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Review Article

Lactose Intolerance with Special Emphasis on Probiotics for Management

Devaraja Gayathri* and Vasudha M

Department of Microbiology, Davangere University, Davangere, Karnataka, India

***Corresponding Author:** Gayathri Devaraja, Department of Microbiology, Davangere University, Davangere, Karnataka, India.

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Abstract

Intestinal lactase deficiency leads to Lactose intolerance (LI) and this condition is found across the globe. The activity of Lactase has been found to be high and vital during early stages of life, but in most mammals, lactase activity declines gradually with aging process. In addition, beta galactosidase activity exists entire lifespan of human. And this dominant inherited genetic trait is called lactase persistence. Generally, primary lactase deficiency is an inherited deficit persists in majority of the world's population, while secondary lactase deficiency could be the consequence of intestinal disease. Recent studies have shown that the risk of lactose ingestion symptoms depends on the dose, lactase expression, intestinal microbiota and the extent of sensitivity GIT. Conventional diagnosis pattern for lactase include; basis of a history of gastrointestinal symptoms, occurrence of after and aggravated by milk ingestion, a breath test demonstrating abnormal hydrogen levels, an abnormal lactose tolerance test, and stool sample for reducing substances or acidic pH and small intestinal biopsy to assess direct lactase enzyme activity. A number of research groups have carried out study on alternate approaches, such as exogenous β-galactosidase, usage of probiotics, pharmacological and non-pharmacological strategies that can enhance contact time between substrate and enzyme that delay gastrointestinal transit time, and colonic adaptation through chronic lactose ingestion. This review majorly highlights the diagnosis of LI with special emphasis on probiotic usage and treatment.

Keywords: *Lactose; Lactase (lactase-Phlorizin Hydrolase); Hydrogen Breath Test (HBT); β-galactosidase; Probiotics*

Abbreviations

SCFA: Short-Chain Fatty Acids; H_2: Hydrogen; CO_2 Carbon Dioxide; CH_4: Methane; LI: Lactose Intolerance; LHP: Lactase-Phlorizin Hydrolase; LCT Gene: Lactase Gene; SNP: Single Nucleotide Polymorphism; HBT: Hydrogen Breath Test

Introduction

Milk and their products are nutritionally rich foods providing protein, minerals such as calcium and magnesium, several B vitamins and fat-soluble vitamins A and D. The lower intakes of these nutrients through milk, specifically Calcium and vitamin D, could result in not just increased risks of osteoporosis but also to several other chronic diseases, such as hypertension, stroke, and colon cancer [1].

Double sugar Lactose (β-D-galactopyranosyl-(1→4)-D glucose), (glucose and galactose), which exits in mammalian milk (0.72 g/10 ml in human milk, 0.47 g/100 ml in cow milk) and also, minimum in some marine mammals [2,3]. In food industry, lactose has been used in wide way. Because of its physiological properties, as only 30% as sweet as sugar, it is used in sweets, confectionery, bread and sausages since lactose provides good texture and act as binder. It is used in infant milk formula and it is also used as a binder and filler in tablets [4,5].

Lactose has to be degraded into its absorbable monosaccharides as glucose and galactose. Lactose digestion takes place in the small intestine by Lactase-phlorizin hydrolase (EC 3.2.1.23), is a β-galactosidase enzyme responsible for the hydrolysis of lactose into glucose and galactose. Lactase is found most abundantly in jejunum and synthesised in microvillus membrane of small intestinal [5,6].

The enzyme Lactase, has two active sites, the initial site hydrolyse lactose into glucose and galactose monosaccharides, therefore absorbable by intestinal mucosa, the later site hydrolyse phlorizin. Hydrolysis takes place in jejunum, as it contains meagre number of bacteria; so marginally lactose gets fermented. Later, these monosaccharides are selectively absorbed by enterocytes further to blood stream, glucose serves as a source of energy and galactose moves to the liver, hence glycolipids and glycoproteins [7,8].

During lactase deficiency either alactcia/hypolactsia, disaccharide lactose osmotically fluid out to intestinal lumen leading to increased liquid accumulation. Through Salvage pathway, lactose breaks down to SCFA (short chain fatty acid, production of H_2, CO_2 and CH_4) perhaps leading to GIT ailments [9,10].

It has been well documented that, during infant stage, intestinal lactase activity would be maximum. Later, 2 - 12 years of age two patterns grows out, as hypolactasia/"lactase non-persistence" group, and "lactase persistence" individuals who will have lactase activity in their adulthood [11,12].

Lactose intolerance

Lactose intolerance (LI) is an inability to digest the lactose, caused by a deficiency of lactase (β-galactosidase) in the small intestine with typical clinical symptoms including abdominal pain and distension, borborygmi, flatus, and diarrhoea occur between 30 minutes and 2h after the ingestion of lactose [13]. Due to acute diarrhoea symptoms immediately after milk consumption, their results in LI, and one need to avoid a lactose containing diet such as milk and milk products [14]. Milk is a nutrient dense food and an important part of a healthful diet [15]. Avoidance of milk is a significant risk factor which would result in low bone density. Those who avoid milk, due to LI, consume significantly less calcium and have poorer bone health and probable higher risk of osteoporosis [16].

Long before 400 years BC, Hippocrates once said about lactose intolerance, however, typical clinical signs/symptoms have been reported in recent times [2,17]. Typical symptoms of LI especially among infants, kids and adolescents include acute diarrhoeal illness and related complications. Due to cow milk-protein hypersensitivity, often results in intestinal mucosal membrane injury [18]. Stages of Lactase deficiency can be classified as - primary, secondary, congenital and developmental lactase deficiency.

During primary lactase deficiency condition, marginal otherwise total absence of lactase observed in childhood at different ages of various races leading to lactose malabsorption and lactose intolerance. It has been estimated that around 70% of the population across the globe suffer from primary lactase deficiency. This percentage varies according to ethnicity and type of diet [18]. Primary lactase deficiency also referred as adult type hypolactasia, lactase non-persistence, or hereditary lactase deficiency, is an autosomal recessive condition resulting from the physiological decline of the lactase-phlorizin hydrolase (LHP) enzyme activity in intestinal cells which occurs in a significant proportion of the global population. Generally, lactase deficient persons exhibit typical symptoms at late adolescence and adulthood. However, primary hypolactasia conditions are seen before 20 years of their age. Additionally, a gradual decline in lactase activity may on rare occasions continue after 20 years of age. Treatment suggested include, limited intake or even elimination of lactose or dairy foods. Otherwise, exogenous β-galactosidase intake have been recommended [2,18].

In secondary lactase deficiency, small bowel injury would be much evident followed by acute gastroenteritis, persistent or continuous diarrhoea, overgrowth of enterobacteria, or for other reasons a damage to small intestinal mucosa, and these symptoms though temporary (sometimes) can be observed at any age but are more often during infant stage [18]. Several recent studies and a meta-analysis have shown that kids with rotaviral (and related infectious) or diarrheal illnesses who have no or only mild dehydration can safely continue human milk or standard (lactose-containing) formula without any significant effect on outcome, including hydration level, nutritional condition, duration of illness, or success of medication. However, the World Health Organization recommends avoidance of lactose-containing milks in children with persistent post infectious diarrhoea (> 14 days) whenever there is a failure of dietary trial with milk, yogurt related foods. Since milk and other dairy products are major source of calcium, perhaps continue its intake once the primary symptoms are stopped [2,18].

Further, Congenital lactase deficiency is less incident disorder documented in few infants. Before 20th century, rate of survivability of these patients would have been meagre as there was no ready availability of nutritionally adequate lactose-free milk substitute [18]. Further, data on single autosomal disorder related molecular mechanisms are scarce. Although, histological study using biopsy small intestinal contained normal report, however, very low or total absence of lactase concentration was determined. If this stage of lactase deficiency was not detected early and treated, the situation would be worsening with electrolyte loss on diarrhoea. For which, treatment included with removal and substitution of diet with a commercial lactose-free formula [2,18].

Developmental lactase deficiency has been defined as the relative lactase deficiency observed among preterm infants of less than 34 weeks' gestation [18]. Although lactase is a non-inducible enzyme, in preterm infant's lactase supplemented feeding would favour the production and the expression of the enzyme. In addition, it has been reported that in neonates/young infants only 20% of dietary lactose would get the access to colon. Upon bacterial digestion/degradation of this lactose the faecal pH decreases (5.0 - 5.5 is normal), which has a beneficial effect, favouring certain good bacteria (anaerobic *Bifidobacterium* spp and microaerophilic *Lactobacillus* spp) in lieu of enteropathogens (*Proteus* sp, *Escherichia coli*, and *Klebsiella* sp) [2,18].

Genetics

LCT gene encoded for lactase approximately 50 kb in size and that maps on chromosome 2 (2q21) (Swallow, 2003). For Wild-type lactase non-persistence, two single nucleotide polymorphisms (SNPs) in the lactase gene have been associated along with lactase persistence. There occurs C/T-13910 and G/A 22 018 substitutions at 14 and 22 kb upstream of the 5' end of the said gene in a DNA region, now directly functions as a cis-acting element influencing the promoter. Dominant polymorphism of C/T-13910 with C allele linked to decline in lactase mRNA expression, although, the mechanism of decline after weaning has not been properly elucidated [10,19]. LI persons having heterozygous for either SNP have intermediate lactase activity and observed to be more susceptible to lactose intolerance during stress or gastrointestinal tract infection [2,3].

Prevalence

Lactose intolerance prevalence shows diversity among regions, human populations, continents and across the globe. Although, 70% of global population has lactase non-persistence, not all are intolerant to lactose, as such many nutritional and genetic factors directly influence. Prevalence of lactase non-persistence condition in Asian and African countries ranges between 80-100%, however, among Northern European countries the LI prevalence (adult-type) observed to be very low. Further, data on hypolactasia in Asian populations are sparsely reported, while in western countries, prevalence is higher. In India (north) the frequency of maldigesters was observed to be 48% per 200 subjects during breath test, but among south Indians it was comparatively higher (66%). And in North-west Russia the lactase non-persistence ranges between 16% - 23% [2,5,7,20]. Thus, LI management is a worldwide issue in terms of public health management.

Generally, decrease in lactase expression will be complete during childhood but the decline has also been documented to take place during later stages of adolescence. The rate of loss of lactase activity also varies according to ethnicity, however, exact physiological mechanism is yet to be resolved. Chinese/Japanese lose 80 - 90% of lactase activity within 3 - 4 years after weaning, while, Jews and Asians lose 60 - 70% in many years post weaning, though, in white Northern Europeans it would be 18 - 20 years to reach its lowest expression [3,21].

Use of Probiotics

Probiotics are dietary supplements containing live beneficial microorganisms that can improve the health of host when consumed at appropriate amount. WHO/FAO (2002) defines "probiotics are live microorganisms which when administered in adequate amounts confer health benefits to the host". The most common probiotics are lactic acid bacteria belonged to the genera of *Lactobacillus* and *Bifidobacterium* [22,23]. The mechanism of probiotic action mainly includes competition with harmful bacteria in the gastrointestinal tract for adhesion and nutrients, enhancing host immunity to pathogens, by producing antimicrobial metabolites [24].

Probiotics as health promoters help to improve the lactose digestion and symptoms of intolerance [25]. Fermented milk Yogurt or buttermilk/sweet acidophilus milk is based on the presence of endogenous lactase activity of probiotic bacteria. It has been well tolerated by lactose intolerant people and other fermented products such as buttermilk, kefir and ropy milk also had a 20 - 26% reduction in lactose content [26]. Several studies have shown better lactose digestion and less hydrogen production in patients with lactose intolerance who consumed milk containing probiotics [27]. Probiotic bacteria are also beneficial in the lactose intolerance, a physiological state in human beings where they lack the ability to produce an enzyme named lactase or β-glycosidase, which is essential to assimilate the disaccharide present in milk and needs to be split into glucose and galactose [28].

In this context, some species of lactic acid bacteria, such as *Streptococcus thermophilus, Lactobacillus bulgaricus* and other lactobacilli in fermented dairy products, perhaps alleviate symptoms of lactose intolerance by secreting lactase to the intestine and stomach. It has been estimated that, LI affects almost 70% of world population; consumption of such food would be an ideal method to incorporate dairy products and allied nutrients into the dies of lactose intolerants [26].

However, consumption of lactose by those lacking adequate levels of beta galactosidase would result in symptoms of diarrhoea, bloating, abdominal pain and flatulence. The affected can be elaborated with following reasons. Firstly, lactose digestion would be improved by bacteria if intracellular β-galactosidase has been released. The role of increased permeability in bacterial cell wall and their intracellular lactose hydrolysis caused by bile acids is yet to be resolved. Further, the residual β-galactosidase in the small intestine and decrease the osmotic load of lactose and delaying gastric emptying, thus slows down the intestinal transit. Moreover, short term and long term ingestion of lactose and bacteria in fermented dairy foods would affect the intestinal pH, intestinal resident microbiota or the sensitivity of the individual to intestinal disorders [29]. Generally, these symptoms are due to non-digested lactose entering to the large intestine and being fermented by the intestinal microorganisms. These microorganisms can produce a variety of gases and products that lead to watery stool. Further, *L. acidophilus* and bifidobacteria have been shown to improve digestion of lactose [30].

When intolerant patients were given the unfermented milk supplemented with *L. acidophilus* improved the LI condition or tolerance. *L. acidophilus* strains are the source of lactase, the enzyme needed in the digestion of milk products, which lacks in lactose intolerant people [31]. *L. delbrüeckii* in a dairy food could deliver β-galactosidase activity. Similar bacteria may be alive as long as their membranes are intact which promotes to protect β-galactosidase during gastric transit. These information would reveal that LI can be reduced by incorporating fermented dairy products in their diet consistently and β-galactosidase enzyme by lactic acid bacteria help to overcome LI.

Another problem associated with lactose intolerance is calcium deficiency. A person consuming non-milk diet will naturally develop calcium deficiency, leading to osteoporosis. Calcium absorption is better and more in acidic conditions; hence, if lactose is converted to lactic acid, pH of the gut decreases, i.e. it becomes acidic favouring enhanced absorption of calcium. So, if probiotics are fed to lactose intolerance patients, then milk lactose is hydrolysed by probiotic strains and lactose is assimilated and calcium absorption is also favoured [32]. These controversial results are perhaps due to differences in specific probiotic strains, concentrations, and preparations, as well as due to the subject's susceptibility to gas, osmotic pressure or the individual responsiveness to probiotics [26]. In addition to this, clinical trials of beneficial bacteria and their concentrations standardisation are developed in order to delineate the therapeutic effects of probiotics.

Diagnosis

There are many methods to measure lactose digestion in humans. These methods are based on different principles causing variable accuracy and diagnostic reliability. The possibilities and limitations of the available test methods are discussed below.

Hydrogen breath test

A hydrogen breath test (HBT) is currently the gold standard for assessment of lactose intolerance, as it is sensitive, non-invasive, and cost effective, thus can be performed in subjects of all ages. When lactose is not hydrolysed in the small intestine, undigested lactose will reach the colon and colon bacteria would ferment it. This fermentation process leads to the production of gases including Hydrogen (H_2), methane (CH_4) and carbon dioxide (CO_2) and of lactate and short chain fatty acid that can all be absorbed by the enterocytes. In this test, participant drinks a lactose bolus. In those who are lactose intolerant, the non-digested lactose will be partially fermented in the colon, to produce short-chain fatty acids (lactate, acetate, propionate and butyrate), together with gases (hydrogen, methane and carbon dioxide). Subsequently the gases are transported via the blood and exhaled. Since other biological processes in the human body do not produce hydrogen, the exhaled breath concentration of H2 represents the fermentation of carbohydrate in the colon. Unfortunately the carbohydrate fermentation process in the colon is not restricted to lactose as substrate. Other carbohydrates such as fibre or undigested starch can also be subjected to the similar bacterial fermentation process and lead to the same products in breath. Apart from this, many fac-

tors can influence the composition of the colonic flora, such as medication, colonic acidity and thereby the capacity to form H_2 would be analysed. The result of the test is expressed as positive or negative (i.e. maldigestion yes or no) with a cut-off point of 10 or 20 parts per million H_2 concentration rise above base line levels [33-35]. In understanding the mechanisms behind this test its reliability with respect to the quantity of fermented lactose can be doubted. Despite such uncertainty about results it is until today the most frequently used test to study lactose digestion.

Lactose tolerance test

After consumption of lactose (milk), the substrate will be hydrolysed into glucose and galactose. After intestinal absorption of both monosaccharides the galactose is converted into glucose in the liver. Depending on the feeding state of the individual, glucose is either stored in the liver or released to the blood. In the fed state most glucose will be stored, while in the fasting state the substrate derived glucose will mainly be released to the blood. The rise in serum concentration of glucose directly reflects to the amount of lactose that is hydrolysed. However, the total glucose concentration in blood partly consists of glucose derived from body stores, which makes the test unreliable. Therefore this test has been abandoned in clinical practice [2,11,35,36].

Determination of faecal reducing carbohydrates

Although this test is simple, non-specific, but a positive result indicates the absence of the enzyme. After ingestion of a lactose-containing drink, a stool sample is collected and Fehling's solution is added. The presence of lactose will cause a change in colour from blue to red. A simple kit is available to perform this test [35,36].

Determination of Faecal pH test

Since this test is a non-specific marker, this test is less commonly. Collected stools after ingestion of a lactose-containing drink will be acidic (< pH 6) as in case of lactose intolerance. This indicates fermentation of undigested sugars by the colonic bacteria [11,35].

Plasma glucose test

When lactose is digested in the small intestine, the hydrolysis products are galactose and glucose, subsequently enter the liver where the galactose will be primarily converted into glycogen. Glucose will mostly enter the peripheral bloodstream and induce a prompt rise in blood glucose concentration. Lactose-intolerant subjects will not show such a rise, although there may be a smaller and later increase in blood glucose originating from gluconeogenesis of lactate and/or propionate generated from colonic fermentation of lactose. A rise in blood glucose at least 1.5 mmol/L is indicative of lactose tolerance. The specificity and sensitivity of this lactose tolerance test ranges from 76 to 96%. The magnitude of the increase in blood glucose is subject to several hormonal influences, therefore reducing the reliability of the test compared to the breath hydrogen test [33].

Plasma galactose test

A plasma galactose test, in which a lactose bolus is administered with a 500 mg/kg dose of ethanol to prevent the conversion of galactose to glycogen in the liver is much more reliable than the plasma glucose test, although the necessary invasive sampling (to obtain sufficient blood for the galactose assay) makes the test more difficult to administer on large number of subjects [33].

Genotyping test

Genotyping is a quick, specific and sensitive for detecting lactase gene. This method helps to demark persons with primary hypolactasia from that of lactose intolerance caused by secondary hypolactasia. Further, genotyping could be conducted either on blood or in saliva, although, it is not routinely been available in clinical practice. In addition, identification through simple genetic test for adult hypolactasia has a distinct advantage over conventional methods. While the later methods are cumbersome, requires skilled workers with specialised facilities [2,39].

Treatment and management

Treatment for LI majorly depends on the kind of deficiency exists in one's system. During primary lactase deficiency, the initial set of symptoms depends on actual quantity of lactose needs to be ingested prior to the available lactase is saturated. In this situation, most people with primary lactase deficiency can consume up to 240 ml of milk or 12 g of lactose with no typical symptoms of LI [10].

General approach without following standard guidelines is to restrict milk and other dairy products from their regular diet. As stated earlier, this strategy would have serious disadvantages, chiefly reduced intake calcium, phosphorus and vitamins, and that would result in low bone mineral density. In this line, several strategies have been proposed for alternative approaches, for e.g. lactase enzyme preparations (exogenous β-galactosidase), yogurt and usage of probiotics with lactase activity. These strategies could prolong contact time between enzyme and substrate, therefore delaying gastrointestinal transit time, and chronic lactose condition prevails [2,37,38].

Enzyme-replacement therapy with microbial exogenous lactase represents a possible strategy for primary lactase deficiency. Enzymes could be mixed (liquid form) to milk before its consumption otherwise, can be taken in, in a solid form (capsules or tablets) along with milk and related products [10,39]. Several studies were conducted to obtain "pre-incubated milk" done by mixing soluble enzyme to milk few hours before consumption. This technique is efficient in reducing Hydrogen breath excretion and manifestation of discomfort after milk consumption. However, these trials were carried out on relatively small subjects. In addition, solid lactase preparation in capsules or tablets is an alternative that are commercially available for enzyme replacement treatment. Several studies in this line have investigated and ascertained their efficacy, although, comparative studies have shown that these preparations are costly and less effective than liquid form. This may be due to the enzyme gastric inactivation. Usage of exogenous lactase, especially during mealtime, appears to be effective, practical feasible with no side effects.

As it is already proved that bacterial β-galactosidase activity is the main factor responsible for improving lactose digestion. Further, the enzyme's osmolality and energy density perhaps play an important role, thus the action of β-galactosidase in the small bowel and would decrease the osmotic load of lactose [2,40].

Conclusion

Lactose intolerance has been recognized as a major problem in many children and most adults throughout the world. Milk and dairy products are often assumed to be the cause of gastrointestinal symptoms and inappropriate avoidance can lead to nutritional inadequacy. To overcome lactose intolerance dietary elimination and more-formal testing is typically with faecal pH during watery diarrhoea and hydrogen breath testing has been done. In this context, Probiotics that are beneficial organisms that can alleviate lactose intolerance symptoms through increased hydrolysis of lactose in intestine. In this way applications of probiotics for treatment of lactose intolerance could lead to a promising method in prevention or management of lactose intolerance.

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