UTILIZATION OF WHEY & WHEY PROTEINS: EITHER TO REUSE OR TO REDUCE COST OF CHEESE MAKING PROCESS BY USING ADVANCE TECHNIQUES

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ABSTRACT

Whey is still considered as a dairy waste by dairy industries, and therefore, does not have any value. At the start of 21st century, regulations were developed for the disposal of why waste. Recently, scientific studies unrevealed the secrets whey protein and develop a number of techniques and methods to get whey protein with best nutritional and functional value. Advance technologies such as ultrafiltration, bio fermentation, transformation, have increased the again use of whey waste. Cost effectiveness is only the reason of not processing the whey protein and therefore, removes the whey protein as a waste by dairy industries. This paper discusses the production, types, forms, applications, separation of whey protein.

Keywords: Whey, Whey Protein, Ultrafiltration, Bio Fermentation, Transformation.

INTRODUCTION & BACKGROUND

Proteins of milk may be distributed into two groups; i) casein, ii) whey protein. [41] Caseins are present (approximately 80%) in milk in the form of spherical complexes with a radius of about 100 nm that are called casein micelles. During the manufacturing of cheese, the casein micelles get agglomerated. The liquid that is exuded during this process is called whey (approximately 20%) and comprises of numerous types of globular proteins that are, different from caseins, stiff, compact and complex structured. [46, 49] Whey was discovered 3000 years ago, when calves' stomach was used to carry, handle or transport milk. During the storage or transportation of milk, some chemical reactions took place in milk because of an enzyme "chymosin" (rennet), which is present in the calves' stomach. This was actually the start of foundation of cheese (or whey) industry. [19]

Composition

Whey proteins are byproducts of casein or cheese manufacturing industry and they contain proteins of high nutritional value as well, therefore, their use for the development of new varieties of food material and pharmaceuticals has been increased. [12] It is mainly a mixture of β-lactoglobulin, α-lactalbumin, bovine serum albumin (BSA) and immunoglobulins, with the isoelectric point being at pH ~5. Whey consists of a number of chief nutrients such as lactose (4.5 to 5% w/v), soluble proteins (0.6 to 0.8% w/v), lipids (0.4 to 0.5% w/v) and mineral salts (8 to 10% of dried extract). [23] The chemical composition of whey proteins is different for different type of cheeses and especially for different kinds of milk used for cheese production. [27, 50]

Forms of WP

Whey contains only small and diluted amounts of proteins but several industrial processes can be used to obtain different fractions from it, as whey protein concentrate (WPC, having protein contents 35-85%), whey protein isolate (WPI, protein content >90%) and enzymatic whey protein hydrolysate. [6] Whey protein isolate, concentrate and hydrolysate results from the separation of different fractions from whey and during the manufacturing of cheese. [44]

Whey Protein Concentrate (WPC)

This form is further available in different concentrations of protein as; with 80% protein (WPC80), 50% protein (WPC50), and 34% protein (WPC34). Although WPC contains low level of fat and cholesterol, but has high level of many bioactive components as well as carbohydrate (lactose). On the other hand, abovementioned 70-80% of protein could also be more available in the form of supplements of protein powder. [17, 43]

Whey Protein Isolate (WPI)

It is the purest form of whey protein and it contains 90% or more of the protein contents, but less lactose (<1%). Fat and cholesterol are present in minute quantities. These isolates are processed to remove fat, cholesterol and lactose, but these contain fewer amounts of bioactive compounds. [43]

Whey Protein Hydrolysate (WPH)

Hydrolysate whey proteins are more easily absorbed because these are predigested and partially hydrolyzed to their simpler peptide chains. WP hydrolysates are costlier but less allergic than other forms of whey. The purpose of this article is to enlighten the importance of the whey protein that was actually considered as a dairy waste from a long time ago. The present articles also have some of the techniques and methods that can be used to increase the nutritional as well as functional properties of whey protein. Overall, this paper will have proved as an attempt to increase the recovery and purification of valuable components from WP, which will result in a reduction of polluting load (polluting factors are as, it has 30 to 50 gL^{-1} BOD and 60 to 80 gL^{-1} COD) on the outside environment due to its disposal. [12]

Production

The production of whey increasing with an increase in the rate of milk production (>2% per year, FAO 2006). Whey is produced in significant amounts, approx. 10 L kg^{-1} of cheese produced. [31, 45, 57] There is a high volume of whey is produced in the world i.e. 10^8tons $year^{-1}$. [30] The major producers of whey are EU and the U.S. [21, 51] G.W. Smither (2008) studied that 50% of milk solids appears in the whey, in which present 100% of lactose and some of the 20% of proteins.

Benefits of WP

Benefits of consuming whey protein focus on its natural origin being recognized as a harmless foodstuff with higher nutritious value, biodegradability and technical functional properties. [52] Whey resulting from cheese making is available in large volumes (approximately 80-90% of the volume of milk used) and its disposal represents a major industrial concern. Nonetheless, whey is a source of valuable nutrients which could be recovered and used individually. [14]

Advance Techniques

The most commonly used technique in the dairy industry is membrane filtration for the removal and separation of different fractions of dairy waste. Ultrafiltration is also being used to effectively recover protein contents of waste before their disposal to outside environment.[5] Microfiltration is also used as a pretreatment, just before the ultrafiltration, to make the waste free from all the suspended or colloidal particles, fat, and casein particles, so as to minimize the fouling effect on the ultrafiltration membrane.[10] For the removal of charged or uncharged ions particle from the waste and to concentrate the valuable components of the whey, use of Nano-filtration technique is common in some of the dairy industries. Ultrafiltration in a diafiltration mode using water as a buffer is a useful technique that helps to get the maximum concentration of solutes as of lactose concentration, protein concentration, and other biomolecules. [13, 14] For lactose separation, techniques such as enhanced crystallization, isolation and purification, hydrolysis and transformation.

UTILIZATION OF WHEY PROTEIN
Edible Films from Whey Protein

Different edible films and coatings of different materials are used such as proteins, polysaccharides, lipids or combinations of these components. Among them, protein-based materials seem to be most attractive because they also provide nutritional value. [11] Whey proteins are generally used in infant formulas and sports food, and additionally exhibit good film- forming capacity. [24] Whey protein films are characterized by good mechanical resistance and constitute a good gas barrier at low relative humidity [39] as well as WPI have good aroma and oxygen barrier properties. WPI, depending on manufacturing process, contains approx. 30% α-lactalbumin, 60% β-lactoglobulin and 7% other whey proteins. Besides, protein-films have brittleness, so therefore, a small amount of glycerol (a plasticizer) can be used to reduce brittleness and increase strength. Some recent studies show that the barrier properties of protein-film can be increased by different concentrations of glycerol, as Kokoszka, Debeaufort, Lenart & Voilley (2010) observed that these film properties were better at 40% (w/w, of WPI) of glycerol. Whey proteins have amphiphilic character. [49] The functional properties of whey protein can be change with the help of addition of certain components as polysaccharide. Whey protein edible film could also be modified by using almond and walnut oil in different concentrations to improve the functional properties. [24]

Antioxidants from Whey Protein Hydrolysate (WPH)

The scientists from different fields including food science were attracted towards the ability of protein to get assembled itself and to form a fibrillary system.[1] These structures of proteins have many functional properties such as gelation property, emulsification property, foaming property, viscosity increasing property, etc. [38]The most commonly studied protein was whey protein, because it has high nutritional value, not expensive, different functional characteristics.[42] Whey proteins are a good source of peptides that promotes the good health of humans.[42] Peptides derived from whey proteins have antihypertensive, antithrombotic, immunomodulatory, antimicrobial and antioxidant properties.[41]Enzymatic hydrolysis method is most commonly used, among all the other methods, to get antioxidant peptides from the whey protein.[37] The exposure of amino acids that were buried in the native conformation of protein, increasing of hydrogen ions availability and concentration of carboxylic acid groups due to enzymatic hydrolysis also contributes in the augmented antioxidant activity of protein hydrolysates.

Delivery of Nicotinic Acid by Using WPI

Nicotinic acid, or pyridine-3-carboxylic acid, is also known as niacin or vitamin B_3 and produces aqueous alkaline solutions, which are highly stable to oxygen, heat and light. The most important and major benefits of this vitamin include cell growth and metabolic regulation of humans. Insufficient intake of niacin (or nicotinic acid) results in the severe photosensitive dermatitis, which is also known as pellagra.[9] Due to limited consumption of essential nutrients by almost a complete population, the process of fortification of processed foods just remains a subject of interest.[26] Biopolymer based materials have been employed, using a variety of protocols, to deliver water or fat-soluble vitamin fortification.[12] Advantages of using whey protein include the ability to control the release rate of small molecules with pH variation via the carboxylic and ammonium groups in polypeptide chains, which adjust their protons to acidic or neutral medium.[32]

Reuse of WP into Cheese

The reuse of WP into cheese matrix has been an issue of utmost importance for the increase of yield, the improvement of nutritional value, the refinement of texture and the increase of profit. Native WP can be reuse in cheese, although heat-denatured WP (HDWP) are preferred for their high water-binding capacity and better physical retention in the curd during drainage. Therefore, heat-induced association of WP and casein is exploited to increase the cheese yield. Anyway, the adverse effect of in situ denaturation of the WP means the progressive worsening of the rennet coagulation properties due to the interaction of WP with k-CN, impairing its accessibility with the coagulant. Another approach is represented by ultrafiltration of unheated milk and subsequent coagulation by acidification or renneting of the retentate enriched in native WP. Combination of heating and membrane technologies to entrap HDWP in the cheese matrix is the solution which is preferentially adopted for soft cheeses. The inclusion of protein-based powders into the cheese milk is obtained by the addition of milk protein concentrate (MPC) and whey protein concentrate (WPC). Moreover, a microparticulated whey protein concentrate (MWPC) can be used in cheese making mainly as a fat replacer.

Particulation is a technology by which heated WPC is submitted to a simultaneous shearing force allowing to aggregate WP into particles the size of which depends on composition and process conditions. The size of these last aggregates is up to the mm scale and can be reduced by shearing of the heated whey up to 0.5-10 mm. HDWP can be formed also upon heat treatment of the cheese milk. This process promotes unfolding of WP and subsequently, via disulphide-thiol exchanges and hydrophobic bonds (Kelly et al., 2008), the formation of WP-coated casein micelles and/or WP aggregates. Milk pH during heat treatment plays an important role in the distribution of these two types of interactions. The diameters of complexes between CN and WP is from 30–100 nm up to <1 mm. Finally, the increase in WP content into cheese can be achieved by addition of Ricotta cheese into cheese milk. The large flocks of WP in Ricotta remain entrapped into the coagulum and create a soft texture with an increased capacity to absorb water into the cheese.

Bromine and Iodine from WP

S.V. da Silva *et al.*, 2016 studied that the whey protein contains appreciable amounts of bromine and iodine. Iodine is an essential element for growth, development, and well-being, as it is an important component of the thyroid hormones, but adverse reactions have been associated with excessive intake.[23] Bromine is not an essential element and has been used in pharmaceuticals, disinfection products, and many polymers as a flame retardant, and is associated with endocrine disruption and subject to bioaccumulation in food chains.[54] Therefore, the determination of both Br and I is important to assure the quality of food and the ingestion of safe levels of these elements. Studies showed that the casein contains not much amount of iodine as compared to whey protein, therefore, casein can be enriched with iodine obtained from the whey protein. As the Estimated Average Requirement (EAR) from iodine were estimated to be 95μg.d^{-1} from diet and the Recommended Dietary Allowances (RDA) is 150μg.d^{-1}.[34] For casein, the intake of 100 g only reaches 37.1 μg.d^{-1}, while for whey proteins the intake ranges from 253 to 804 μg.d^{-1}.[49] Bromine is not an essential trace element and scientific literature has not established its requirements.[54] From studies with human volunteers, a non-observed adverse effect level value of 4 mg kg^{-1} of body weight was identified. Based upon this value, a provisional acceptable daily intake (ADI) for Br of 0.4 mg kg^{-1} of body weight could be derived. [53] Therefore, the concentration of Br in whey proteins and casein is far from ADI (e.g., 28 mg for a 70 kg man) ranging from 0.145 to 9.04 mg per 100 g of protein.

Used as a Substrate

WP protein can also be used as a substrate for growth of microorganisms. WP is also a good source of carbon and expect these, it also has such properties that microorganisms, especially algae (microalgae), can grow on this. A.P. Abreu et al. (2012) researched that a green algae *Chlorella vulgaris*were cultivated on a whey protein substrate and produce appreciable amount of biomass at low cost. Microalgae can be used for animal feed, as well as for high-valued health supplements. [15] Microalgae is also being used for the treatment of waste water, to lessen the CO_2 gas danger. [8]

Biotechnological transformation to Bioethanol

As WP contains an appreciable amount of lactose, therefore, it can be used for the production of bioethanol together with some amount of scotta (a dairy waste too). [22] To date, fermentation of WP is hardly economically competitive in comparison with the fermentation of other sources as sugar cane and so on. A yeast strain, named as *Kluyveromyces marxianus*, was used for the production of ethanol from whey protein at a temperature above 40^{o}C. [29, 3]

CONCLUSION

Whey protein (WP) contains appreciable amounts of proteins (WPI, WPH & WPC). WP can be used again for the production of cheese, as it contains protein that is available at low cost. It can be used as a carrier of certain vitamins and supplements into the body through capsule. Whey protein is also used as a substrate by certain microorganisms for the production of ranges of products i.e. biomass, enzymes, glucose, galactose, etc. But the most important and major use of WP is as edible films and coatings.

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