ENCAPSULATION OF NEUTRACEUTICALS IN DAIRY PRODUCTS

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Abstract - Due to the awareness of health benefits of probiotics, there has been an increased use in different health based products. Probiotics have been incorporated into a wider range of dairy products, including yogurts, cheese, ice cream and dairy desserts. The ability of probiotic cells is of greater importance because of having beneficial effects on health, they must stay alive until they reach their site of action. There are some problems regarding survivability of probiotic bacteria in dairy foods due to various factors. This encouraged developing of different innovative ways to improve the probiotic cells viability in the product incorporation. Microencapsulation of probiotics is one of the best approaches which currently is receiving considerable attention. Microencapsulation of probiotic bacteria can be applied to enhance and also to improve the viability during processing and also in gastrointestinal tract.

Keywords: Encapsulation, dairy products, iron, vitamins, chitosan.

I. INTRODUCTION

Milk and dairy products comprised an important part of the human diet all over the world since ancient times. Due to the presence of almost all essential nutrients, milk is popularly called as a balanced food for all groups of age. Recent research mentions that apart from delivering basic nutrients, milk is a well known source of bioactive components. A number of dairy products have already found their ways in several food items for nutritional improvement. Milk is enriched with various nutritional elements, making it one of the important and complete food in nature, however, recently some bioactive components and other few beneficial health components have been incorporated into dairy items. In this modern period, people expect their food, besides providing all basic nutrients, should nourish their health, also ensuring protection against certain diseases. As a consequence, dairy companies are continuously launching new improved products supplemented with specific bioactive components. A large number of research studies are being conducted to develop adoptable dairy products aimed at particular groups who are deficient in specific nutrients as well as at the general population. In addition, there has been a significant research that are helping the large numbers of lactose-intolerant people all over the world. In this regard, microencapsulation have evolved as advanced technology with huge potential. Microencapsulation is new to the dairy industry and has already found several applications, such as encapsulation of omega-3 polyunsaturated fatty acids, chitin, peanut sprout, lactase, iron, vitaminC, probiotic bacteria and many more [118], [62], [23], [27], [1], [34].

Bioactive ingredients are very much sensitive to environmental and processing conditions, such as temperature, light, pH, acid etc. Moreover, these ingredients can't remain in the gastric condition and in most of the cases are unable to reach the absorption sites. To address this problem, Many encapsulation techniques have been proposed to entrap the bioactive ingredients so as utilize in the dairy industry. The choice to coat material is also an important factor to be considered for microencapsulation. Eventually, a good deal of research has been carried out with regard to supplementing functional/nutritional ingredient using microencapsulation for milk and dairy products.

II. MILK

1.2.1 Microencapsulation of functional ingredients

Consumers today are more aware about the beneficial health aspects of the food they consume. Beyond satisfying hunger, it is expected that the foods should also have properties to prevent disease and ameliorate physical and mental well-being [95], [126].

A wide range of encapsulation methods have been proposed for the microencapsulation of the bioactive components to be fortified in milk but no single procedure has become universally applicable [17]. The suitability of any microencapsulation technique relies primarily on the molecular structure and characteristics of an individual bioactive substance [11]. Coating materials are chosen, based upon the characteristics of the individual functional components and the type of vehicle products. For the application of microencapsulated nutraceutical materials into milk, several methods have been investigated so far

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[111]. As described in the next sections, the nutraceutical ingredients that have been applied to milk are chitosan, iso-flavone, mistletoe, conjugated linoleic acid, Inonotus obliquus, and peanut sprout extract.

1.2.1.1 Chitosan

Chitin is the second most abundant natural biopolymer on earth [119]. The deacylated form of chitin, called chitosan, has been of great interest for the past few years due to its broad range of health-promoting functions. Following the approval of chitosan as a feed additive by the United States Food and Drug Administration (USFDA), the food industry also is trying to utilize its nutritional effects, and consequently, some developed countries have recognized chitosan as a functional food ingredient [119]. Milk is one of the potential vehicles for chitosan supplementation, but the characteristic bitter taste, along with its off-flavour and the colour of chitosan or chitin-derived products, is a big constraint on its application to milk. As a result, the incorporation of microencapsulated chit oligosaccharide into milk has been studied using polyglycerol mono stearate (PGMS) as a coating material [27]. Having 88.08% encapsulation efficiency, the chitosan microcapsules were found to be very stable and only 7.6% of chit oligosaccharide was released from the microcapsules during 15 days of storage at 4∘C. With regard to the physico chemical and sensorial properties of milk , microencapsulated chitooligosaccharide had a very insignificant effect. Therefore, there is a huge prospect of developing microencapsulated chitooligosaccharide-supplemented milk for the mass market.

1.2.1.2 Mistletoe extract

Mistletoe has been used as a folk medicine for several centuries in Far Eastern as well as in European countries for it's the rapeutic value [97]. Lectin is the principal functional component of mistletoe, and temperature, pH, and other processing conditions make it very unstable. Therefore, it is crucial to protect lectin from environmental or processing conditions during food application Kim etal Demonstrated that if encapsulated with PGMS, mistletoe extract could be successfully applied to milk [55]. As a coating material, PGMS was efficient in ensuring the stability of lectin as well as in releasing lectin in the intestinal ambience. This study concluded that the addition of microcapsules containing mistletoe extract did not produce any undesirable quality changes in the milk.

1.2.1.3 Inonotus obliquus

Inonotus obliquus, commonly known as Chaga mushroom, is a parasitic fungus, growing on the living trunks of mature birch. For several centuries, this fungus has been used as a treatment for gastrointestinal cancer, diabetes, and cardiovascular disease in Russia, Poland and the Baltic countries. Furthermore, some of recent research has revealed that this fungus has unique beneficial health functions, such as protection of the DNA from oxidative stress, anti-inflammatory, and antitumor activities [48], [102], [103]. Therefore, beyond its traditional use as a therapeutic medicine, this mushroom can be applied to milk and dairy products in order to make its beneficial effects available to the mass population. Ahn et al. Investigated how to improve the functionality of milk by adding microencapsulated I. obliquus extract [3]. In order to prevent the off-flavor and high viscosity of I. obliquus, a microencapsulation technique was developed in that study, using MCT as a coating material. The optimum conditions of microencapsulation were ascertained, based on response surface methodology (RSM). Evaluating the resulting data, this study drew the conclusion that the microcapsules of I.obliquus could be applicable in milk without affecting the consumers' acceptance of milk in terms of its physicochemical and sensorial properties.

1.2.1.4 Peanut sprout extract

Resveratrol, a famous anti-aging polyphenol, is also known for its anti-arthritic, anti-inflammatory, and anti carcinogenic properties [57]. A number of other health-promoting functions of resveratrol have been discovered through some recent investigations. Due to its potential beneficial health effects, incorporation of resveratrol into food products is becoming a popular practice in the food industry. Various sources of resveratrol have been reported but the greatest source is known to be peanut sprout extract [49] from the peanut sprout (Figure1.1). The extreme sensitivity of resveratrol to environmental conditions is considered a major challenge in its food application. Water-in-oil-in-water (w/o/w) technology has been emerged recently as an advanced microencapsulation procedure which is currently being used to protect functional components [104], [133]. To protect the functional effects of resveratrol, a study demonstrated that peanut sprout extract could be microencapsulated with a greater efficiency of 98.74% under conditions optimized by response surface methodology [73]. To produce w/o/w emulsion, MCT was used as primary coating material and whey protein concentrate (WPC), maltodextrin, and gum Arabic were used as secondary coating material to entrap the peanut sprout extract. This invention was followed by another work which was conducted regarding the viability of w/o/w emulsion technology to preserve the functional effects of resveratrol as well as the quality of milk [76]. This study demonstrated that higher concentrations of peanut sprout extract microcapsules (PPSEM) had a significant effect on the pH and color values of milk while lower concentrations of PPSEM did not produce any noticeable change in the physicochemical properties of milk. It was determined that the concentrations of PPSEM of up to 0.1% (w/v) could be used in formulating functional milk while maintaining the physicochemical and sensory properties of milk.

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Figure 1.1 : peanut sprout

Milk and dairy products comprise the major portion of the functional food market and these are essential parts of our daily diets. Hence, the application of functional ingredients to milk is worth while. For the efficient delivery of bioactive ingredients into milk, microencapsulation is regarded as a feasible solution. Using this technology the proper delivery of the bioactive ingredients can be ensured by protecting the functionality until the ingredients reach the target site in the intestine. Reduced dose and cost efficiency are two other potential advantages of microencapsulation.

1.2.1.4 Conjugated linoleic acid

Of several bioactive lipid components in milk, conjugated linoleic acid (CLA) has attracted considerable attention for having beneficial health functions, such as being anti carcinogenic and anti atherogenic [16]. In order to utilize the bioactive functions of CLA, it should be protected from oxidation during food application. Recently, one study reported that whey protein concentrate (WPC) has been a very effective coating material to prevent the oxidative deterioration of CLA [46]. This study revealed that the application of WPC-coated CLA did not cause any objectionable change in the sensorial properties of the vehicle. Lateron, Choietal. Manifested that the Maillard reaction products (MRPs) of whey proteins and maltodextrin showed better microencapsulation efficiency and solubility as a coating material [31]. However, in vitro and in vivo studies have to be performed to confirm the efficiency of microcapsules in releasing CLA in the gastrointestinal condition.

1.2.1.5 Isoflavone

Isoflavones are soy-derived phytoestrogens with a potent biological activity. Studies have reported that isoflavone could play a significant role in lowering blood cholesterol level [10], [36]. However, its bitter taste along with a beany off-flavor and brown color of isoflavone has limited its direct application into milk.

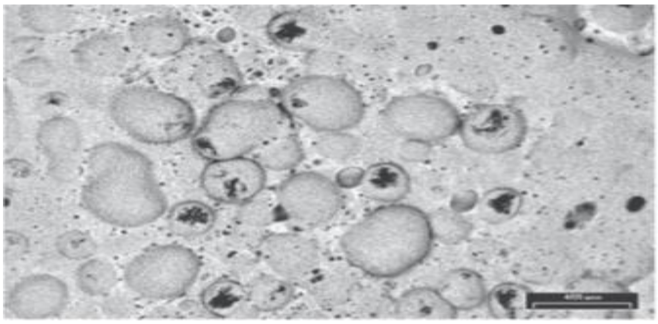

Figure 1.2 Photomicrograph of microencapsulated chitooligosaccharide with polyacylglycerol monostearate. The photograph was taken at 50× magnification. Source: Choi et al. (2006). Reproduced with permission of Asian-Australasian Association of Animal Production Societies (AAAP) and Korean Society of Animal Science and Technology (KSAST).

Jeon et al. developed microencapsulated isoflavone-added milk by entrapping the chitosan particles with medium chain triglyceride(MCT) [45]. This study determined that more than 70% encapsulation efficiency could be obtained, given the coating to core material ratio as 15:1. The release rate of the isoflavone during 3-days of storage at 4∘C was limited up to 8%. In simulated gastric conditions, the release rate was recorded as 4.0–9.3% while in the simulated intestinal condition, the release rate was 87.6%. The sensory analysis revealed no undesirable quality changes of the milk after the addition of the isoflavone. Furthermore, Jeonetal. provided evidence that microencapsulated isoflavone-supplemented milk had strong cholesterol-lowering effect in rats. Later, Kim et al. showed that if using either MCT or PGMS as coating material, isoflavone together with milk β-galactosidase could be supplemented into milk without affecting each other's delivery to the absorption site [54].

1.2.2 Microencapsulation of iron

Iron is an essential microelement and has several important functions in the human body. Lack of this element leads to one of the most prevalent nutritional deficiencies around the world called iron deficiency anemia (IDA) which affects nearly 20% of the world's population [84]. Iron deficiency usually results from the inadequate supply of iron in the diet, poor bioavailability of iron from the digested foods, or a combination of the two [37]. According to the National Institute of Health (NIH), the average daily requirement of iron for adult men and women are 8 and 18mg, respectively, whereas the requirement for children and pregnant women are 8 and 27mg, respectively [91]. Therefore, it is suggested that the intake of iron could be enhanced, especially through foods fortified with iron. Milk and dairy products are being considered as suitable iron-fortifying vehicles due to their high consumption and as an outstanding source of essential nutrients. Moreover, milk is well known for its low content of iron (0.2mg/kg), despite being abundant with other nutritional elements [35].

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Therefore, fortification of milk with iron could be an important solution in the fight against iron deficiencies. For an effective iron supplementation, it is crucial to select the appropriate iron compound with a high degree of bioavailability, and more importantly, to select the suitable technique which is safe, does not change the organoleptic qualities of the food vehicle and helps provide iron with high level of stability and bioavailability. It has been suggested that microencapsulation can prevent the negative effects, such as fat oxidation, the off-taste from the iron, the off-color, and sedimentation from iron fortification, and can secure an effective supplementation of iron in milk and dairy products.

For a feasible iron fortification in milk, microencapsulation of iron salts has begun using a type of phospholipid called SFE-171 as a coating material and afterwards a series of studies were carried out to improve this technology [19], [134], [128], [80]. Ferrous sulfate microencapsulated with lecithin (SFE-171) has been studied in animal and human subjects and it was reported that this product had the same bioavailability as ferrous sulfate and did not deteriorate the organoleptic quality of the fortified products [20]. A few investigations revealed that the constitutive ingredients of milk, such as casein, calcium, and whey protein lessen the bioavailability of ferrous ion by interacting with this ion, and this negative interaction has been shown to be avoided by microencapsulating iron with SFE-171 [42], [87], [19]. Kwak et al. revealed that encapsulating the iron salts with fatty acid ester (FAE) can increase both the microencapsulation efficiency and iron fortification efficiency. Using PGMS, they obtained 75% microencapsulation efficiency. The microcapsules could effectively protect the iron until it reached the intestine. Moreover, the resultant milk, after the addition of the PGMS-coated iron capsules, did not show any change in the organoleptic quality [63].

The feasibility of the liposome technology to encapsulate ferrous sulfate to incorporate into milk has also been investigated [132]. Previous studies suggested that, compared to other encapsulation techniques, liposome has some advantages because the liposomal system is well characterized, easily made and composed of food-acceptable ingredients [2], [58]. On the other hand, liposome is thermodynamically unstable and the encapsulation efficiency is quite low, because of which [132] postulated that these downsides can be checked by the addition of cholesterol and Tween 80 to the liposome system. In a recent comparison study, it was shown that maintaining the optimum conditions encapsulation efficiency could be obtained up to 85.5% and 81.1% in case of liposome and the FAE method, respectively [1]. However, based on the sensory analysis, this study demonstrated that the milk fortified with the FAE-microencapsulated iron had the higher resemblance to the control. Due to its simplicity, low cost, and speed, the FAE method is considered a practicable iron fortification method.

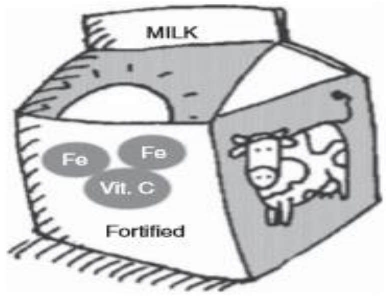

Figure 1.3 Milk fortified with encapsulated Fe and Vit. C.

In recent years, though on a small scale, this technique is being practiced industrially in South Korea and is expected to expand in near future. As w/o microcapsule emulsion is not suitable for long storage, the w/o/w technique has been implemented to solve the problems regarding storage of iron microcapsules [74]. MCT and whey protein isolate (WPI) were used as the primary and the secondary coating materials to make w/o/w emulsion. Iron salts coated with MCT and WPI can be easily modified into powder form which guarantees a longer shelf-life, moreover, by virtue of the hydrophilic nature of the outer coating material (WPI), the microcapsules are susceptible to dispersion into milk [75]. W/O/W technology is the most advanced technology so far to be used in microencapsulated iron fortification in milk and speculatively is an appropriate method for industrial application.

1.2.3 Microencapsulation of vitamins

Supplementation of micronutrients into foods has become a common approach to increase the daily consumption of essential vitamins and minerals. Food fortification began long ago in the industrialized countries for the efficient control of several deficiencies, such as vitamin A and D, several B vitamins, iodine, and iron. In recent years, some developing countries have also started implementing food fortification to combat micronutrient deficiencies. Given the enormous success of iodine fortification in salt, a number of other food products are considering supplying specific micronutrients. Fortifying milk and dairy products can play a vital role in the supply of essential micronutrients. Vitamin D fortification of milk started in the United States in the 1930s on the recommendation of the American Medical Association's Council on Food and Nutrition in order to fight the prevalence of rickets in children [124], [90]. The initiative proved successful and currently, in the United States and Canada, it is mandatory to fortify milk with vitamin D [23]. This success has led to the initiation of vitamin A fortification of milk as required by regulation in the 1940s. Even though apart from these two types of vitamins, the addition of other trace elements into milk has not become common practice, food scientists are trying to incorporate some other important trace elements into milk using the appropriate technology. The

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purpose of vitamin fortification in food products is mainly to meet the special nutritional needs of infants and aged people and to prevent diseases in nutrient deficient populations. This aim cannot be attained unless a proper delivery system is determined, which can supply the nutrient to the appropriate site by protecting it from both environmental and gastrointestinal conditions until it reaches the appropriate site. Water-soluble vitamins are sensitive to environmental conditions, such as temperature, pH, moisture, light, etc. and are susceptible to processing and storage. The sensitivity and the risk of degradation of vitamins can be prevented by encapsulating them with suitable coating materials. Ascorbic acid (vitamin C) is a water-soluble vitamin and is one of the essential micronutrients to be incorporated in to milk. This element has very important functions in the human body. Besides its antioxidant role, ascorbic acid plays a crucial role in enhancing the absorption of iron by converting ferric iron into a ferrous state [89]. In order to overcome some of the shortcomings regarding the instability of ascorbic acid during its incorporation into milk, microencapsulation has been proposed as a possible solution. Both dry powder and liquid microcapsule technologies are applicable to encapsulate ascorbic acid. Of several factors, the choice of coating materials, the physicochemical properties of the core materials, the encapsulation process, and the ultimate properties of microcapsules all have to be considered. Lee et al. have determined the suitability of PGMS as a coating material for ascorbic acid during its incorporation into milk [72]. In their research, the encapsulation efficiency was 94.2% and the release of ascorbic acid during 5 days of storage was limited up to 6.7% with no significant change in the sensorial properties of the fortified milk. In an earlier study, these authors demonstrated that along with PGMS, MCT could also be a potential candidate as a coating material [71]. In vitro examination carried out in this study indicated that these two coating materials showed very high levels of efficiency in both preserving the ascorbic acid in the simulated gastric fluid and releasing it in the simulated intestinal fluid. This study has revealed that the encapsulated ascorbic acid, when consumed with milk, increased the serum iron, which implied that the microencapsulated ascorbic acid increased the bioavailability of iron.

1.2.4 Microencapsulation of lactase

Lactose, a type of carbohydrate, is the second biggest component in milk (Figure 1.4). Generally, this component comprises about 4.8–5.2% of milk. After intake, lactose is hydrolyzed into glucose and galactose, which are easily absorbed in the small intestine. However, a large number of non-Caucasians, aged people in many Western countries, and various ethnic population groups are intolerant to lactose, as they lack adequate amount of lactase (β-D-galactosidase EC 3.2.1.23) in their gastrointestinal tract [121].

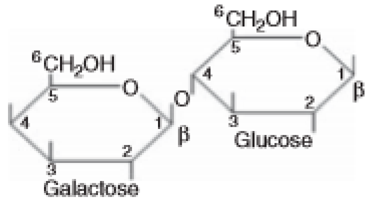

Figure 1.4 Chemical structure of lactose.

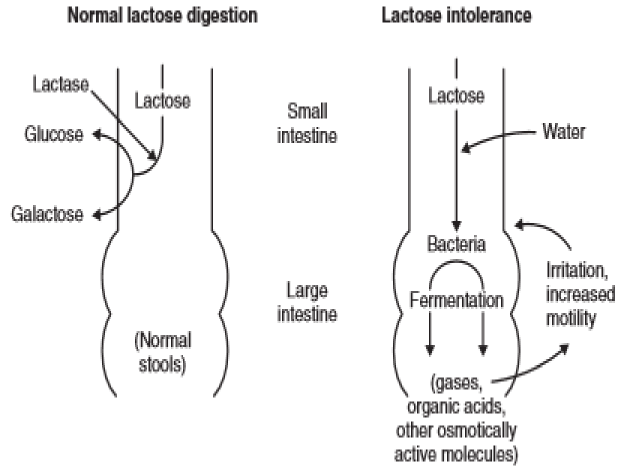

Figure 1.5 Digestion of lactose. Left: normal level of the small intestinal lactase. Lactose is hydrolyzed and the component monosaccharides are absorbed. Right: lactase deficiency with resultant diarrhea from failure to hydrolyze the lactose.

When lactase-deficient people consume milk, they have one or more of the following clinical symptoms: abdominal pain, watery diarrhea, bloating, and cramping, and these symptoms are generally known as lactose intolerance [39]. The reduction of lactase activity is a genetically programmed process which occurs after weaning when lactose is no longer an essential element in the human diet [125]. Nearly two-thirds of the populations in the world have trouble digesting milk, and reportedly, 97% of Thais and 84% of Koreans are lactose-intolerant, compared to only 5% of Northern Europeans with the same problem [63]. To assist the large number of lactose-intolerant people, several efforts have been undertaken so far. For the very first time, β-D-galactosidase was added to the milk which hydrolyzed lactose during processing. A technique called "lactase immobilization," where lactose was continuously hydrolyzed, further promoted the process. In terms of lactose hydrolysis, this method was simply ingenious. But lactose-hydrolyzed milk did not attract much attention due to the high sweetness resulting from the hydrolysis of lactose. Therefore, there was a demand for hydrolyzing the lactose after consumption. In the 1980s, milk was sold together with powdered lactase on US market. Many lactose-intolerant people found it uncomfortable to take lactase along with milk and eventually this attempt failed to meet consumers' demand. A few years later, a further advanced technique known as

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UF/RO, was introduced whereby lactose was removed from the milk through ultra-filtration (UF). However, removal of lactose resulted in the loss of some important macro/micro nutrients from milk and the loss of the original sweetness of milk. Moreover, milk with lactose removed was inefficient in calcium absorption and translocating it into bone. Microencapsulation is the most efficient method so far to aid lactose maldigesters as it is capable of withholding the activity of lactase in the stomach and releasing the entrapped lactose in the intestine. This method has been recognized as one of the most advanced and commonly used techniques in the food industry in order to protect the beneficial effects of certain food ingredients. Due to being entrapped, β-D-galactosidase cannot hydrolyze lactose until it reaches the human gastrointestinal tract. For the microencapsulation of lactase, a wide range of research has been conducted with different methods and a variety of coating materials. One of the preliminary research studies was done by Baik et al they used polymerized 1, 6-diaminohexane and sebacoyl chloride as a coating material [12]. To prevent the immediate hydrolysis of lactose, entrapment of β-galactosidase in a lipid vesicle was also tried [109]. The vesicle is generally called liposome; in this strategy β-galactosidase is entirely enclosed by a phospholipid membrane which is later destroyed in the stomach by the presence of bile salts. However, according to their claim, the entrapment efficiency was only 28%. To overcome the instability of the conventional liposome suspension, dried liposomes have been produced using the dehydration-rehydration method in some instances [53], [108]. Kim and his fellow researchers developed dried liposomes containing β-galactosidase in the presence of trehalose and they found it had a very high entrapment efficiency; in addition, trehalose could prevent the fusion of liposomes and the leakage of the entrapped enzyme [53]. In order to advance the microencapsulation technique, one of the most innovative works was revealed by Kwak et al. whereby lactase was microencapsulated with fatty acid ester [62], [67]. To microencapsulate the lactase, the choice of coating materials is very important. Inparticular, it is necessary that the coating materials should be stable in the gastric ambience, while in the intestinal condition they essentially need to be hydrolyzed easily. Kwak and his group have determined MCT, PGMS or a combination of those two as suitable coating materials. In a subsequent study, it was shown that the β-galactosidase microcapsules coated with MCT or PGMS remained intact in the gastric juice and are hydrolyzed when they reached the upper intestine. The reason is attributed to the sensitivity of the microcapsules to bile salt sand pan creatin present in the intestinal fluid.The maximum efficiency of microencapsulation was found when the ratio of coating to core material was 15:1. As a coating material, MCT was inexpensive, but the quality of MCT was not so satisfactory. On the other hand, PGMS is a very high quality material while it is expensive. Therefore, inorder to obtain aviable coating material the research group of Kwak tried a combination of MCT and PGMS with a ratio of 5:5. The yield of microencapsulation was maximal with the combination product when the ratio of coating and core material was 15:1. Through some functional tests and clinical demonstrations, milk containing β-galactosidase coated with fatty acid esters did not show any change in sweetness during storage, meanwhile it maintained the characteristic taste and flavor, and was digestible to lactose-intolerant people [64]. To encapsulate β-galactosidase, a number of coating materials including milk fat, agar, gelatin, etc. have been tried out [82], [83], [21], [120]. While most of the coating materials were inefficient in terms of microcapsules yield, storage, and the quality of microcapsules, the invention of encapsulating β-galactosidase with MCT and PGMS has been shown to solve nearly all of those problems. Those coating materials could protect the hydrolysis function of β-galactosidase in milk while in the human body β-galactosidase was released to hydrolyze the lactose. Therefore, the milk containing microencapsulated β-galactosidase does not change in sweetness and is digestible to lactose-intolerant people. Moreover, the milk can maintain its characteristic taste and flavor due to the splendid feature of the fatty acid ester.

During microencapsulation a small amount of β-galactosidase remained uncapsulated to partially hydrolyze the lactose. This partial breakdown increased the sweetness of milk to some extent. To solve this, centrifugation of the residual enzymes has been suggested [62]. However, this process involved a high cost and was not suitable for large-scale production. Later, from the same inventors, a special technique called "ozone treatment" made it possible to inactivate the residual enzymes without affecting the sensory profile [66], [65]. Furthermore, this method could eliminate the pathogenic microorganisms from the food system. These inventions have proved a huge success in solving lactose-intolerance problems; however, one crucial aspect regarding microencapsulation of β-galactosidase still remained unaddressed. Since the water-in-oil (w/o) emulsion technique was used to encapsulated β-galactosidase, the resulting microcapsules made with MCT or PGMS were in liquid form. And these w/o microcapsules were found to be easily oxidized under normal environment conditions. Hence, the storage of microencapsulated β-galactosidase was a major concern; besides, the liquid type of microcapsules was inconvenient to transport. In order to overcome the disadvantages relating to w/o emulsion, recently an innovative work has been done by Kwak and his fellow researchers. They applied the water-in-oil-in-water (w/o/w) technique to encapsulate β-galactosidase. The w/o/w emulsion technique facilitates longer storage of microcapsules as it can be dried into a powder form; as a result, the delivery of the substance is now remarkably convenient. In this double emulsion technique, two coating materials were used, whereby the inner phase was

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hydrophobic in nature and the outer phase was hydrophilic. MCT was used as the inner coating material and whey protein isolate (WPI) was chosen as the outer coating material. The hydrophilic characteristics of the WPI allow it to be dispersed in milk easily; the conversion of powder form is also associated with the hydrophilic nature of the outer phase of the w/o/w emulsion (Figure1.6). It was also shown that the yield and quality of microcapsules were further increased with this new technology.

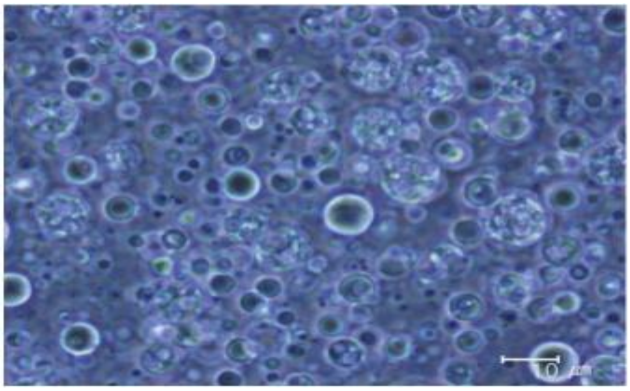

Figure 1.6 Water in oil in water (w/o/w) emulsion of microencapsulated lactase. Source: Ahn et al. (2013). See plate section for color version.

It is obvious from the latest research results discussed that some of the inventions have great potential to aid lactose maldigesters and can make meaningful progress in the processing of dairy products. In particular, the, double emulsion technique invented by Kwak et al. is potentially a prime candidate for industrial application.

III. YOGURT

1.3.1 Microencapsulation of functional ingredients

Yogurt is one of the six traditional snacks that has showed a historic rise in consumption in recent years and it is among the few top popular snack foods for children aged 2–17 years. There is an ever increasing trend in yogurt consumption throughout the world and this trend seems set to continue in the coming years. Due to the high nutritive value and consumers' preference, yogurt can be a perfect medium for supplementing functional ingredients. Like many other carrier foods, the addition of bioactive substances to yogurt can be challenging with regard to the stability of the bioactive compounds and the quality of the yogurt. However, the incorporation of microencapsulated functional ingredients into yogurt has not become popular yet. Very few studies have been attempted to incorporate the microencapsulated functional components in yogurt. Marine fish oil and peanut sprout extracts are the two substances that have been used to develop nutraceutical yogurt.

1.3.1.1 Marine fish oils

Marine fish oils are the biggest source of polyunsaturated fatty acids (PUFAs). Many scientific publications suggest that regular consumption of PUFAs has preventive effects on cardiovascular diseases and an increase in the intake of these substances is recommended by supplementing them with certain foods [15]. A number of studies have reported the beneficial effects of long-chain polyunsaturated fatty acids (LCn-3PUFAs) in preventing CVD [13], [25], [61], [47]. Marine fish oils are the principal source of LCn-3 PUFAs, especially eicosa pentaenoic acid (EPA) and docosa hexanoic acid (DHA). Since the average daily intake of these essential fatty acids is far below the minimum daily requirement, enrichment of food products with these acids can ensure the minimal daily intake [60]. As one of the most popular snacks, yogurt is very suitable for enrichment of LCn-3 PUFAs. Attempts have been made to incorporate omega-3 fatty acids directly into yogurt [26], [85]. As PUFAs are easily oxidized in the food system and give rise to undesirable sensory attributes along with a fishy smell, it was demonstrated that encapsulation of these acids could decrease oxidation and minimize the undesirable taste and flavor [14]. Maltodextrin and gum Arabic are excellent at encapsulating food ingredients. Estrada et al. revealed that the unwanted sensorial changes including the fishy smell of marine fish oil were inhibited by coating it with those two substances [34]. As it was encapsulated, the addition of marine fish oil did not significantly change the pH, color, water-holding capacity, and other physicochemical properties of yogurt. These authors suggested that strawberry yogurt fortified with microencapsulated fish oil can be manufactured with the direct addition of microencapsulated fish oil before homogenization and pasteurization. Spray-drying of emulsion is the most common technology for the microencapsulation of marine fish lipids, so far. However, higher surface oil levels in the microcapsules obtained using spray-drying is often problematic for longer storage. Recently, a number of food products containing microencapsulated fish oil prepared as spray-dried complex coacervates have been launched. Complex coacervation is a liquid–liquid associative phase sparation that occurs due to poly electrolyte complex [69]. Tamjidi et al. demonstrated that the complex coacervate of gelatin/gum acacia was very effective in encapsulating omega-3 PUFAs in order to prevent oxidation and fishy smells [127]. After drying and extraction of the surface oil, the resultant microcapsules achieved better stability, and it was found that the addition of the semi crocapsules to yogurt caused to are duction in the whey separation and gave higher viscosity. Sensory panellists reported observable smell or taste in the omega-3 PUFAs microcapsules-supplemented yogurt.

1.3.1.2 Peanut sprout extract

Peanut sprout extract, with numerous reports on its beneficial health functions, is a potential candidate for application to yogurt. But direct addition of peanut sprout into milk products is problematic A recent study has demonstrated that microencapsulated peanut sprout extract

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supplementation with a concentration of upto 0.5% had a very limited or no undesirable change in the physicochemical properties of the yogurt [77]. Encapsulation of the peanut sprout extract can prevent oxidation; moreover, it ensures the protection of resveratrol, the main functional ingredient in peanut sprout. Based on this finding, a viable technology may come up through future extensive studies to develop nutraceutical yogurt supplemented with peanut sprout extract. Food scientists emphasize the utilization of peanut sprout mainly because of its exceedingly high content of resveratrol. Microencapsulation has the potential to facilitate the supplementation of this important functional component by protecting it from environmental and processing conditions.

1.3.2 Microencapsulation of iron

Like other typical dairy products, yogurt contains a negligible amount of iron, and this product is primarily consumed by women, children, and adolescents who are often regarded as iron-deficient [18], [32]. Hence, to reduce the incidence of iron deficiency, fortification of iron in yogurt would be worthwhile. But iron fortification in yogurt is a little more challenging compared to other dairy products, because it may create an imbalance between the lactic acid bacteria (LAB) and other bacteria [38]. This microbial imbalance is attributed to the fact that LAB do not require iron for their growth, while some other bacteria, such as pseudomonas spp and E. coli, have the mechanism and the affinity to uptake iron in order to enhance their growth [93], [43]. More than a decade ago, one study reported that yogurt could be fortified with protein chelated iron without any substantial change in the organoleptic properties [38]. Even so, due to the enormous success of the microencapsulation technique in protecting the nutritional elements in the food industry, it was realized that the encapsulated iron could be more efficient in terms of protecting the quality of iron as well as keeping up the microbial balance in the fortified yogurt. Consequently, a group of researchers applied microencapsulated iron to drinking yogurt for the first time [56]. This study had a far-sighted view of encapsulating the iron and ascorbic acid separately with PGMS to fortify both of these nutrients in yogurt. Fortification of iron together with ascorbic acid served a dual purpose: it replenished the iron and vitamin C content in milk, and additionally, the ascorbic acid played a huge role in the absorption of nonheme iron in the duodenum by forming a chelate with ferric iron [79], [56] suggested that as a result of being encapsulated, iron and ascorbic acid did not change the sensorial quality of the yogurt; furthermore, iron reached the gut with a higher level of bioavailability aided by the ascorbic acid. Soy yogurt which is considered a good alternative choice for lactose-intolerant people has been reported to be fortified with iron without any adverse effect on the rheological and sensorial properties [24]. A recent finding provided evidence that microencapsulation had no negative effect on the probiotic bacteria in yogurt, nor did it produce any oxidized flavor in the fortified yogurt [44]. Of the very few methods of iron fortification in yogurt, it is obvious that microencapsulation is the most suitable way to carry, protect, and deliver this element efficiently so far. The delivery system depending on nanotechnology is another possible way of iron fortification in yogurt. However, based on current knowledge, further extensive studies involving microencapsulation and nanotechnology are required to establish an effective way of supplementing iron into yogurt.

IV. CHEESE

1.4.1 Microencapsulation for accelerated cheese ripening: Ripening is an important process in cheese manufacturing which is entirely responsible for texture and flavor development. Ripening time varies from 4 weeks to 3 years, depending on the type sand varieties of cheeses. During this period cheese satta in their characteristics by going through several chemical, biochemical, and microbiological changes where by protein, fat, and lactose are broken down into primary and secondary products [52]. Acceleration of cheese ripening has been studied using various factors, such as enzyme, temperature, and microorganisms for a long time. Shortening the ripening time would provide both economic and technological advantages to the cheese manufacturers. Eventually, the addition of proteolytic and lipolytic enzymes to cheese started long ago as a popular process. If the enzyme is added to the milk, only a small portion of enzyme is retained in the curd and increases the cost, moreover, the addition of the enzyme to milk results in the reduction of the cheese yield and flavor defects [33]. Addition of the enzyme in curd also gives rise to undesirable property changes. It has been suggested that encapsulation is an efficient way to solve the drawbacks associated with direct addition of enzymes. Various methods have been investigated to encapsulate cheese-ripening enzymes. In one of the preliminary attempts, cell free extracts (CFE) of L. lactis ssp. diacetylactis were encapsulated using milk fat for Cheddar cheese ripening [82]. Approximately eight times more diacetyl and acetoin were produced in the cheese with the added encapsulated enzyme. Later, Braun and Olson added the encapsulated CFE of St. lactis var. multigens and Gluconobacter oxydans and obtained a higher amount of flavor compounds [22]. During cheese manufacturing in this study, a 16% loss of capsules in the whey was observed. Till today considerable research has been conducted to develop a viable microencapsulation technology to accelerate cheese ripening. There have been several reports on encapsulating proteinases, chymosin, and lipases using liposomes, carrageenan, hydrogel beads, and milk fat as encapsulating materials [105], [51], [70], [50], [9]. Most of the studies mentioned above dealt with a single enzyme or a mixture of either lipolytic or proteolytic enzymes. However, Kheadr et al. revealed that a liposome-

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encapsulated enzyme cocktail containing flavourzyme, neutral bacterial protease, acid fungal protease and lipase could be incorporated into cheese milk for a balanced ripening [52]. But liposome technology has some constraints for industrial application, as this technology is expensive and has uncontrolled release during ripening. However, besides improving the liposome technology, other approaches are also being applied in order to develop a more suitable technology for the microencapsulation of cheese ripening.

1.4.2 Microencapsulation of iron

In addition to milk and yogurt, a few other dairy products have also been emphasized with a view to facilitating their on fortification program. As a consequence, considerable research has been conducted so far to supplement iron into different types of cheeses. In a comprehensive review, [37] has pointed out that when supplementing in free form, iron has the tendency to aggregate with casein and other ingredients in milk to affect the sensorial quality of cheese, and contrary to that, encapsulated iron showed no significant changes in the cheese quality. During fortifying Havarti cheese, [43] observed better organoleptic properties when their on was microencapsulated. Moreover, they found less amount of iron was drained off through the whey when it was microencapsulated. An animal study using Petit-Suisse cheese as a food vehicle has revealed that the ferrous sulfate encapsulated with SFE-171 had the same degree of bioavailability as free ferrous sulfate [80]. Generally, free iron is known to produce an unpleasant flavor and color in the food vehicle due to its oxidizing effect while iron coated with PGMS was found to have no deteriorative effects on the flavor characteristics of cheese [63].

V. CONCLUSION

Owing to the dramatic increase in functional foods, dairy products are widely utilized as vehicles for supplementing bioactive ingredients. To improve the incorporation process of health functional ingredients into dairy products, the application of microencapsulation has recently started. Microencapsulation has remarkably improved the efficiency of the supplementation procedure of bioactive materials in milk and dairy products. The advantage of microencapsulation lies in the fact that this technique can protect both the ingredients and the vehicle product from undesirable property changes. The advantages of micro encapsulation has opened up new opportunities that can revolutionize dairy products processing. Based on recent research, it is worth noting that some of the inventions are not only suitable for small-scale processing but also are potential candidates for commercial applications

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