



## Significance of Whey as a Biofunctional Asset in Dairy Industry

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DOI: <https://doi.org/10.55248/gengpi.4.1023.102902>

### ABSTRACT

In recent years, whey protein, a byproduct of cheese production, has garnered substantial attention owing to its wide-ranging biofunctional properties. This review seeks to offer an all-encompassing overview of whey protein, emphasizing its potential for promoting health and therapeutic applications. Whey protein stands out as a rich source of essential amino acids and bioactive peptides, including lactoferrin, immunoglobulins, and lactoperoxidase, which contribute to its antimicrobial, antioxidant, and immunomodulatory attributes. Furthermore, whey protein's ease of digestion makes it a valuable protein source suitable for individuals of all age groups. Whey, produced as a byproduct during cheese production, is a notable feature of India's dairy landscape, reflecting the country's extensive cheese manufacturing. The trend toward increased processing of whey into whey derivatives, such as whey protein concentrates (WPCs) and whey protein isolates (WPIs), has gained momentum. Whey ingredients are experiencing substantial growth in the global market, especially in the context of opportunities related to infant formulas, nutritional foods, and medical applications (IDF Bulletin, 2023).

Keywords: Whey protein, Biofunctional, health-promoting, therapeutic potential, whey derivatives, growing market

### 1. Introduction

Cow's milk stands as one of the most nutritious foods, widely consumed by humans. It is a bountiful source of nutrients with various biological properties that exert influence over biochemical processes within our bodies, support the development and functioning of specific organs, and confer protection against diseases. Milk offers a broad spectrum of biologically active components, including bioactive proteins and peptides, oligosaccharides, immunoglobulins, and fats/lipids, all of which contribute to safeguarding against pathogens and maintaining health when regularly incorporated into one's diet (Shayanti M and Sanjeev A, 2020).

"Whey is a residual product that arises during the cheese manufacturing process. The composition and attributes of whey are contingent on both the production techniques employed for the final product and the quality of the milk utilized. Liquid whey is primarily comprised of approximately 93% water and encompasses nearly 50% of the total solids found in milk, with lactose being the predominant component. While proteins constitute a minor fraction, making up less than 1% of the total solids, trace amounts of minerals and vitamins are also present."

The production of whey-based beverages commenced in the 1970s, and since then, a diverse array of whey beverages has been developed. These beverages can be crafted from various sources, including native sweet or acid whey, deproteinized whey, native whey diluted with water, whey powder, or through whey fermentation. Non-alcoholic whey beverages encompass a wide range of products created by blending native sweet, diluted, or acid whey with different additives. These additives may include tropical fruits, such as apples, pears, strawberries, or cranberries, grains and their derivatives, primarily bran, isolates of plant-based proteins, carbon dioxide (CO<sub>2</sub>), as well as chocolate, cocoa, vanilla extracts, and other flavoring agents.

This trend within the country stands as a major catalyst for the market's expansion, as whey protein is a prominent ingredient widely utilized in a variety of functional products, including healthy snacks and ready-to-eat meals. The whey protein market in India, in particular, experiences substantial growth driven by several key factors. These include the escalating demand for dairy-based ingredients, increasing consumer awareness of the benefits of a healthy diet, the rising popularity of health clubs and fitness centers, a growing percentage of elderly individuals throughout the nation, and the keen interest of the youth in sports and nutrition.

India, known as one of the world's fastest-developing nations, has witnessed a threefold growth in its food sector over the past decade, with expectations of continued growth over the next ten years. Functional foods and beverages have gained significant traction among health-conscious Indian consumers. The Indian whey protein market is anticipated to expand from USD 91.78 million in 2023 to USD 120.29 million by 2028, exhibiting a Compound Annual Growth Rate (CAGR) of 5.56% during the forecast period (2023-2028) (source: mordorintelligence.com).

Notably, there is a special focus on the development of whey beverage production through whey fermentation with probiotic bacteria. The crucial step in this process involves selecting a suitable bacteria culture to produce functional beverages with high nutritional value and favorable sensory attributes. Non-alcoholic whey beverages encompass dietetic beverages, drinks with hydrolyzed lactose, milk-like beverages, and powdered drinks. Meanwhile, alcoholic whey beverages include drinks with a modest alcohol content (up to 1.5%), whey beer, and whey wine. Whey beverages are suitable for a wide range of consumers, from children to the elderly, due to their high nutritional value and beneficial therapeutic qualities.

## 2. WHAT IS WHEY?

Whey is the yellowish-green liquid that separates from curdled milk during the cheese-making process. It is also referred to as lactose serum, as it contains approximately 70% lactose. Liquid whey boasts a mildly tart yet pleasant flavor profile. Sweet whey is typically produced during the manufacturing of hard cheeses like cheddar and Swiss cheese, whereas acid whey is derived from the preparation of soft cheeses, such as cottage cheese.

For many decades, whey was regarded as a significant dairy waste due to challenges associated with its disposal, primarily stemming from its high biological oxygen demand and substantial organic content (Ahn, W.S et al., 2001). However, in contemporary times, whey proteins have been acknowledged as a valuable source of nutrients and are harnessed for their bioactive components. Owing to its rich nutritional profile, whey is now extensively utilized in various commercial food products and plays a pivotal role in the dairy industry.

In its typical form, fresh liquid whey derived from cheese-making comprises approximately 94.2% water and contributes to 50% of the total solids. Within this composition, you'll find about 0.8% whey proteins, 0.5% minerals, 0.1% fat, and 4.3% lactose, which serves as the primary constituent (Almeida, G. et al., 2013). Table 1 provides a comprehensive breakdown of the general composition of sweet whey and acid whey.

**Table 1. Composition (%) of sweet and acid whey**

Component	Sweet whey	(%w/w)	Acid whey	(%w/w)
Water	93-94		94-95	
Dry Matter	6-6.5		5-6	
Lactose	4-5.5		3.8-4.3	
Lactic Acid	Traces		≤ 0.8	
Total Protein	0.8 – 1		0.8 – 1	
Whey Protein	0.6 – 0.65		0.6 – 0.65	
Citric Acid	0.1		0.1	
Minerals	0.5-0.7		0.5-0.7	

(Source: Judy A. Driskell. 2007. Sports Nutrition: Fats and Proteins. New York: CRC, p. 155.)

Whey proteins are a type of globular proteins characterized by a substantial presence of  $\alpha$ -helix patterns, with hydrophilic and hydrophobic, as well as acidic and basic amino acids, evenly distributed along their polypeptide chains (Evans, E.W., 1982). The primary components of whey proteins encompass  $\alpha$ -lactalbumin ( $\alpha$ -LA),  $\beta$ -lactoglobulin ( $\beta$ -LG), bovine serum albumin (BSA), immunoglobulins (IG), bovine lactoferrin (BLF), bovine lactoperoxidase (LP), and smaller quantities of glycomacropeptide (GMP). The composition of each constituent can be found in Table 2. However, it's important to note that the composition of whey proteins can vary depending on various factors, including the type of whey (sweet or acid), the source of milk (bovine, ovine, or caprine), the cattle's diet, lactation stage, and the specific processing method. Whey, which tends to be acidic in nature, typically has a pH of around 5.1 and is usually produced through direct acidification. In contrast, sweet whey has a slightly higher pH of approximately 5.6 and is typically generated through rennet coagulation, notably during the cheese-making process (Pintado, M.E et al., 2001)

**Table 2. Whey protein constituents and its composition**

Whey Protein Constituent	Concentration (g/L)	Molecular Weight in kDa	Number of Amino Acid Residues
Lactalbumin	1.2	14,175	123
Lactoglobulin	1.3	18,277	162
Bovine serum albumin	0.4	66,267	582
Immunoglobulins (A, M, and C)	0.7	25,000 (light chain) and 50,000–70,000 (heavy chain)	-
Bovine lactoferrin	0.1	80,000	700
Glycomacropeptide	1.2	6700	64
bovine Lactoperoxidase	0.03	70,000	612

(Source: Shayanti M and Sanjeev A, 2020)

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### 3. Benefits of Whey

Whey protein is widely embraced as a dietary supplement, finding popularity among a diverse range of individuals, including bodybuilders, athletes, and those seeking an extra protein source in their nutrition (source: Healthline). Whey is particularly rich in leucine, a branched-chain amino acid known for its remarkable growth-promoting, or anabolic, properties. This is why whey protein is recognized for its effectiveness in countering age-related muscle loss and enhancing overall strength. Muscles, which are predominantly composed of proteins, undergo structural changes in response to exercise. These changes encompass increased size, strength, speed, and resistance to fatigue. The supplementation of one's diet with whey protein accelerates and enhances these beneficial muscle adaptations. Additionally, it contributes to improved heart and immune health, as highlighted by research (Kinsella and Whitehead, 1989)

As the global trend towards healthier eating continues to gain momentum, there is a growing worldwide demand for food products that are rich in protein. For a sedentary individual, the recommended daily average protein intake stands at 0.8 grams per kilogram of body weight per day (g/kg/day). This protein quantity is necessary to maintain a positive nitrogen balance and support healthy metabolic functions in the body. A variety of supplemental protein sources are available, including options such as egg, soy, hemp, whey, and casein. Among these choices, milk whey stands out due to its high concentration of readily available amino acids that are easily digestible. This feature makes whey protein a highly effective choice for incorporation into the body's cells (source: Shayanti M and Sanjeev A, 2020)

Whey proteins offer advantages not only to athletes but also to the general population. They prove to be beneficial for physically active individuals in terms of repairing and rejuvenating muscle fibers, helping to maintain a healthy body weight, and contributing to overall heart and immune system well-being. Adequate consumption of whey protein is known to play a role in maintaining body weight by helping to balance blood sugar levels. Normal blood sugar levels are associated with increased fat loss, higher energy levels, and a reduced tendency to overeat.

Scientific research has also highlighted several other health benefits of whey protein. It has been shown to promote heart health, aid in cancer prevention, and improve the immune system, particularly in individuals with AIDS (source: Kinsella and Whitehead, 1989). Furthermore, incorporating whey proteins into the diet has been found to reduce cholesterol levels and support the maintenance of strong bones and muscles during the aging process. In fact, a study demonstrated that older men who regularly consumed whey protein experienced an increase in protein synthesis, which helps mitigate muscle loss over time. Additionally, the same study revealed that elderly individuals with lower protein intake were more prone to experiencing a loss of bone density, especially in the hip and spine (source: Kinsella and Whitehead, 1989).

Research also suggests that whey protein may contribute to reducing the rate of bone density loss. In a study conducted in 2000 at the University of Arkansas by Hakkak and colleagues, diets rich in both whey and soy were found to protect against mammary tumors in rats. According to the study, soy protein in the diet reduced tumor incidence by 20%, while whey proteins were twice as effective as soy in this regard (source: Hakkak et al., 2000). In summary, whey protein consumption offers benefits regardless of age and fitness goals

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### 4. Whey Utilization in Food Products

The Indian whey protein market can be categorized based on product type and application. In terms of product type, it is segmented into whey protein concentrate, whey protein isolate, and hydrolyzed whey protein. When considering its application, the market is further divided into sports and performance nutrition, infant formula, and functional or fortified food (source: mordorintelligence.com)

Over the last few decades, alternative methods for utilizing whey have emerged. Due to the challenges of high transportation costs and the susceptibility of fresh pasteurized liquid whey to deterioration during storage, it is infrequently used in its original form for food products. Instead, various processes such as evaporation, reverse osmosis, or ultrafiltration are employed to concentrate whey into condensed products, or it is maximally concentrated through drying. Derived from whole whey, different valuable products have been created, including whey powder, whey protein concentrates, whey protein isolates, reduced-lactose whey, and demineralized whey (source: Akshay Aswani, 2010)

In the realm of food applications, whey proteins and their derivatives have garnered substantial attention due to the multitude of advantages they offer, owing to their diverse functionalities. These functionalities include gelation, foaming, emulsification, solubility, and thermal properties. Incorporating whey proteins into food products is known to enhance their sensory quality and texture. For instance, whey proteins have been successfully added to a wide range of foods, including yogurt, bakery products, energy bars, pasta, and beverages, where they positively influence overall quality and nutritional value. An illustrative study reported on the efficacy of incorporating a complex of non-heat-treated whey protein and high methoxyl pectin in low-fat yogurt, with the whey protein serving as a commendable fat-replacer and texturing agent for the yogurt (source: Silviya R. Macwan et al., 2016).

Whey proteins are incredibly versatile and can benefit an array of food products, including bread, processed meats, dried soups, surimi, whipped toppings, and confectionery. Whey protein concentrates, for instance, are used to create gels suitable for both meats and sweet treats like custards, puddings, and confectionery. Certain formulations possess excellent whipping and foaming properties, making them invaluable in applications like whipped toppings. Whey proteins also find utility in stabilizing various food emulsions, from salad dressings to soups and processed meats. In today's baking industry, whey proteins serve as nutritious moisture binders, enhancing the quality of baked goods.

It's worth noting that the main protein found in whey, beta-lactoglobulin, has a pronounced ability to bind flavors. This feature has led researchers at Cornell University to suggest that food processors may need to adjust flavor levels when working with ingredients high in beta-lactoglobulin (source:

Kinsella and Whitehead, 1989). Moreover, whey, as a whole, serves as a valuable component in the creation of functional foods, with alpha-lactalbumin and beta-lactoglobulin being extracted and utilized as functional food ingredients (source: Akshay Aswani, 2010)."

In general, whey from various sources can be employed, including sweet whey, sour whey, or powdered whey. However, the utilization of powdered whey tends to raise the overall cost of the final product unnecessarily. Whey, along with whey derivatives sourced from cheese, casein, paneer, and more, can be effectively used in beverage preparation. In the food industry, whey can be harnessed in various ways, but it is predominantly dehydrated into whey powder or used in the production of whey protein concentrates and lactose or protein isolates. Whey beverages are recognized as excellent thirst-quenchers. Unlike many soft drinks, whey-based beverages are light and refreshing, with lower acidity compared to fruit juices. They are often considered a healthier option for quenching one's thirst (source: Prendergast, 1985)

Top of Form

#### 4.1 Whey beverages

Ready-to-drink (RTD) beverages, which are marketed to reach a broad consumer base, often include some form of protein fortification. In this regard, whey proteins are a commonly preferred ingredient. High-protein beverages, typically containing more than 4% protein content, are typically consumed for the replenishment of amino acids and muscle-building, particularly during and after exercise. On the other hand, low-protein drinks, typically containing less than 4% protein, are tailored for young children as a health-conscious alternative to carbonated sugary beverages (source: Wong and Watson, 1995).

##### a. Alcoholic Beverages

Due to its high lactose content, which makes up approximately 70% of its dry matter, whey is an ideal raw material for the production of alcoholic beverages. Alcoholic whey beverages are categorized into those with low alcohol content, typically equal to or less than 1.5%, including whey beer and whey wine (source: Silviya R. Macwan et al., 2016). The production of whey beverages with low alcohol content typically involves the process of deproteinizing whey, concentrating the whey, fermenting the lactose (usually using yeast strains like *Kluyveromyces fragilis* and *Saccharomyces lactis*), or adding sucrose until the desired alcohol content of 0.5% to 1% is achieved. Flavoring, sweetening, and bottling are then carried out (source: Sienkiewicz & Riedel, 1990).

Furthermore, Patel (2012) introduced the concept of self-carbonated probiotic whey beverage using chhana whey. This unique beverage is prepared by inoculating cultures of *Kluyveromyces marxianus* and *Lactobacillus helveticus* MTCC 5462, along with the addition of 7% sugar. It is then incubated at 25°C until the acidity reaches 0.5% Lactic Acid (L.A.), followed by storage at refrigeration temperature for self-carbonation.

##### b. Non-alcoholic Beverages / Lactic fermented beverage

These beverages are created through the fermentation of lactose in whey by lactic acid bacteria (LAB) and can be primarily categorized into three groups: whey-based fermented drinks, whey-based fermented carbonated drinks, and whey-based cultured dairy products. Worldwide, various fermented and carbonated whey beverages are produced, and they can be grouped based on their primary substrate and the types of cultures used (source: coursesonline.iasri.res.in).

The current global popularity of fruit-flavored drinkable yogurt presents an excellent opportunity for incorporating whey into these products. The compatibility of whey, especially acid whey and/or further fermented sweet whey, with the characteristics of cultured dairy beverages makes this approach particularly appealing for traditional dairy processors. Apart from whey-based yogurt drinks, such as 'yor,' and 'thin buttermilk' products like 'interlac,' which are prepared by culturing a blend of regular milk and an ultrafiltration whey protein retentate with the same protein content as milk, are additional examples of whey-containing cultured dairy beverages. Table 3 highlights some well-known whey-based fermented beverages from around the world.

Table 3. Characteristics of various whey beverage in world.

Product	Type	Characteristics
Rivella	Whey based fermented carbonated drink	35% deproteinated whey serum+water, sugar and Flavour
Surelli	Whey based fermented carbonated drink	Almost same as rivella
Fauna-fit	Soft drink type	Approx. 85% sweet whey UF permeate fermented & after second UF mixed with fruit juice (mango, pineapple, strawberry)
Whey kwas	Alcoholic kefir like drink	Deproteinated whey inoculated with thermophillic starter and then treated with yeast
Servovit	Whey based cultured dairy products	Carbonated product based on whey and cultured buttermilk
Kumiss like beverage	Whey based fermented drink	Whey + buttermilk inoculated with culture comprised of kumiss yeast <i>L. bulgaricus</i> and <i>L. acidophilus</i>
Milone	Whey based fermented drink (mildly alcoholic and sweet sour in flavour)	Whey +1% lactic acid + equal amount of tea, coppt", remaining liquid is then incubated with lactose fermenting yeast (0.8% ethanol)

<b>Alcoholic beer</b>	<b>whey</b>	Beer like beverage	Whey beer brewed with hops but without added malt
<b>Malted beer</b>	<b>whey</b>	Beer like beverage	Malt and hops both were added to whey and fermented with bottom yeast
<b>Whey malt Beer</b>		Beer like beverage	50% whey + malt, hop, sugar and coloring matter (produced with top fermentation)
<b>Whey beer</b>	<b>nutrient</b>	-----	Whey+hops and nutrient salts (low alcoholic product)
<b>Whey champagne</b>		Alcoholic wine like	Deproteinized whey inoculated with 0.1% fresh baker's yeast + addition of coloring and flavouring substance
<b>Whevit</b>		Alcoholic soft drink	Whey is fermented with yeast culture ( <i>Saccharomyces cerevisiae</i> ), can be carbonated and non carbonated
<b>Acidowhey</b>		Non carbonated soft drink type	Whey fermented with lactic acid bacteria and free from preservative and synthetic color

## 5. Bioactive properties of whey protein

In 2005, researchers at Lund University in Sweden made a significant discovery regarding whey's potential to stimulate insulin release, particularly in individuals with type-II diabetes. Whey's biological components, including lactoferrin,  $\beta$ -lactoglobulin,  $\alpha$ -lactalbumin, glycomacropeptide, and immunoglobulins, exhibit a wide range of immune-enhancing properties. Whey is known to possess antioxidant, antihypertensive, antitumor, antiviral, and antibacterial qualities. One of the mechanisms through which whey exerts these effects is the intracellular conversion of the amino acid cysteine into glutathione, a potent intracellular antioxidant. Glutathione serves as a guardian against free radical damage and certain toxins.

Numerous clinical trials have successfully employed whey in the treatment of various conditions, including cancer, HIV, hepatitis B, cardiovascular disease, and osteoporosis, as well as its use as an antimicrobial agent. Whey is an abundant source of branched-chain amino acids (BCAAs) that support muscle function and stimulate protein synthesis. Leucine, in particular, plays a pivotal role in initiating the transcription of protein synthesis, potentially expediting recovery and alleviating stress. Whey protein also acts as a precursor to the antioxidant glutathione, providing protection against the adverse effects of stress factors.

Furthermore, bioactive peptides derived from whey protein concentrates, isolates, and hydrolysates with angiotensin-converting enzyme (ACE) inhibitory or antihypertensive activity are closely linked to the renin-angiotensin system. This suggests that foods containing antihypertensive peptides should be consumed regularly to help regulate blood pressure and reduce the risk of cardiovascular disorders (source: Shayanti M and Sanjeev A, 2020).

The presence of cysteine has been observed to play a crucial role in managing glycemia and controlling inflammation in individuals with diabetes. Several studies have indicated that the consumption of whey protein offers beneficial effects for cancer patients (source: Shayanti M and Sanjeev A, 2020). Reports suggest that whey protein hydrolysates may provide an enhanced anticancer effect compared to other forms of whey protein. Additionally, whey protein derivatives in the form of concentrates are recognized for their ability to enhance innate mucosal immunity and provide protection against immune disorders (source: Shayanti M and Sanjeev A, 2020).

Regular consumption of whey protein supplements containing amino acids like hydrolysates has been found to be effective in repairing muscle damage. Furthermore, the intake of  $\beta$ -hydroxy- $\beta$ -methyl butyrate, a compound derived from leucine, has been demonstrated to enhance muscle recovery.

## 6. Conclusion

Whey protein products are currently the focus of research for developing new pharmaceuticals and functional food ingredients that support gut health and regulate the absorption of nutrients due to their biofunctional properties. Many bioactive peptides isolated and purified from whey proteins demonstrate strong antioxidant, antihypertensive, anticancer, antidiabetic, and hypocholesterolemic properties. Once absorbed, these peptides target specific organs to exert their beneficial effects. The growth of the beverage market is driven, in part, by the convenience and effectiveness of beverages. In order to satisfy modern consumers and remain competitive in the beverage industry, beverages must be cost-effective, convenient, nutritious, and enjoyable. Product developers seeking to capitalize on the substantial growth in the beverage market can explore the use of whey and whey-based beverages for various applications and functionalities.

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