	5-10%	Large-sized protein with good profile of essential amino acids Fat-binding properties
Glycomacropeptide	10-15%	Does not contain amino acid phenylalanine, so is often used in infant formulas for infants with phenylketonuria Inhibits formation of dental plaque and cavities

converts lipid peroxides into less harmful hydroxy acids. In addition to the above-mentioned properties, the alpha-lactalbumin component of whey chelates heavy metals[13] and reduces oxidative stress because of its iron-chelating properties.[14]

Immune Enhancement

An *in vitro* study demonstrated bovine milk-derived IgG at levels as low as 0.3 mg/mL suppresses human lymphocyte proliferative response to T cells. The authors conclude IgG in bovine milk typically ranges between 0.6-0.9 mg/mL and is therefore likely to confer immunity that could be carried to humans.[15] Studies show raw milk from non-immunized cows contains specific antibodies to human rotavirus, *E. coli*, *Salmonella enteriditis*, *S. typhimurium*, and *Shigella flexneri*.[16,17] In a murine study, alpha-lactalbumin, in both the native and hydrolyzed state, enhanced antibody response to systematic antigen stimulation.[18] Alpha-lactalbumin also has a direct effect on B-lymphocyte function, as well as suppressing T cell-dependent and -independent responses.[19]

Antihypertensive/ Hypolipidemic Activity

Antihypertensive peptides have been isolated in bovine beta-lactoglobulin, suggesting whey reduces blood pressure.[20] These peptides provide whey with significant angiotensin I converting enzyme (ACE) inhibitory activity, which blocks the conversion of angiotensin I to angiotensin II, a highly potent vasoconstrictor molecule.[21]

beta-Lactoglobulin has been described by Nagaoka et al as a cholesterol-lowering agent. In animal studies, beta-lactoglobulin inhibited cholesterol absorption by changing micellar cholesterol solubility in the intestine.[22]

Clinical Indications

Exercise

Whey's amino acid profile makes it attractive for body composition and to support protein synthesis and muscle growth. In a double-blind trial, 42 men (ages 18-31) familiar with weight training followed the same weight resistance-training program for 12 weeks while consuming whey protein (1.2 g/kg body wt/day), a multi-ingredient whey protein sports supplement (1.3 g/kg body wt/day), or maltodextrin placebo (1.2 g/kg body wt/day). After 12 weeks, men who received whey protein in combination with resistance training showed greater improvements in at least one of four muscle-strength measurements and in lean tissue mass compared to the placebo group.[23]

Lands et al examined the effect of three-month whey protein supplementation (10 g twice daily) versus the same amount of casein as placebo on muscular performance in 18 men. When compared to baseline, peak power and 30-second work capacity improved significantly in the treatment group with no change in the placebo group. Body weight remained unchanged in both groups, but the supplemented group experienced a decrease in percent body fat. Enhanced biosynthesis of intracellular glutathione, shown by an increase of lymphocyte GSH levels in test subjects, is thought to be responsible for the improved muscular performance observed in this study.[24]

Another study examined the effects of casein and whey proteins on the acute response of muscle protein balance and synthesis in 25 young men and women randomly assigned to receive a 300-mL solution of whey protein (n=9), casein (n=7), or placebo (n=7) one hour after leg resistance exercise. Subjects in the whey and casein groups demonstrated a significantly increased uptake of phenylalanine after exercise (casein = 84±10 mg; whey = 62±18 mg; placebo = 5±15 mg) as well as stimulation of net muscle protein synthesis compared to placebo.[25] Cribb et al reported significant improvements in muscle strength and hypertrophy in 26 healthy male body builders supplemented with 1.5 g whey protein daily for 11 weeks; the control group was given an identical dose of a carbohydrate supplement.[26]

Two small studies on HIV-positive women examined exercise, whey supplementation, and body composition.[27,28] In the 2000 study the author notes an increase in body mass composition with whey supplementation; whereas, the 2001 study found no effect. Both studies note an improvement in quality of life with increased exercise and whey protein intake. Treatment groups received 1.0 g/kg daily of undenatured whey protein powder.

While moderate exercise enhances immunity,[29] intense athletic training has been shown to stress the immune system.[30-32] Free radical production and increased inflammatory activity are thought to contribute to impaired immune activity in over-trained athletes. In highly trained individuals muscular performance and recovery can be hindered by oxidative stress.[33-35] Lower levels of sIgA and glutamine[36,37] have also been observed after intense exercise and in over-trained individuals, and have been correlated with increased frequency of infection.[38] In addition, glutamine deficiency may contribute to gastrointestinal complaints experienced by highly trained individuals.[39] Whey protein is an excellent source of glutamine and together with its antioxidant and immune-enhancing properties suggests supplementation in this population may provide therapeutic benefit.

Pediatric Bowel Health

Creating a substitute for mother's milk has proven challenging. It is well accepted that nursing infants have a richer gut flora than bottle-fed infants, particularly with Bifidobacteria and Lactobacilli.[2] Such flora is normally associated with increased resistance to colonization of the digestive tract with pathogenic bacteria.[40] In a double-blind study, 102 healthy infants less than two weeks old were randomized to receive either a standard cow's milk formula or an infant formula containing partially hydrolyzed whey protein.[41] The whey-protein fed infants had significantly more Bifidobacteria in their stools, ultimately affording improved gastrointestinal immunity. It has been observed in previous studies that higher levels of Bifidobacteria in the digestive system decrease the potential for developing atopic conditions in at-risk infants with family history.[42]

A randomized, double-blind, placebo-controlled study of 43 infants with diagnosed infantile colic compared a hypoallergenic, hydrolyzed whey formula to a standard cow's milk formula for one week.[43] A clinically relevant result was observed in the whey formula group, with colic-related crying time reduced to less than one hour per day in the group taking whey formula – a one-hour greater reduction than the cow's milk formula group.

Thirty-eight infants with constipation (median age=1.7 months) were randomized to receive standard infant formula (n=18) or a partially hydrolyzed whey formula plus prebiotics (n=20) for three weeks, then crossed over to the other formula for an additional three weeks; only 24 subjects completed the crossover portion of the study. Infants on the hydrolyzed whey formula improved to softer stools at a higher rate (90 percent) than those consuming the standard infant formula (50 percent). Frequency of bowel movements improved significantly in both groups and similar growth rates were reported in both groups compared to baseline.[44]

A small, randomized, double-blind, crossover study examined the effect of hydrolyzed versus non-hydrolyzed whey protein on growth and development of 10 children with short bowel syndrome.[45] In children experiencing bowel resection, food introduction and promotion of normal growth and development is of utmost importance. Therefore, the increased transit time of whey proteins in the small intestine makes it an ideal protein source for this subset of children. Hydrolyzation of the whey protein did not have an effect on nitrogen balance, weight gain, or intestinal permeability compared to non-hydrolyzed whey, suggesting patients should utilize whichever whey protein is best tolerated.

Obesity

Research has shown a five-percent reduction in body fat mass can reduce the risk of obesity-related disease. Because high-protein, lower-carbohydrate diets appear to be the most effective for promoting weight loss, whey is an attractive source of dietary protein. The amino acids in whey act as substrates for protein synthesis and may improve body mass index in individuals participating in exercise and weight reduction programs.[23]

In a 12-week, randomized, double-blind study, a specialized whey protein high in leucine, bioactive peptides, and calcium was studied for its effect on fat loss and lean muscle in 106 obese women (ages 25-50) on a restricted 500 calorie/day diet. Subjects received a whey protein drink (n=53) or an isocaloric maltodextrin control beverage (n=53) twice daily, and body weight, body fat, and lean muscle mass were measured at 0, 4, 8, and 12 weeks. Responders were classified as subjects who lost at least 2.25 kg body weight. Blood samples were obtained and analyzed at the beginning and end of the study period and chemistry panels, lipid profiles, insulin, and complete blood counts were measured.[46] Significant weight loss was reported in both groups with no statistically significant difference at the end of 12 weeks. However, the whey protein group lost significantly more body fat than the control group (2.81 kg versus 1.62 kg in the completer group and 3.63 kg versus 2.11 kg in the responder group), and responder subjects in the whey-protein group lost significantly less lean muscle mass than those in the control group (1.07 kg versus 2.41 kg, respectively). Overall, subjects in the whey protein group lost 6.1 percent of their body fat mass. Blood profiles revealed a significant decrease in cholesterol, triglycerides, and LDL cholesterol in the treatment group compared to the control group.[46]

Diabetes

Whey protein reduces postprandial glycemia and promotes insulin release in healthy subjects.[47] In one trial, blood glucose and insulin response were measured in 14 type 2 diabetic men and women (ages 27-69) after consumption of a high-glycemic-index meal with or without whey protein supplementation. Insulin levels after breakfast were 31-percent higher when whey was added to the meal, an effect even more pronounced after lunch, as evidenced by a 57-percent increase in the insulin response and 21-percent reduction in blood glucose with whey compared to no whey. Glucose-dependent insulinotropic polypeptide responses were also higher after supplementation with whey protein. These results demonstrate that whey protein added to meals with a high glycemic index stimulates insulin release and reduces postprandial glucose levels in type 2 diabetic patients.[48]

Cardiovascular Disease

Whey protein may provide benefit for normalizing blood pressure and lipid levels. A study on a group of 20 healthy adult males investigated whether a fermented milk supplement containing *Lactobacillus casei* and *Streptococcus thermophilus* with added whey protein concentrate would affect serum lipids and blood pressure.[49] When volunteers consumed 200 mL of fermented milk with whey protein concentrate or placebo (non-fermented milk without added whey protein) twice daily for eight weeks, the whey-protein group had significantly higher HDL cholesterol and lower triglycerides and systolic blood pressure than the non-whey group. The difference in total- and LDL-cholesterol levels between groups was not statistically significant.

Human Immunodeficiency Virus (HIV)

Glutathione deficiency is common in individuals infected with HIV. A study by Micke et al demonstrated significantly elevated glutathione levels in 30 subjects with HIV supplemented with 45 g whey protein daily for two weeks.[50] The same researchers, in a subsequent six-month study, again found significantly increased glutathione levels compared to baseline.[51]

Cancer

Whey protein concentrates have been researched extensively with respect to cancer prevention and treatment. Glutathione stimulation is thought to be the primary immune-modulating mechanism. In a review of whey protein concentrates in the treatment of cancer, Bounous discusses the antitumor and anticarcinogenic potential. The amino acid precursors to glutathione in whey might increase glutathione concentration in relevant tissues, stimulate immunity, and detoxify potential carcinogens.[52] Other authors conclude the iron-binding capacity of whey might also contribute to anticancer potential, as iron may act as a mutagenic agent causing oxidative damage to tissues.[53]

Many animal studies have examined the effects of whey and its immune-enhancing components, including lactoferrin and beta-lactoglobulin.[54-56] In an animal model of colon cancer, animals given whey components demonstrated significantly lower incidence of tumors and fewer aberrant crypts.[55,57,58] A hamster study demonstrated fractionated whey has the ability to prevent and treat 5-fluorouracil chemotherapy-induced oral mucositis.[59] This protection is thought to occur via induction of tumor growth factor-beta (TGF-ß), which reduces basal epithelial cell proliferation. An *in vitro* study by Kent et al demonstrated that an isolate of whey protein, when compared to a casein-based protein, increased glutathione synthesis and protected human prostate cells against oxidant-induced cell death.[60]

Other Uses of Whey Protein

The wide range of essential and non-essential amino acids, minerals, fats, and biologically active proteins in whey provide extensive application in clinical nutrition. Adequate protein intake is essential for post-surgical wound healing, and protein depletion delays healing time.[61] Surgery stresses the body, altering natural defenses and leading to various post-surgical complications. Zimecki et al demonstrated the lactoferrin component of whey protein regulates the immune response and provides protective measures in post-surgical patients.[62]

Human studies demonstrate whey protein improves cognitive function and coping ability in highly stressed individuals.[63,64] Because a rise in serotonin is thought to improve adaptation to stress,[65] the authors propose tryptophan in whey provides a substrate to increase brain serotonin levels. The treatment groups for both studies received alpha-lactalbumin-enriched whey protein because it has the highest tryptophan concentration of whey protein fractions.

Side Effects and Toxicity

For individuals with frank milk allergies, whey products may not be suitable. On the other hand, many dairy-sensitive individuals find that casein is the culprit and they can tolerate whey, particularly if it is partially hydrolyzed and therefore less allergenic. Other dairy-sensitive individuals are lactose-intolerant. Most whey proteins are processed to remove lactose, with finished products only containing trace amounts. A challenge test with a small portion of a particular whey supplement for an individual with dairy sensitivities is indicated before beginning therapeutic amounts.

Dosage

Dosages of whey protein published in clinical trials range from 30-90 g daily, depending on the condition being treated.

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