

**ANTIBACTERIAL EFFECTS OF PROBIOTICS
ISOLATED FROM MILK PRODUCTS AGAINST SOME
COMMON BACTERIAL PATHOGENS***

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ABSTRACT

Since the beneficial effects of viable probiotic bacteria as dietary supplements have gained huge research interest, Lactic Acid Bacteria (LAB) including *Lactobacillus* spp. are Generally Recognized as Safe (GRAS) bacteria that have been widely used to prepare fermented dairy products such as yoghurts and fermented milk drinks. This research was conducted to isolate and test the antibacterial effects of the probiotics of different commercial fermented dairy products against some common bacterial pathogens. Lactic acid bacteria (LAB) isolated strains were divided into three groups as *Lactobacillus* spp. (*L. casei*, *L. acidophilus*) and *Bifidobacterium* spp. (*B. lactis*) with mean count (Log cfu/ml) of total LAB 7.00 ± 0.70 and 6.00 ± 0.50 in probiotic yoghurts and fermented milk drinks respectively. The spectrum of their antibacterial effects varied against the selected pathogen, the most potent antagonistic microbe *Salmonella typhimurium* and *Staphylococcus aureus* was described as bactericidal effect with mean diameters of zones of inhibition ranging from 26.00 ± 2.00 mm to 12.30 ± 0.40 mm. While, the isolates exert a mild antibacterial effect against *E. coli* was described as bacteriostatic effect with mean diameters of zones of inhibition ranging from 10.00 ± 0.0 mm to 11.33 ± 0.55 mm for yoghurts and fermented milk drink samples respectively. Therefore, results from our present study are expected to encourage people to consume more probiotic dairy products in Najran city, south region of Saudi Arabia as it was revealed that these products contain some probiotic lactic acid bacteria which play a major

role for the beneficial health effects of consumers. Consequently, will discuss the findings and public health importance.

INTRODUCTION

Good health starts with good nutrition, and good nutrition can be protects against diseases later in life. Liquid milk is a common health drink consumed by people of all age groups. A large population in our countries depends on milk from local suppliers. Milk is a product of biological evolution, its role in human nutrition is well known, and its biochemical complex which appears to be the only material to function solely as a source of food. As a consequence, almost complete freedom from nutritional diseases among infants, children, young and adults **Cashman (2002)** and **Hoppe et al. (2006)**.

Human health is a highly attractive word, so food safety remains a major challenge to food producers and to legislators to make an effort to adequately protect the consumers (**Allam et al. 1999-a; 1999-b & 2002**) and (**Abdelhamid et al. 2002**).

Probiotic organisms are live microorganisms principally bacteria thought to be safe and beneficial to the host organism beyond basic nutrition according to the currently approved definition by the United Nations Food and Agriculture Organization and the World Health Organization **FAO/WHO (2001&2002)**. Probiotic lactic acid bacteria (LAB) include (*L. acidophilus*, *L. johnsonii*, *L. casei*, *L. rhamnosus*, *L. gasseri* and *L. reuteri*) and Bifidobacteria include (*B. bifidum*, *B. lactis*, *B. longum* and *B. infantis*) are the most common types of microbes used as probiotics; but certain yeasts and bacilli may also be used **Heller (2001)**. Lactobacilli can produce different antimicrobial components including organic acids (lactic, acetic, propionic acids), hydrogen peroxide, carbon dioxide, low-molecular weight antimicrobial substances, bacteriocins and adhesion inhibitors and thus have gained prominence as probiotics **Stiles & Holzapfel (1997)** and **Quwehand & Vesterlund (2004)**.

Probiotics are commonly consumed as part of fermented foods or cultured foods with specially added active live cultures; such as in some yogurts and fermented milk drinks, or as dietary supplements. Probiotics can be defined as mono or mixed culture of living microorganisms, which beneficially affect the human being by improving the balance of the indigenous microflora, when consumed in an adequate amount as part of the food **FAO/WHO (2001)** and **Isolauri (2001)**. LAB are amongst the most important groups of microorganisms used in food industry **Sonomoto & Yokota (2011)**, many species are involved in the daily manufacturing of dairy products **Ayad et al. (2004)**. Various commercial probiotic preparations are also available in market in the form of

capsules, liquid/gel and powdered that claims for prevention of infectious diseases. Commercially available probiotic preparations include *Lactobacillus* alone (Lactiflora, LactoBacil, Lactocap, Lactovit, etc.) or in combination with *Streptococcus* (Lacticin) or *Saccharomyces* (Laviest), showed beneficial effects **Saggio (2004)**.

Probiotic claimed beneficial effects include treatment of diarrhoea associated with antibiotic therapy, viral infections, chemotherapy and even foodborne diseases; inhibition of pathogenic microorganisms; hypocholesterolemic properties; strengthening of the intestinal mucosal barrier; antimutagenic and anticarcinogenic activities; stimulation of the immune system **Havenaar et al. (1992)**; **Schrezenmeir & Vrese (2001)** and **Chiang & Pan (2012)**. Food and Agriculture Organization and the World Health Organization also states that the appropriate selection of probiotic cultures requires compliance with the following criteria, ability to survive in the gastrointestinal tract; bile salt hydrolase activity; adherence to mucosal or epithelial cells; exclusion or reduction of pathogenic adherence and antimicrobial activity against pathogens **FAO/WHO (2002)**. Also, it has been reported that these probiotics can play an important role in immunological and respiratory functions and could have a significant effect in alleviating infectious disease in children **FAO/WHO (2001)**.

At the start of the 20th century, probiotics were thought to beneficially affect the host by improving its intestinal microbial balance, thus inhibiting pathogens and toxin producing bacteria **Metchnikoff (1907)**. Today, specific health effects are being investigated and documented including alleviation of chronic intestinal inflammatory diseases **Mach (2006)**. One important limitation is that only one kind of probiotic bacteria may not exert protection against all harmful strains that cause gastrointestinal pathogenic infections **Boyle et al. (2006)**. Probiotics are beneficial in gastrointestinal disturbances such as diarrhoea, dysentery and, typhoid etc. **Fuller (1991)**. The rise in antibiotic resistant bacteria has awakened the scientific community to the prophylactic and therapeutic uses of probiotics and to reconsider them as alternatives to antibiotics **Ahmed (2003)**. Various *Lactobacilli*, *Bifidobacteria* and *Streptococcus* species have been evaluated for the prevention or treatment of various infectious diseases and these were found to be safe **Chapoy (1985)**.

Recently, illness resulting from food-borne pathogens has become one of the most widespread public health problems in the world **Mor-Mur & Yuste (2010)**. As a result, there is a need to look for solutions to break the transmission of harmful bacteria along the food chain. Beneficial bacteria, mainly lactic acid bacteria (LAB) and *Bifidobacteria*, may be a useful and effective strategy to prevent or reduce the incidence

of pathogens, thus improving food safety and consumer health **Caplice & Fitzgerald (1999)**; **Callaway et al. (2008)** and **Gaggia et al. (2010)**. Probiotics are involved in synthesis of biotin, vitamin K and B and, in ions absorption as Mg^{2+} , Ca^{2+} , and Fe^{3+} **Holzappel et al. (2001)**. Additionally, some LAB possess antimicrobial properties against pathogens, spoilage bacteria, yeasts, molds and viruses by different mechanisms **Naidu et al. (1999)**; **Alvarez-Olmos & Oberhelman (2001)**; **Rodriguez et al. (2003)**; **Deegan et al. (2006)**; **Settanni & Corsetti (2008)** and **Todorov et al. (2010)**.

Therefore, the an attempt was made to isolate Lactobacillus strains as probiotics from some commercial fermented dairy products and compared it's in vitro probiotics potential with some pathogenic bacterial strains.

MATERIAL AND METHODS

Sampling:

A total of 40 samples of fermented dairy products, (20 each of probiotic yoghurt and fermented milk drink samples) of different brands were randomly collected from different local markets and supermarkets in various districts of Najran city. The collected samples were transported immediately to the laboratory in an insulated container at about 4°C to avoid deterioration and, microbiological analyzed before their expiration dates exceeded.

Methods:

Preparation of yoghurts samples:

The samples of yoghurt were shaken vigorously to suspend the bacterial contents. Then, Ten grams of each sample were dissolved into 90ml of normal saline 0.9% w/v.

Preparation of fermented milk drink samples:

The samples of fermented milk drink were shaken vigorously to suspend the bacterial contents. Then, 10ml of each sample were dissolved into 10ml of normal saline 0.9% w/v.

Isolation of lactic acid bacteria (LAB):

The standard media accepted by the International Dairy Federation for lactic acid bacteria differential the fermented dairy products, is de Man Rogosa Sharp (MRS) **IDF (1983)**. The samples were inoculated into MRS agar medium **De Man et al. (1960)**. Plating was done within 3-6hrs of arrival of the samples in the laboratory and incubated anaerobically in anaerobic jar (BBL, GasPak Plus anaerobic system) at 35°C for 48hrs with replications. The suspected colonies were enumerated (cfu/ml) and purified by successive streaking on MRS agar medium before being subjected to identification. It was adopted according to **Therzaghi &**

Sandine (1975); De Man et al. (1960); Florez et al. (2006) and Roissart & Luquet (1994).

Identification of the lactic acid bacteria (LAB) isolates:

The pure isolates selected were identified on the basis of their cultural, morphological, and biochemical characteristics. Stock culture were also, kept on MRS agar slant at 4°C and streaked every 4 weeks **Herrero et al. (1996)**. Confirmation of the identification was based on the use of API 50CHL (analytical profile index is a system, based on quick identification of clinically anaerobic bacteria).

Pathogenic bacterial strains:

The pathogenic strains used in this work were maintained in brain heart infusion (BHI) agar butt-slants in screw-capped tubes kept at 4°C. They included both Gram-negative (*Escherichia coli* and *Salmonella typhimurium*) and Gram-positive (*Staphylococcus aureus*) clinical laboratory strains.

Antimicrobial activity of probiotic isolates:

For antibacterial effects determination:

The probiotics from the stock cultures were inoculated into brain heart infusion (BHI) broth and turbidity of the broth culture was then adjusted to equal that of 1 McFarland standard **McFarland (1907)**.

The test pathogens from the stock cultures were subcultured in BHI broth under aerobic condition at 37°C for 18h. The turbidity of the broth cultures was adjusted to equal that of 0.5 McFarland standards. The MRS agar medium **De Man et al. (1960)** overlay method was used to test for the presence of antibacterial effects among the probiotics isolates. The prepared probiotics were individually inoculated into the plates by swabbing area in the center of each plate. The plates were incubated anaerobically, at 37°C for 72h for yoghurt and fermented milk drink samples.

The growth in each plate was then overlaid with ≈ 10 ml of molten and cooled in MRS Agar previously seeded with 1ml of 1% the prepared selected pathogen cultures (approximately 1×10^7 cfu/mL⁻¹ of pathogenic bacteria). The agar was allowed to solidify at 4°C for 1h, and the plates were incubated aerobically at 37°C for 24h. The plates were then examined for the presence of zones of inhibition around the wells **Shah & Dave (2002)**. Inhibition was considered positive when the width of the clear zone around the colonies of the LAB isolates was 0.5mm or larger where, resistance was defined as the absence of a growth inhibition zone around the well. Zones of inhibition were measured and recorded.

To further determine whether the selected pathogens were inhibited or killed by probiotics, the growth inhibition zone was swabbed. The swab was then inoculated into BHI broths and incubated aerobically under 37°C for 24h. The BHI broths were then checked for growth. The

presence of growth in the broth was interpreted as an inhibitory property in the agar plate to be as a result of the bacteriostatic effect, while no growth was interpreted to be as a result of the bactericidal effect. Each of the tests in the determination of antibacterial effects of the probiotics was conducted in two trials, and in duplicate as control without using the pathogenic bacteria **Chuayana et al. (2003)**; **Millette et al. (2006)** and **Lim & Dond-Soon, (2009)**.

Statistical analysis:

Analysis of data was performed according statistical programmed of **SPSS (1997)**.

RESULTS AND DISCUSSION

Results listed in **table, (1)** show that out of examined 40 samples of fermented dairy products, 20 (100.00%) of probiotic yoghurts and 20 (100.00%) fermented milk drinks samples were contained with probiotics that isolated, identified and confirmed into three groups as *Lactobacillus* spp. (*L. casei*, *L. acidophilus*) and *Bifidobacterium* spp. (*B. lactis*) as reported in table, (2). Nearly similar findings were reported by (**Tabasco et al. 2007**) and (**Dardir 2012**). Elsewhere, the presence of multiple and closely related species in these products made the differential enumeration of probiotic and yoghurt starter bacteria difficult due to similarity in growth requirements and covering biochemical outlines of the species.

Table, (1): Occurrence and frequency distribution of examined “fermented dairy products” samples (n=40):

Fermented dairy products type samples	No. of positive samples	Positive samples percentage	No. of groups of probiotics isolates
probiotic yoghurt (bio-yoghurt)	20.00	100.00%	3.00
fermented milk drink	20.00	100.00%	3.00

Data outlined in table, (2) showed the viable numbers of the LAB present in the examined samples of probiotic fermented dairy products were determined and given the mean count (Log cfu/ml) of total LAB in examined samples were 7.00 ± 0.70 and 6.00 ± 0.50 in probiotic yoghurts and fermented milk drinks respectively. To some extent higher findings were stated by (**Dardir 2012**). *Lactobacillus* spp. and *Bifidobacterium* spp. viable cells in adequate numbers, namely the ‘therapeutic minimum’ need to be consumed regularly for transferring ‘probiotic’ effects to consumers. However, the minimum amount of probiotics needed to obtain a clinical effect has not been established (**Roy 2005**). Consumption should be more than 100 g/daily of probiotic dairy products

containing more than 10^7 – 10^9 cfu/gL⁻¹ (Kurmann & Rasic 1991); (Rybka & Kailasapathy 1995); (Tamime et al. 1995) and (Shah 2000). Other authors stipulate more than 10^8 cfu/gL⁻¹ as satisfactory level (Davis et al. 1971); (Kailasapathy & Rybka 1997) and (Kim 1988). Recommended dosage of 10^9 – 10^{10} cfu/gL⁻¹ is considered a minimum for healthy enteric system (Sanders & Veld 1999).

Table, (2): Mean count, of Lactic acid bacteria (LAB) strains discovered from examined “fermented dairy products” samples (n=40):

Fermented dairy products type samples	LAB Mean count (Log cfu/ml±SD)	Groups of discovered probiotic LAB isolates
probiotic yoghurt (bio-yoghurt) (n=20)	7.00±0.70	Lactobacillus acidophilus Lactobacillus casei Bifidobacterium lactis
fermented milk drink (n=20)	6.00±0.50	Lactobacillus acidophilus Lactobacillus casei Bifidobacterium lactis

The results tabulated in **table, (3)** show that all probiotic strains isolated from the different fermented dairy products samples were able to prevent or inhibit the growth of the selected bacterial pathogens. Probiotics of yoghurts and fermented milk drinks were inhibited the growth of the pathogenic Escherichia coli selected against them giving a bacteriostatic effect with mean diameters of zones of inhibition $10.00±0.1$ mm and $11.33±0.55$ mm for yoghurts and fermented milk drink samples respectively. Variable records were stated by (Kaboosi 2011) and (Anshumala & Behera 2011).

Table, (3): Antibacterial effects and the mean diameters of zones of inhibition (in mm) of probiotics isolated from examined “fermented dairy products” samples against some common bacterial pathogens (n=40):

Fermented dairy products type samples	Salmonella typhimurium*	Staphylococcus aureus*	Escherichia coli*
probiotic yoghurt (bio-yoghurt) (n=20)	Bactericidal 22.00±0.35	Bactericidal 15.30±0.50	Bacteriostatic 10.00±0.01
fermented milk drink (n=20)	Bactericidal 26.00±2.00	Bactericidal 12.30±0.40	Bacteriostatic 11.33±0.55

*Clinical laboratory strains.

Probiotics were have bactericidal effect for Salmonella typhimurium and Staphylococcus aureus and showed zones of inhibition with mean diameters were strong $22.00±0.35$ mm and moderate $15.30±0.50$ mm for yoghurt samples respectively. Similar findings were

described by **Catherine & Cabrera (2011)**. While, showed zones of inhibition with mean diameters were strong 26.00 ± 2.00 mm and moderate 12.30 ± 0.40 mm for fermented milk drink samples respectively. These finding nearly coincides with that of **Thirabunyanon et al. (2009)**. Otherwise, lower finding was mentioned by **Assefa et al. (2008)**.

Antimicrobial activity of Probiotics may be due to a number of factors. The adhesion of LAB to host intestinal epithelium might result in the competitive or exclusion of adhesion of pathogenic bacteria (**Velraeds et al. 1996**). The mechanism of inhibition on the pathogen invasion might also be due to hindrance of human enterocytic pathogen receptors by whole cell Lactobacilli rather than to a specific blockade of receptors (**Coconnier et al. 1993**). On the other hand, several other mechanisms for LAB to antagonist some common bacterial pathogens have also been suggested, among these were: contribution to mucosal barrier function, modulation of the immune response, competition for substrates, co-aggregation with pathogens, decreasing of the luminal pH via the production of lactic acid and secretion of specific compounds such as bacteriocins (**Coconnier et al. 2000**).

Regarding the public health significance, some probiotics have been shown in preliminary research to possibly treat various forms of gastroenteritis **King et al. (2003)**. Recent studies have indicated that the antagonistic activities against intestinal pathogens are produced by antimicrobial substances from several probiotic strains **Thirabunyanon et al. (2009)**. They might reduce both the duration of illness and the frequency of stools **Allen et al. (2010)**.

Probiotics consumption has been promoted as an effective means of preventing infections as antibiotic-associated diarrhea (AAD). Studies to date have not been definitive, but there is some support in the literature for the possibility that consumption of probiotics reduces the incidence and severity of AAD as indicated in several meta-analyses **Cremonini et al. (2002)**; **D'Souza et al. (2002)**; **Szajewska & Mrukowicz (2005)**; **Mcfarland (2006)**; **Sazawal et al. (2006)** and **Szajewska et al. (2006)**. Potential efficacy of probiotic AAD prevention is dependent on the probiotic strain (s) used and on the dosage **Doron et al. (2008)** and **Surawicz (2008)**. Up to a 50% reduction of AAD occurrence has been found in preliminary studies **Sazawal et al. (2006)** where, no side-effects have been reported in any of these studies. A study showed that a commercially available probiotic drink containing Lactobacillus casei DN-114001 and yoghurt bacteria might reduce the incidence of AAD and Clostridium difficile-associated diarrhea **Hickson et al. (2007)**. Milk and milk products are usually associated with probiotic bacteria, which provide supplements in maintaining beneficial intestinal balance **Isolauri (2001)**.

Ingestion of certain active probiotic strains may help lactose intolerant individuals tolerate more lactose than they would otherwise have tolerated **Sanders (2000)**. Animal studies have demonstrated the efficacy of some strains of Lactic acid bacteria (LAB) to be able to lower serum cholesterol levels, probably by breaking down bile in gut, thus inhibiting its reabsorption **Sanders (2000)**. Although, not a confirmed effect, some studies have indicated that consumption of milk fermented with various strains of LAB may result in modest reductions in blood pressure, an effect possibly related to the angiotensin-converting enzyme, inhibitor-like peptides produced during fermentation is involved in the regulation of blood pressure **Sanders (2000)** and **Sarkar (2003)**.

A study showed that lactic acid bacteria isolated from dairy products suppress the growth of *Salmonella typhimurium* and *Salmonella enteritidis*, the main cause of *Salmonella* food infection **Thirabunyanon et al. (2009)**.

A novel approach to controlling *Listeria monocytogenes* in food is the use of antimicrobial compounds from LAB **Vugst & Leroy (2007)** and **Singh & Prakash (2009)**

In laboratory investigations, some strains of LAB (*Lactobacillus delbrueckii* subsp. *bulgaricus*) have demonstrated anti-mutagenic effects thought to be due to their ability to prevent DNA damage, which in effect prevents tumors **Wollowski et al. (1999)** and also, bind with heterocyclic amines, which are carcinogenic substances **Wollowski et al. (2001)**. Some human trials hypothesize that the strains tested may exert anti-carcinogenic effects by decreasing the activity of β -glucuronidase **Brady et al. (2000)** and **Paul & Somkuti (2010)**. **Kailasapathy & Rybka (1997)** reported on several animal studies confirming that the intake of yoghurts and fermented milks, containing probiotic bacteria inhibited tumor formation and proliferation.

Some strains of LAB may affect pathogens by means of competitive inhibition and there is evidence to suggest that they may improve immune function by increasing the number of IgA-producing plasma cells, increasing or improving phagocytosis, as well as increasing the proportion of T lymphocytes and natural killer cells **Quwehand et al. (2002)** and **Reid et al. (2003)**. Specific rotavirus IgA was shown to be produced when certain probiotics were ingested **Kaila et al. (1992)**, viable probiotics were more efficient in inducing these responses than inactivated probiotics **Kaila et al. (1995)**. How probiotics may influence the immune system remains unclear, but a potential mechanism under research concerns the response of T lymphocytes to pro-inflammatory stimuli **Braat et al. (2004)**.

Some strains of LAB may affect *Helicobacter pylori* infections which may cause peptic ulcers in adults when used in combination with

standard medical treatments, but there is no standard in medical practice or regulatory approval for such treatment **Cats et al. (2003)**; **Hamilton (2003)**; **Mylyluoma et al. (2007)** and **Lin et al. (2009)**. Several in vitro and in vivo experiments on antagonism of different Lactobacillus strains against Helicobacter pylori and Clostridium difficile, Campylobacter jejuni, E. coli were performed by **Nowroozi et al. (2004)**. All tested human Lactobacillus strains were able to inhibit the growth of all strains of anaerobic human gastrointestinal pathogens **Strus et al. (2001)**.

Evidence on beneficial preventive effect of traveler's diarrhea by probiotics is inconsistent. A few studies have reported a prophylactic effect with Lactobacillus GG **Hilton et al. (1997)** and **Oksanen et al. (1990)** and Saccharomyces boulardii **Kollaritsch et al. (1993)**.

In one study, a commercial strain of Bifidobacterium infantis improved some symptoms of irritable bowel syndrome (IBS) in women **Whorwell et al. (2006)**. A separate study showed that a strain of Lactobacillus plantarum may also be effective in reducing IBS symptoms **Niedzielin et al. (2001)**. Other study focused on Bifidobacterium animalis showed a reduction in discomfort and bloating in individuals with constipation-predominant IBS, as well as helping to normalize stool frequency in said individuals **Guyonnet et al. (2007)**.

All patients responded to standard antimicrobial therapy, Saccharomyces boulardii has been used without complications to treat chronic diarrhea in AIDS patients **Saint-Marc et al. (1991)** and **Elmer et al. (1995)** while, Lactobacillus reuteri has been safely given to HIV-infected patients **Wolf et al. (1998)**.

Several clinical studies suggest that specially selected Lactobacillus strains reduced the frequency of recurrent urinary-tract infections (UTIs) in a group of high-risk women **Bruce & Reid (1988)**; **Reid et al. (1992)** and **Reid et al. (1995)** and an effective means of preventing genitourinary infections **Falagas et al. (2007)**. Several clinical studies suggest that using Lactobacillus acidophilus vaginal suppositories can help treat bacterial vaginosis **Martinez et al. (2009)**. Lactobacilli can also be used to restore particular physiological balance and protect the vaginal eco-system from Gram-negative and Gram-positive bacteria **Pascual et al. (2008)** and **Reid et al. (2009)**.

Lactobacillus paracasei could act as a potential barrier to prevent Staphylococcus aureus- associated injury and might exert its effect on the Staphylococcal enterotoxins or their target **Bendali et al. (2011)**.

Recent clinical trials on the use of probiotic as a potential and clinically applicable anti-caries measure was performed in preschool children and elderly demonstrated prevented fractions of between 21% and 75% following regular intakes of milk supplemented with Lactobacillus rhamnosus where, no adverse effects or potential risks

were reported **Twetman & Keller (2012)** and the effect of daily intake of milk supplemented with fluoride and/or probiotic bacteria on primary root caries lesions may reverse soft and leathery in older adults **Petersson et al. (2011)**. The single study carried out in early childhood reported a significant caries reduction in 3- to 4-year-old children after 7 months of daily consumption of probiotic milk **Twetman & Stecksén-Blicks (2008)**. Probiotics have been proven effective for preventing caries, lactic acid bacteria show promising properties to be used as potential probiotics for improving oral health **Bosch et al. (2011)**. Studies show probiotics can allow beneficial Lactobacilli to populate sites on teeth, preventing Streptococci pathogens from taking hold and inducing dental decay **Makarova et al. (2006)**.

From the point of view of risks, probiotic that offer a health benefit to their host indicates that consuming these microbes in foods or supplements would have few risks. Although, the available probiotic microorganisms are considered nonpathogenic, nontoxigenic, but even benign microorganisms. To date, there have been only isolated reports linking probiotics with adverse effects. Caution should be implemented when administering probiotic supplements to immunocompromised individuals or patients who have a compromised intestinal barrier, ulcerative colitis or other forms of intestinal damage or who are already very ill **Sanders (2008)**.

Cases of lactobacillemia have been reported in patients with severe underlying conditions **Horwitch et al. (1995)** and **Saxelin et al. (1995)**. A case of a liver abscess was associated with *Lactobacillus rhamnosus* in a 74-year-old diabetic **Rautio et al. (1999)** and cases of fungemia involving *Saccharomyces boulardii* have been reported **Pletincx et al. (1995)** and **Rautio et al. (1999)**. In a therapeutic clinical trial conducted by the Dutch Pancreatitis Study Group, the consumption of a mixture of six probiotic bacteria increased the death rate of patients with predicted severe acute pancreatitis **Besselink et al. (2008)**.

Lower rates of colon cancer among higher consumers of fermented dairy products have been observed in one population study, but confirmation of such an effect does not occur **Sarkar (2003)**.

Studies indicate that probiotic products as yogurts and fermented milk drinks could be a cause for obesity trends. However, this is contested as the link to obesity and other health related issues with yogurt may link to its dairy and calorie attributes **Ehrlich (2009)** and **Raoult (2009-a & 2009-b)**.

CONCLUSION AND RECOMMENDATION

Recently, many studies are now in progress on the applicability of probiotic bacteria as an alternative biotherapeutic treatment for and

protection against pathogenic infections. Although, probiotics have an excellent overall safety record, they should be used with caution in certain patient groups particularly neonates born, prematurely or with immune deficiency. Therefore, clinical trial results from one probiotic strain in one population cannot be automatically generalized to other strains or to different populations. Further, studies are needed to explore mechanistic issues and probiotic interactions. In view of the increasing use of probiotics as health supplements and therapeutic agents, clinicians need to be aware of the risks and benefits.

Government regulations differ among countries however, the status of probiotics as a component in food is currently not established on an international basis. For the most countries, probiotics come under food and dietary supplements because most are delivered by mouth as foods, whereas foods, feed additives and dietary supplements can only make general health claims. In only a few countries, regulatory procedures are in place or sufficiently developed to enable probiotic products to be allowed to describe specific health benefits. The Consultation Recommends that the Codex General Principles of Food Hygiene and Guidelines for Application of HACCP be followed CAC (1997).

In conclusion, present study showed that traditional dairy products of Najran region of Saudi Arabia can be used as a good source of potentially probiotic bacteria. Adequate scientific evidence exists to indicate that there is potential for the source of health benefits from consuming food containing probiotics. However, it was felt that additional research data are needed to confirm a number of these health benefits in humans.

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دراسة تأثير النشاط الميكروبي لبكتريا البروبيوتك المعزولة من منتجات الألبان المتواجدة في السوق المحلي على بعض الميكروبات المرضية الشائعة*

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المُلخَص العربي

نظراً للأهمية الغذائية والصحية للألبان ومنتجاتها أجريت هذه الدراسة للكشف عن تواجد
بكتيريا البروبيوتك في بعض منتجات الألبان في مدينة نجران بجنوب السعودية، وقد هدف
البحث إلى عزل وتصنيف بكتيريا البروبيوتك واختبار تأثيرها على بعض الميكروبات الممرضة
الشائعة، حيث دلت النتائج على عزل وتصنيف عدد ثلاث مجموعات من بكتيريا البروبيوتك
(بكتيريا حمض اللبن - Lactic Acid Bacteria)، وكانت كالتالي جنس *Lactobacillus*
spp. وتشمل *L. acidophilus* و *L. casi* والجنس *Bifidobacterium* spp. ويشمل *B. lactis*
من عينات الفحص وعددها ٤٠ عينة عبارة عن ٢٠ عينة من الزبادي و ٢٠ عينة من
اللبن جمعت من أماكن بيع الألبان ومنتجاتها المختلفة، وقد تم دراسة تأثير النشاط الميكروبي
لبكتيريا البروبيوتك على بعض أصناف من البكتيريا الممرضة والشائعة وهي *Salmonella*
typhimurium و *Staphylococcus aureus* و *Escherichia Coli*، حيث كان التأثير
قوي وقاتل ومانع لنمو البكتيريا الممرضة bactericidal للنوعين الأولين، بينما كان التأثير
مثبط ومحبط bacteriostatic للنوع الأخير وذلك على التوالي، هذا وقد تم مناقشة النتائج
والأهمية الصحية لبكتيريا البروبيوتك ومدى تأثيرها على صحة الإنسان.