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Review Article
Role and Mechanisms Lowering Cholesterol by Dietary of Probiotics and Prebiotics: A Review

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Abstract
Cholesterol plays a pivotal role, since it is a necessary compound for functions of brains and heart. Moreover, cholesterol is an important precursor for biosynthesis of some important hormones. Notwithstanding, high cholesterol is a leading risk factor for human cardiovascular diseases such as stroke, coronary heart disease and atherosclerosis. Most pharmacological agents that are utilized in the control of hypercholesterolemia are bile acid sequestrates, fibrates, niacin and cholesterol absorption inhibitors. Nevertheless, these drugs have also been correlated with many adverse effects that limit treatment agreement as well as the state of life. Live micro-organisms that possess healthy benefit upon consumption are namely probiotics being different from prebiotics. The latter are defined as non-digestible food components and selectively stimulate the growth of probiotics. Probiotics and/or prebiotics could be used as alternative supplements to exert health benefits, including cholesterol-removal effects. In addition, there are several articles on the cholesterol-lowering ability of probiotics. Many mechanisms for cholesterol removal by probiotics have been suggested. Some of these mechanisms include: deconjugation of bile via bile salt hydrolase, binding of cholesterol to cellular surface of growing cells, coprecipitation of cholesterol with deconjugated bile and incorporation of cholesterol into the cellular membrane and short-chain fatty acids produced by oligosaccharides. The current paper reviews the mechanisms of action of the cholesterol-lowering potential of probiotic micro-organisms and functional food contain probiotics and prebiotics, with the aim of lowering the risks of cardiovascular and coronary heart diseases and also focus on the ability of combination of probiotics and prebiotics (synbiotics) to be novel substitutional to chemical drugs for elimination of hypercholesterolemic problems.

Key words: Cholesterol, cholesterol-lowering, bile salt hydrolase enzyme, probiotics, synbiotics, hypocholesterolemic effect, conjugated bile salts

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INTRODUCTION

Epidemiological investigations have exhibited that higher than regular serum cholesterol level is fully associated with the increase of cardiovascular disease (CVD), which is one of the leading causes of death and inability in advanced countries[1]. The World Health Organization (WHO) has predicted that CVD will stay the prime problem of death by 2030 and will affect almost 23 million people[2].

In the human body, there are two sources of cholesterol; (i) Internal biosynthesis by the liver and (ii) From the diet, principally from foods including animal fats[3]. Although drugs such as statins, efficiently lower cholesterol, side effects are generally published, leading to patients seeking alternative choices, such as weight loss, exercise, supplements and diet[4]. It is worth to mention that, the use of traditional cholesterol-lowering drugs, adds additional great costs to the individual, as most often, anti-cholesterol drugs are taken for life. The bioactive ingredients of natural health products and functional foods are responsible for their potency in health improvement and disease prevention, consequently, functional foods have been approved as a favourable dietary approach to reduce total cholesterol, especially for those patients with borderline blood cholesterol levels who do not demand cholesterol-lowering medications[5-7].

In addition of the affordable prices of functional foods, no side effects have been traced for consumption of these food products containing probiotics. Moreover, they have been claimed to possess health benefits including: antimicrobial activity against pathogens, reduction of serum cholesterol, anticarcinogenic properties, antimutagenic properties, immune system stimulation, anti-diarrhoeal properties, improvement of inflammatory bowel disease[8-13]. Health benefits awarded by probiotic bacteria are strain specific. It is eligibility to remark that no strain will give all proposed benefits and not all strains of the same species will be effective against defined health conditions.

According to Gibson and Roberfroid[14], prebiotics are non-digestible ingredients present in food and possess health benefits. On the other hand, probiotics are defined as live microorganisms that have health benefits for human[15]. It is worth to mention that combination of prebiotics and probiotics, namely synbiotics, can be applied in formulating novel functional food. The effect of synbiotic functional food (combination of prebiotic and probiotic) on several aspects of health have also been documented[14,16,17]. It is worth to mention that exact mechanisms of reduction of serum cholesterol by probiotics are still unclear[18].

The proposed mechanisms of serum cholesterol lowering by probiotics include (1) Cholesterol binding to cellular membrane of probiotics[19], (2) Deconjugation of bile salts by probiotic bacteria producing bile salt hydrolase (BSH), (3) Production of short chain fatty acids upon fermentation by probiotics in the existence of prebiotics[20], (4) Co-precipitation of cholesterol with deconjugated bile[21] and (5) Incorporation of cholesterol into the cellular membranes of probiotics through growth[18]. Nevertheless, some of the aforementioned mechanisms were found to be strain dependent and conditions produced under laboratory conditions wouldn't be workable in the *in vivo* systems[22].

Very little attempts have been carried out to develop synbiotic products with a specific need to remove cholesterol in order to exert hypocholesterolemic effect *in vivo*. Most previous studies had emphasized on the cholesterol lowering properties of probiotics or prebiotics individually but not synergistically. The main reasons for that would be the complication raised due to the number of possible probiotic-prebiotic combinations and also the complex evaluation of the synergism from the interaction effects[23].

Although all these would seem unfeasible with the conventional screenings, there is no attempt to develop such cholesterol lowering synbiotic product using statistical approaches. The response-surface approach would be an extremely useful tool for developing, optimizing and estimating interaction effects[24]. This study discovered the beneficial health of dietary probiotics and prebiotics in terms of lowering cholesterol level in blood. Meanwhile, this study will help the researchers to uncover the critical areas of mechanisms of such effect of probiotics and prebiotics that many researchers were not able to explore.

DEFINITION OF PROBIOTICS

The name probiotic comes from the Greek words *'pro bios'* which means 'for life'[25]. Probiotics word was first used by Lilly and Stillwell[26] to explain the 'substances excrete by one micro-organism that stimulate the growth of another'. A number of definitions of the term 'probiotics' have been used over the years but the one derived by FAO. and WHO[27] and established by the International Scientific Association for Probiotics and Prebiotics[15] best explains the extent and scope of probiotics as they are known now: "live micro-organisms which, when administered in enough amounts, exert health advantage on the host".

Probiotics are defined as "living micro-organisms, which upon ingestion in certain numbers exert health benefits on the host beyond inherent basic nutrition" by Guarner and Schaafsma[28].

Health benefits of probiotics: Various health benefits of probiotic organisms have previously described by Parvez *et al.*[29] and Shah[30]:

- Restoration and maintenance of healthy flora
- Nutrient synthesis and bioavailability: Fermentation of food with lactic acid bacteria increases folic acid, niacin and riboflavin levels
- Preventative and therapeutic effects against diarrhea: Competes with pathogenic viruses or bacteria for binding sites on epithelial cells and through their effects on the immune system
- Improves body's natural defense
- Alleviation of lactose intolerance: It is due to the lactic acid bacteria in fermented milk increasing lactase activity in the small intestine
- Enhancement of immune system functions
- Allergies: Probiotics exert a beneficial effect on allergic reaction by improving mucosal barrier function
- Cancer prevention
- Control of blood cholesterol: By the increasing the excretion of de-conjugated bile salts derived from cholesterol

DEFINITION OF PREBIOTICS

Most prebiotics as known now are fermentable, non-digestible carbohydrates with a various number of sugar groups from two up to several numbers. Some examples are resistant starch, maltodextrin, lactulose, fructo and galacto-oligosaccharides[31].

Prebiotics general uses: Prebiotics are described as "non-digestible food components that beneficially influence the host by selectively stimulating the growth of one or a limited quantity of bacteria in the colon and thus improve health"[14]. An increase of *Bifidobacterium* spp. would be exposed for the inulin and exopolysaccharide (EPS). The bifidogenic influence of the EPS would be established by culturing on specific medium.

The potency of a probiotic LAB strain to survive in the Gastro-Intestinal Tract (GIT) might be developed by oligosaccharides facilitating the metabolism and growth of LAB in the lumen[32]. Dietary fiber, fundamentally polysaccharides and oligosaccharides fermented in the colon might act as prebiotics[33,34]. The influence of prebiotics as enhancers of the growth of probiotic lactic acid bacteria has been documented in human[35]. *Bifidobacterium* sp. and *Lactobacillus* sp. particularly produce a positive effect on human health[36,37].

The reduction of cholesterol level could be due to the cholesterol assimilation by *Lactobacillus*[38], as a prebiotic supplementation could improve the lactic acid bacterial count. Related results have been described by Mohan *et al.*[39] and Kalavathy *et al.*[40] and a similar hypocholesterolemic effect would be observed in broiler chicken supplemented with beta fructans from chicory as a source of prebiotic[40]. The mechanism(s) involved in the overall hypocholesterolemic effect of mono, oligo saccharide supplementation is not fully documented. However, malto-oligosaccharide (MOS) is looked as substrate for lactic acid bacteria. Increasing level of MOS also increases the CFU of this lactic acid producing bacteria[41].

IMPORTANCE OF CHOLESTEROL

Cholesterol (cholest-5-en-3-ol) is a steroid alcohol and a crucial structural component of biomembranes, which is closely related with phospholipids. The cholesterol molecule includes itself in the membrane with the same orientation as the phospholipid molecules (Fig. 1). It is a significant part of an animal body because it plays an important role in determining the membrane fluidity, hormones and nerve fiber insulation and it is required for other functions such as production of vitamin D and synthesis of various steroids. Biosynthesis and diet are the two ways that organisms obtain cholesterol.

Cholesterol biosynthesis, a highly-adjusted process occurs in all animal tissues but in higher mammals this machinery is notable in liver. Bile is the major excretion path of cholesterol from the body, as unesterified cholesterol. In human, about 400 mg of cholesterol per day is transformed to bile acids and only approximately 50 mg is changed to hormones. Cholesterol that is produced in the liver is transported to cell membranes by lipoprotein. Excess cholesterol accumulates in fatty insoluble deposits called plaques along artery walls narrowing and clogging them restricting the blood flow causing of coronary artery disease[42].

Biosynthesis of cholesterol: New biosynthesis is responsible for less than 50% of body's cholesterol[43]. Figure 1 shows the steps of cholesterol biosynthesis in the human body[44].

Regulating cholesterol synthesis: In healthy people, approximately 1 g of cholesterol is synthesized and 0.3 g is consumed per day. The body controls a comparatively fixed quantity of cholesterol (150-200 mg dL^{-1})[11]. This is done principally through checking the level of *de novo* synthesis. Biosynthesis of bile acids consumes most of cholesterol. Body's constant supply of cholesterol from cells is regulated by three separated mechanism, namely, HMG-CoA reductase

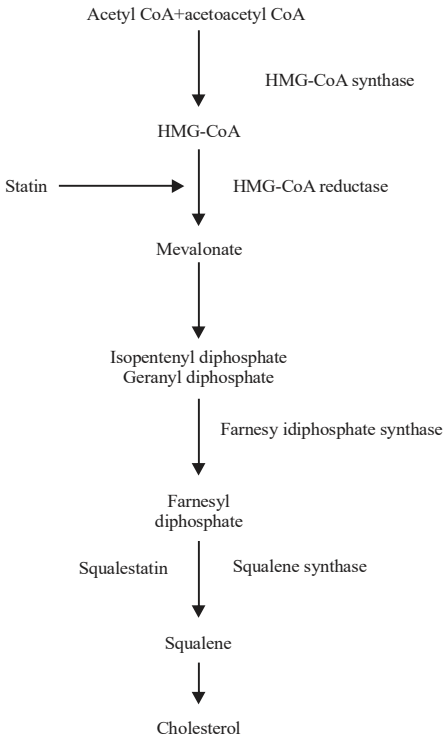

Fig. 1: Cholesterol biosynthesis: (44)

(HMGR), acyl CoA cholesterol acyltransferase (ACAT) and via HDL, mediated reverse transport and receptor-mediated uptake of LDL[43,11]. The liver utilizes part of cholesterol pool to produce bile salts which are stored in the gall bladder. It is known that bile salts play a pivotal role in emulsification ingestion and absorption of fats.

The remainder of the cholesterol is utilized for other requirements of the body. To do that, the liver integrates cholesterol from its pool with triacylglycerols and covers it with a special protein so that it could be dissolved in the blood. The liver then removes them from the blood. Lipoprotein lipase (LPL) exists in excess all over the body, particularly in the walls of the arteries. This enzyme is included in removing triacylglycerols from VLDL cholesterol. In the process, the VLDL shrinks in size and a relatively larger portion of it is made up of what is called intermediate-density lipoproteins (IDL)[45].

Mechanisms of cholesterol removal by probiotics: Many potential cholesterol removal mechanisms have been proposed[18]. Most of the hypotheses suggested to date are based on *in vitro* experiments and few efforts have been made to assess the potential hypocholesterolemic mechanisms based on *in vivo* examinations[23].

De-conjugation of bile salts via BSH: Bile salt hydrolase (BSH) enzyme is one of the most significant mechanisms for cholesterol removal. The enzyme is effective for bile salt deconjugation in the enterohepatic circulation (Fig. 2). It has been discovered in several LAB species common to the gastrointestinal tract (Table 1)[46,47].

Several researchers proposed that BSH activity should be considered in the selection of probiotic organisms with cholesterol-lowering properties, as non-deconjugating organism doesn't has the ability to eliminate cholesterol from the culture medium in any significant extent[48]. It is worth to mention that de-conjugation of bile salts exhibits host specificity. In mammalian host, the de-conjugation site is dependent on the host species. In the small intestine of mice, *Lactobacillus* flora is present where bile salt deconjugation occurs[49], while in mankind, a significant flora starts only at the end of the ileum and fully developed in the large intestine indicating that, bile salt de-conjugation activities begin at the end of the ileum and active in the large bowel[50]. Thus, if the deconjugation of bile is an essential mechanism for lowering cholesterol levels, cultures used for human *in vivo* trials must be selected from suitable origins.

Numerous studies have proposed that the distribution of BSH activity in *Bifidobacterium* and *Lactobacillus* is connected to the type of a genus, species or even strains. Major of the probiotic lactic acid bacteria that isolated from human intestine and faeces revealed BSH activity. Nevertheless, it must also be noted that not all strains isolated from the intestine or faeces have BSH activity suggesting that bacteria without this enzyme could survive and grow through bile acids conditions[51].

Conjugated bile salt is regularly re-circulated into enterohepatic circulation, whilst de-conjugated bile salts are less soluble and excreted in faeces[60]. The secreted bile salts are substituted by new bile salts from cholesterol in the bloodstream. Therefore, the more bile secreted the more cholesterol eliminated from the bloodstream[61]. In recent years, the possibility of using bile salt deconjugation by LAB to lower serum cholesterol level in hypercholesterolemic patients and prevent hypercholesterolemia in normal people received increasing attention[56]. Several strains of *Lactobacillus* and *Bifidobacterium* had the ability to reduce cholesterol through the mechanism of enzymatic deconjugation of bile by bile salt hydrolase enzyme[62-64].

Co-precipitation of cholesterol with deconjugated bile: The *in vitro* elimination of cholesterol during co-precipitation of cholesterol with deconjugated bile salt was described by

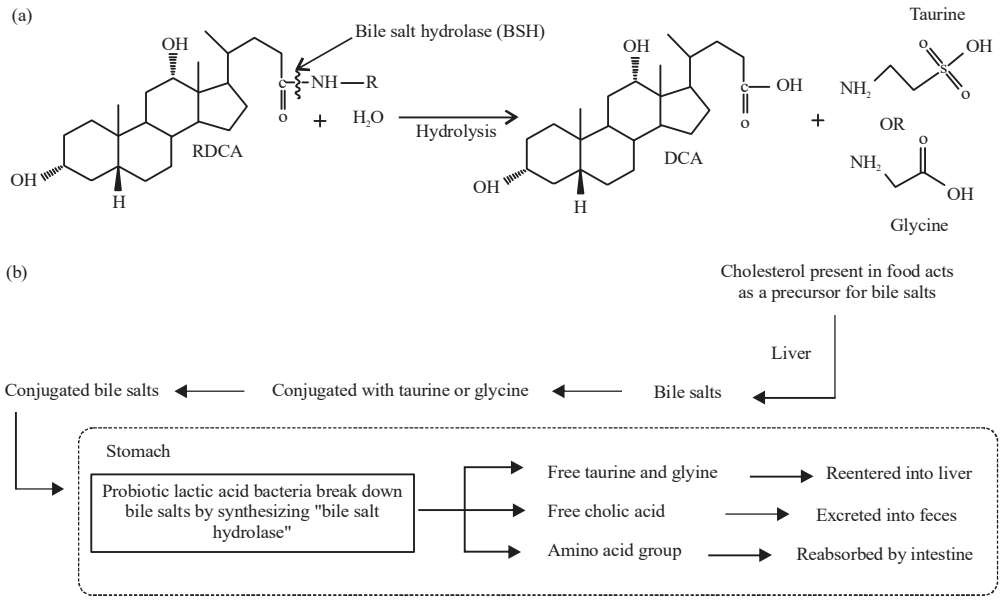

Fig. 2(a-b): (a) Hydrolysis of conjugated bile salts by the bile salt hydrolase (BSH) enzyme R: Amino acid glycine or taurine. RDCA: Glyco or tauro-deoxycholic acid, DCA: Deoxycholic acid and (b) Cholesterol as the precursor for the synthesis of new bile acids and the hypocholesterolemic role of bile salt hydrolase[58,59]
Source: Jones *et al.*[58] and Anandhraj *et al.*[59]

Table 1: Influence of the habitat of a genus, species or even strains of Bifidobacteria and Lactobacilli on the distribution of bile salt hydrolase (BSH) activity

Probiotic organisms	BSH activity[a]	Source of micro-organism	References
Bifidobacterium adolescentis	+	Human faeces	Tanaka *et al.*[51]
B. animalis	+	Human faeces	Tanaka *et al.*[51]
B. breve	+	Human faeces	Tanaka *et al.*[51]
B. coryneforme	-	Human faeces, honey bee gut	Tanaka *et al.*[51]
B. infantis	+	Human faeces	Tanaka *et al.*[52]
B. longum	+	Human faeces	Tanaka *et al.*[52]
Bifidobacterium spp.	+	Intestines of mammals and Insects, human faeces	Grill *et al.*[53]
Lactobacillus acidophilus	+	Faeces, mammalian intestines	Corzo and Gilliland[46]
L. buchneri	-	Human intestine	Moser and Savage[54]
L. casei	+	Human faeces	Brashears *et al.*[55]
L. fermentum	+ (-)	Infant faeces (fermented beets)	Moser and Savage[54]
L. gasseri	+	Human faeces, mammalian intestines	Tanaka *et al.*[51]
L. helveticus	+	Human faeces	Tanaka *et al.*[51]
L. paracasei subsp. *paracasei*	+	Raw milk	Moser and Savage[54]
L. plantarum	-	Human faeces	De Smet *et al.*[56]
L. rhamnosus	+	Human faeces, bowel drain, yoghurt	Tanaka *et al.*[51], Moser and Savage[54]
Lactococcus lactic subsp. *lactis*	+	Boza	Shehata *et al.*[57]

[a] +: Exhibited bile salt deconjugation activities, -: Without bile salt deconjugation capability

several researchers[21,23]. Cholesterol might co-precipitate with deconjugated bile salts which observed at pH values below 5.5, contributed by bacterial fermentation and formation of short chain fatty acids[65]. At acidic pH, deconjugated bile salts are protonated and precipitated, while taurine-conjugated bile salts remained ionized in solution and glycine-conjugated bile salts are partially precipitated without hydrolysis[66].

It was previously reported that the removal of cholesterol from a medium was contributed by the disruption of destabilized cholesterol micelles as a result of bile salt deconjugation, followed by the precipitation of cholesterol with the free bile salts as pH decreased[62,65]. *The in vitro* cultivation of *L. casei* reduced cholesterol mainly by destabilizing cholesterol micelles and co-precipitating cholesterol with deconjugated bile salts[55] at pH>6.0.

The co-precipitation of cholesterol with de-conjugated bile was correlated with the pH of media[52]. Co-precipitation of cholesterol occurred at pH ranging[60] from 3.78-4.69. Cholesterol precipitation was the highest with cholic acid at pH below 5 while minimal co-precipitation occurred with

sodium glycocholate was negligible regardless the pH. So, it can be said that, co-precipitation of cholesterol was not entirely pH dependent.

Incorporation of cholesterol into cellular membrane: The mechanism for cholesterol lowering is the incorporation of cholesterol into bacterial cellular membrane; some *Lactobacillus* strains have the ability to incorporate cholesterol into cellular membranes through growth which would reduce cholesterol absorption from the intestine into the blood resulting in its reduction in serum total cholesterol[60,67].

Lactobacillus acidophilus removed most of the cholesterol from cultivation medium by incorporation cholesterol into cellular membrane[55]. Meanwhile, Kimoto *et al.*[68] investigated the removal of cholesterol by several strains of lactic acid bacteria from media. They observed difference in the fatty acid profile for cells grown in the presence and lack of cholesterol. Lipids of probiotics lactic acid bacteria are essentially concentrated in the membrane, suggesting that cholesterol combined into the cellular membrane had modified the fatty acid composition of the cells. The association of cholesterol into the cellular membrane enhanced the concentration of saturated and unsaturated fatty acids, resulting in improved membrane strength.

Lye *et al.*[18] reported that cholesterol that binds or adheres to the bacterial cells would more likely be less available for absorption from the intestine into the blood. This mechanism involves the correlation between incorporation of cholesterol and pH of the growth medium. Cell membranes from culture grown without pH control was found to contain significantly more cholesterol compared to cultures grown under controlled pH of 6.0. Cell membranes from culture without pH control were reported to be higher in cholesterol concentration as compared to the whole cells grown under the same conditions, while there was no significant difference of cholesterol concentration between cell membranes and whole cells grown at controlled pH 6.0. However, the amount of cholesterol removed in the membrane fraction of *L. acidophilus* did not account for the total amounts removed by the culture. The authors suggested that some cholesterol might loosely associate with the cells and do not get incorporated into the membrane resulting in loss during the isolation procedure. Probiotics caused also cholesterol removal by incorporation into the cellular membranes during growth. Furthermore, this mechanism has been studied by determining the possible sites of cholesterol binding within the membrane phospholipid bilayer of probiotic cells using fluorescence probe.

The *in vitro* investigations on *Lactococcus lactis*, revealed that there was variance in fatty acid distribution pattern for cells grown with and without cholesterol. Cells grown in the presence of cholesterol included significantly higher amount of 18:1, 18:2 and total unsaturated fatty acids, but lower amount of 16:0, 18:0 and total saturated fatty acids compared to those grown in the absence of cholesterol. Alteration of the fatty acid composition of the cells was as a result of cholesterol being combined into the cellular membrane after its removal from the media[59].

Assimilation of cholesterol by probiotics: Assimilation of cholesterol is another proposed mechanism involved in lowering cholesterol level by some Bifidobacteria and lactic acid bacteria[53,69,70]. Cholesterol can be mostly assimilated during bacterial growth, being bound without transformation onto the cellular surface and incorporated within the membrane phospholipid layer[18,47,67,68].

In order to be able to assimilate cholesterol, prebiotics have to be viable and growing. Also, assimilation requires growth under anaerobic conditions and the presence of bile acids[59,70]. The *in vitro* experiments showed that strains that did not grow well in medium containing bile salts were unable to assimilate cholesterol; while those that grew well in the presence of higher concentrations of bile salts, has more ability to uptake of cholesterol from medium[71,72].

It has been reported that the bacterial culture which actively take up cholesterol from the laboratory medium would function *in vivo* to exert a hypocholesterolemic effect. Thus, testing the bacteria for its ability to assimilate cholesterol from the medium would be a prerequisite for the selection of probiotic strain with hypocholesterolemic effect[68].

Binding of cholesterol to cells: Tahri *et al.*[48] found that large amount of cholesterol was retained by growing bacterial cells and more than 40% of cholesterol was exerted by sonication from cells of *Bifidobacterium breve* ATCC 15700. The absorbed cholesterol could not be detached even after several washes indicated that the binding between cholesterol and growing cells was intense. The mechanism of cholesterol removal was studied from different points of view when non-growing Lactococci cells reportedly removed cholesterol *in vitro*. Hypocholesterolemic effect of the probiotic has also been attributed to their ability to bind cholesterol in the small intestine[18].

Dambekodi and Gilliland[69] found that both heat-killed cells and resting cells removed cholesterol although, the amount of cholesterol removed was less compared to growing ones, leading to the hypothesis that cholesterol not only

might be removed by living cells during growth but also via dead cells and some cholesterol was restrictive to the cellular surface. Strains of *Lactobacillus gasseri* were capable of elimination cholesterol *in vitro* via binding onto cellular surface and this capability appeared to be growth and strain specific[64]. Not with standing, ability of probiotics to remove cholesterol during different growth conditions, further strengthened the aforementioned hypothesis[68].

Effects of prebiotics on cholesterol-lowering: Prebiotics may also have an effect on lipid regulation. Notwithstanding the mechanism is currently unknown, studies have shown positive results and mechanistic hypotheses have been developed. Previous studies on diabetic rats revealed that, when xylooligosaccharides (XOS) substituted simple carbohydrates in the diet, the serum cholesterol and triacylglycerols that considerably increase in diabetes were found to decrease and liver triacylglycerols increased to a comparable level to that observed in healthy rats[73]. The reduction of blood lipids might be due to the inhibition of a lipogenic enzymes in the liver, as the result of the action of propionate generated from the fermentation of prebiotics by probiotic lactic acid bacteria[74,75].

Whereas prebiotics could be used in decreasing hyperlipidemia induced by diabetes and other conditions, decreases in lipids have not been observed in healthy subjects[74], which is a useful safety feature as misuse or overdose does not seem to have negative effects. There are many possible mechanisms by which prebiotics may lower cholesterol, such as:

- Increase viscosity in the upper intestinal tract includes the esophagus, stomach and duodenum. This viscosity might act as a natural barrier and reduce the (re) absorption of lipids, containing cholesterol and bile acids. This would lead to improve faecal output of cholesterol and bile acids would be resulted in higher cholesterol catabolism in the liver, which would lead to lower plasma cholesterol concentrations[76]
- Reduce luminal pH level as results of production SCFAs which in turn alter intestinal bacterial population[77]. At low pH, the amount of soluble bile acids decreases and as a result, lipid absorption decreases and faecal bile acid secretion rises
- Adjusting hepatic triacylglycerol synthesis. The lipid-lowering action of inulin is probably due to alterations of hepatic triacylglycerol synthesis, very low-density lipoprotein cholesterol (VLDL-c) secretion and reduced reabsorption of circulating bile acids[78]. Thus, higher bile acid secretion leads to enhance utilization of liver cholesterol to re-synthesise bile acids and as hepatic pools of free cholesterol decrease to reach a new steady state, endogenous cholesterol synthesis will increase. This leads to increase the activities of 7-hydroxylase and 3-hydroxy-3-methylglutaryl-coenzyme A reductase (HMG-CoA reductase) to compensate for the loss of bile acids and cholesterol from liver stores
- Fermentation of prebiotics, the hypocholesterolemic effect of prebiotics is acquired from a metabolic effect[79], as these compounds are fermented in the lower intestinal tract[80]. Meanwhile, Rossi *et al.*[81] observed that butyrate was the major fermentation product from inulin, though acetate was produced from fructooligosaccharides. The hypocholesterolemic effect of prebiotics has been essentially referred to SCFAs. Butyrate is identified to prevent liver cholesterol synthesis and generate a source of energy for human colon epithelial cells and propionate may stop the synthesis of fatty acids in the liver, whereby reducing the rates of triacylglycerol secretion. Propionate is also inclusive in the regulation of hepatic cholesterol synthesis and it decreases the degree of cholesterol synthesis which could result in lowering of plasma cholesterol levels[78]
- Affect serum inulin and oligofructose cause a decrease in hepatic fatty acid and triacylglycerol synthesis through a coordinated decrease in the activity of all lipogenic enzymes. Delzenne and Kok[82] found that the triacylglycerol reducing effect of oligofructose examined in rats was induced by its anti lipogenic action in the liver, that is by decreasing the activity and probably the expression of all lipogenic enzymes, because prebiotic consumption significantly decreases serum insulin and glucose, which induce lipogenic enzymes[82]
- Rats fed on resistant starch for four weeks exhibited decrease in serum total cholesterol. It was obvious that total serum cholesterol was found to decline *in vivo* after probiotic supplementation[83,84]

CONCLUSION AND FUTURE RECOMMENDATIONS

The consumption of synbiotics is winning publicity particularly in the servicing of health and protection of disease. In particular, the function of synbiotics as a cholesterol-lowering agent has been investigated broadly. Growth has been made in the recent years on the production of synbiotic functional foods with a hypocholesterolemic effect. In spite of these assumed advantages from the human clinical investigations accomplished in recent years, a critical outcome has failed to be reached due to controversies raised.

Further investigation on the role of BSH enzyme in cholesterol lowering might open a new prospect to understand the mechanism of cholesterol lowering and formulating an alternative cholesterol lowering food supplement.

SIGNIFICANCE STATEMENT

This study discovered the beneficial health of dietary probiotics and prebiotics in terms of lowering cholesterol level in blood. Meanwhile, this study will help the researchers to uncover the critical areas of mechanisms of such effect of probiotics and prebiotics that many researchers were not able to explore. Consequently, a new theory on mechanisms of lowering cholesterol as influenced by probiotics and prebiotics may be arrived at.

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