Script

1. **Subject: FOOD PRESERVATION TECHNOLOGY**
2. **Paper - 00:**
3. **Paper Code:**
4. **Topic: Bio preservation and chemical preservatives**

PART – 1: MAIN SCRIPT

Dear Students, in to-day's lecture, let's make an attempt to know about the **Bio preservation and chemical preservatives.**

Introduction

Reservoir of enteric pathogens in the environment contributes to disease burden and mortality in population. Enteric pathogens are predominating in sewage manure, polluted water used for irrigation. It is universally accepted that there is no such thing as called "Zero risk" food. Consumers demand food products without preservatives having fresh quality even after storage. Food borne diseases are among the most serious public health concerns world wide. In spite of modern technologies, good manufacturing practices, quality control and hygiene and safety concepts such as risk assessment still food-borne illnesses increased over the past decade.

In recent years, there has been a growing interest in the use of natural antimicrobials, especially nisin. Generally they are active against food spoilage and food borne pathogenic microorganisms including *Bacillus cereus, Clostridium perfringens*,*Staphylococcus aureus*, and *Listeria monocytogenes*. Nisin is a ribosomally synthesized peptide that has broad-spectrum of antibacterial activity including many food-spoilage pathogens. Nisin works by attaching to the plasma membrane of target cells and create pores in the cytoplasmic membrane leading to cell lysis.

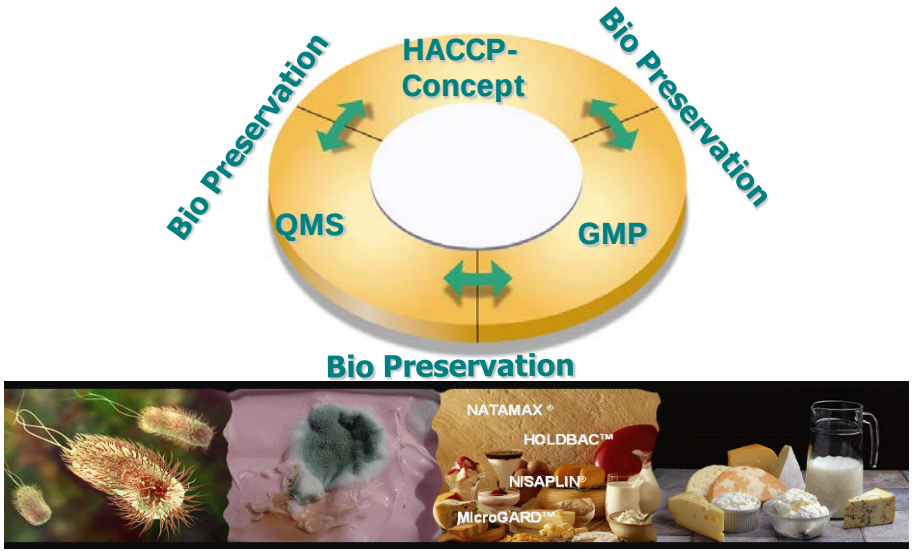
This episode deals with:

1. **Bio preservation and bacteriocins**
2. **Classification of bacteriocins**
3. **Nisin, Properties of Nisin and Applications of Nisin**
4. **Lactic acid bacteria and its application**
5. **Chemical preservation ,organic and inorganic**

1: Bio preservation and Bacteriocins:

Biopreservation is a natural and safe power tool to extended storage life and enhanced safety of foods using the natural antimicrobial compounds.

Gerenally theyt are of plant, animal and microbial origin and have been used in human food for long time, without any adverse effect on human health.



In ancient times, honey was used as a food preservative. It works as a preservative because the high concentration of sugar in honey and osmostic activity will takes place. This processes known as “osmosis,”. The process of osmosis destroys the cells by drying them up. The term bacteriocin is mostly used to describe the small, heat-stable cationic peptides synthesized by Gram positive bacteria, namely lactic acid bacteria.

The use of bacteriocins or bacteriocin-producing strains of LAB are of great interest as they are generally recognized as safe organisms and their antimicrobial products as biopreservatives. One of the most common forms of food bio preservation is fermentation a process based on the growth of micro organisms in foods, weather they are natural or added. These organisms mainly contain lactic acid bacteria which produced organic acids and other compounds that in addition to antimicrobial properties.

Bacteriocins:

Bacteriocins are antibacterial proteins produced by bacteria that kill or inhibit the growth of other bacteria. Many lactic acid bacteria (LAB) produce a high diversity of different bacteriocins. Though these bacteriocins are produced by LAB, found in numerous fermented and non-fermented foods, nisin is currently the only bacteriocin widely used as a food preservative. Bacteriocins are inhibit the growth of similar or closely related bacterial strain(s). They are typically considered to be narrow spectrum of antibiotics.

1. They are generally recognized as safe substances
2. They are not active and nontoxic to eukaryotic cells
3. They are usually pH and heat-tolerant
4. They have a relatively broad antimicrobial spectrum against many food borne pathogenic and spoilage bacteria.
5. They show a bactericidal mode of action on the bacterial cytoplasmic membrane.

Table .1 Bacterial genera that produces bacteriocin

Acetobacter	*Corynebacterium*	*Pediococcus*	*Shigella*
Actinobacillus	*Enterococcus*	*Psedomonas*	*Streptococcus*
Bacillus	*Erwinia*	*Salmonella*	*Propinobacterium*
Brevibaacterium	*Lactococcus*	*Serratia*	*Staphylococcus*
Closteridium	*Lactobacillus*	*Yersinia*	*Haemophilus*
Leuconostock	*Haloferax*	*Listeria*	

2: **Classification of bacteriocins**: Bacteriocins can be grouped based on their structure and on their mode of action. Class I and class IIa bacteriocins are usually very stable at acid-ic pH.

Methods of classification

Class I bacteriocins

Class II bacteriocins

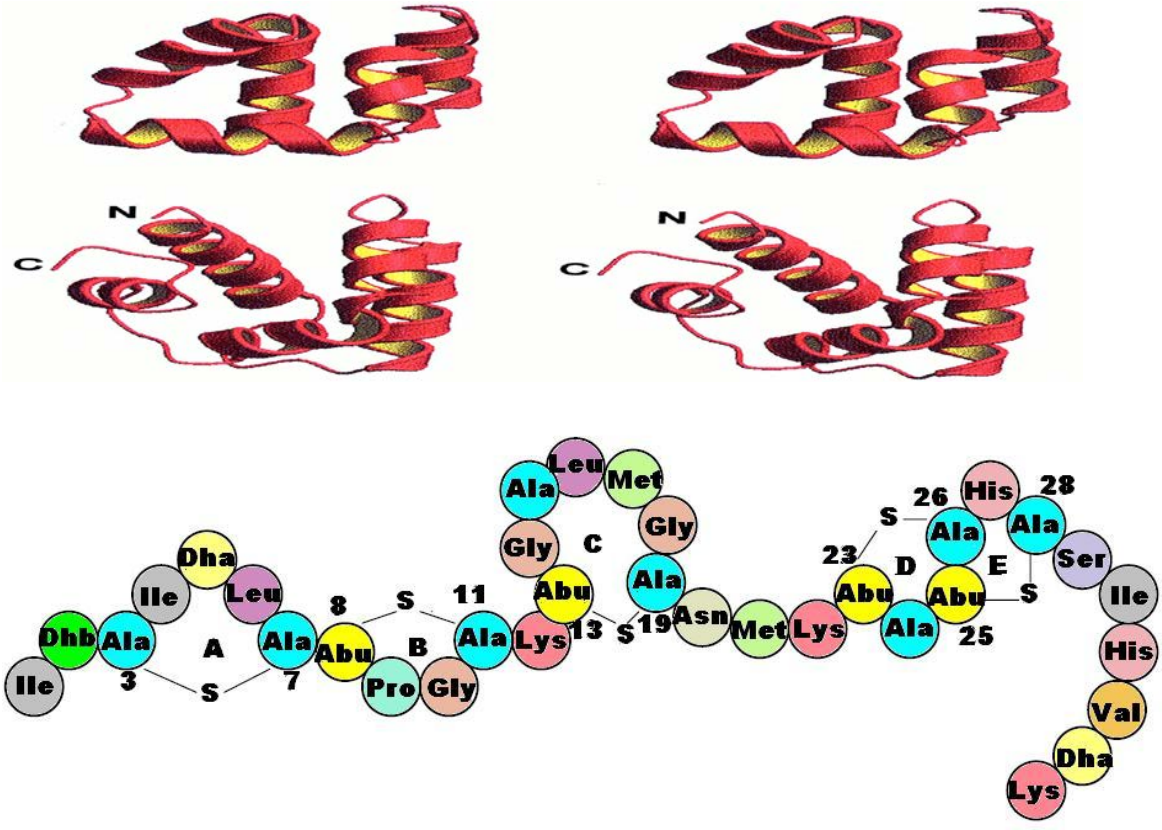
Class III bacteriocins

Databases

3: <u>Nisin and Properties of Nisin and Applications</u>

Nisin.

Nisin is a polycyclic anti bacterial peptide with 34 amino acid residues used as a food preservative and it contains the two uncommon amino acids. The unsaturated amino acids originate from serine and threonine.

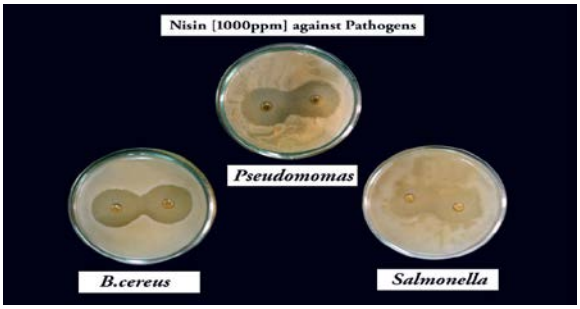


Appearance : It is a Free flowing white powder ,shelf life is 2 years at 4°C to 25°C.

Nisin is produced by fermentation ,using the bacterium *Lactococcus lactis*. In the food industry, it is obtained from the culturing of *L. lactis* on natural substrates, such as milk or dextrose and is not chemically synthesized. It was originally isolated in the late 1930s and produced since the 1950s as Nisaplin from naturally occurring sources by Aplin and Barrett company used as food additives in USA in late 1960s. Nisin was the first bacteriocin derived from fermentation of a lactic-acid bacterium and was approved by the FDA in April 1988 to use in prevention of *Botulinu*m spores in pasteurized process-cheese spreads.

Nisin is used in processed cheese, meats, beverages etc, during production to extend their shelf life by suppressing Gram-positive spoilage and pathogenic bacteria. In foods, it is common to use nisin at levels ranging from ~1-25ppm, depending on the food type and regulatory approval. As a food additive, nisin has an E number of E234. Since it is a natural antimicrobial peptide it has been approved as a natural food preservative by more than 50 countries and considered as generally recognized as safe (GRAS) by Food and Agriculture Organization (FAO)/World Health Organization (WHO) and the European Union.

Fig-Antimicrobial activity against pathogens.



Application of Bacteriocins

Bacteriocins are of interest in medicine because they are made by non-pathogenic bacteria that normally colonize the human body. Loss of these harmless bacteria following antibiotic use may allow opportunistic pathogenic bacteria to invade the human

body. They have shown distinct promise as a diagnostic agent for some cancers. Bacteriocins were tested as AIDS drugs around 1990, but did not progress beyond in-vitro tests on cell lines. Bacteriocins can target individual bacterial species, or provide broad-spectrum killing of many microbes.

Applications in meat products ie a bacteriocin-producing culture for sausage fermentation and/or biopreservation.

The well-studied bacteriocins in meat and meat products include nisin, enterocin AS-48, enterocins A and B and especially pediocin PA-l/AcH, alone or in combination with several physicochemical treatments, modified atmosphere packaging. Pediocin PA-l/AcH is more suitable for use in meat and meat products than nisin. Pediocin PA-1/AcH in salad and fruit juice and enterocin AS-48 against B. cereus in rice and vegetables as well as in fruit juices against other pathogens such as E. coli O157:H7, *S. aureus*, and the spoilage bacterium.

Nisin is non poisonous, natural and effective as antiseptic in food. It is used in powdered form which is grayish white or yellowish in color. It remains stable under the condition of acid-heating and at room temperature. Stability of Nisin mainly depends upon three factors:

1. Storage length
2. Incubation temperature
3. pH

Its antimicrobial spectrum reveals that Nisin can stop or kill G+ that can turn the food putrefactive. It works well against bacterial spore that display tolerance towards *Bacillus acidophilus*. Nisin preservative also inhibits various other bacteria such as *Bacillus cereus*, *Clostridium botulinum*, *Bacillus stearoacidophiles*, and *Listeria monocytogenes.* The nisin-Lipid II complex penetrates the cell wall and breaks into the cytoplasmic membrane and form pores. Through these pores the internal cytoplasmic content oozes out resulting in bacterial inhibition or death. It does not make any changes in the working of normal community of bacteria in the intestine neither it causes drug resistance to antibiotic substances. It also has not shown resistance to antibiotic drugs.

Applications of Nisin:

Nisin is popularly used in dairy products, milk, aquatic products, poultry, meat products, plant protein, fast food, baking food, fruit juice, canned food, cosmetics, beverage drinks, health care products and medicines. It helps by decreasing the time taken for sterilization, lowers temperature during food sterilization, improves food quality, lessens damage to nutrition, and prolongs storage time of food material. For regular use 5% of Nisin solution made by dissolving in pure water is mixed in equal amount of food by using grading extension technique. Sometimes, other types of antiseptics are also used in combination with Nisin**.**

Nisin for meat sausage ham

fish

sausage

Food categories in which Nisin is used?

There are various food categories in which Nisin is used:

Dairy products: In this category, Nisin is used as preservative for increasing the life of yoghurt, natural cheese and processed cheese. The further categories of processed cheese including block cheese, spreads, dips, sauces and slices contain varying amount of moisture content. These contain emulsifying salts and are processed by heating. Processed cheese also comes with additives and flavors such as shellfish, herbs, fish etc. Flavored processed cheese requires use of **Nisin** in different measure for extending the shelf life.

Egg products: Egg products like yellow, whole or white are treated to heat temperature for kill *Salmonella* spp.

Pasteurized soups: There are many heat resistant bacteria that do not get eliminated during pasteurization. Hence Nisin is used in pasteurized soups for preventing or delaying the psychroduric activities of food spoiling bacteria.

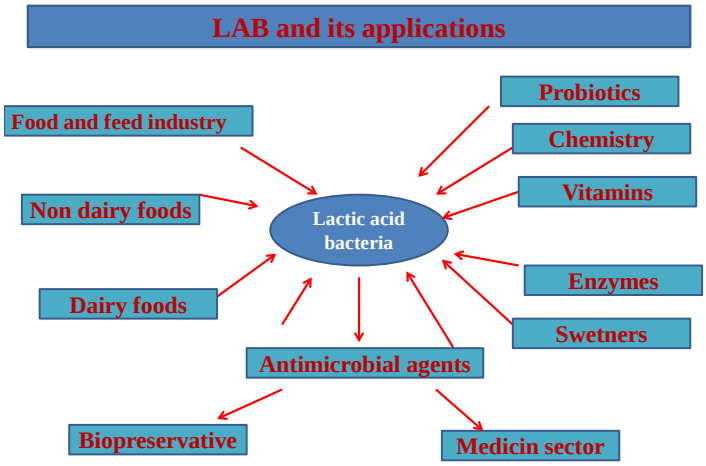
Flour based products: Flour based product do not have long shelf life, even though it is toasted before serving, heat resistant bacteria do not get killed. There have been instances of food poisoning due to crumpets in major part of Australia. So as to stop the growth of *B.cerus* in crumpets or any other flour based products, Australian and New Zealand government has approved to use **of** Nisin and unlimited.

Canned food: Use of Nisin for preventing thermophilic spoilage has been made mandatory in most of the countries. Although low acid canned foods are exposed to heat during packing, so as to guarantee their shelf life at normal temperature use of Nisin is permitted. Since thermophilic spores are very sensitive to the combined treatment of Nisin and heat, this preservative is used preferably with heat treatment. Some of the canned foods that contain Nisin are okra, baby sweet corn, asparagus, potatoes, peppers, carrots, peas, and mushrooms. For canned foods with high acid product like tomato, more amount of Nisin is used.

Meat products: Earlier Nitrite was used as preservative in meat products but at present it has been partially replaced by nisin. It is used in cooked sausages and low fat sausages.

- **Seafoods**: **Nisin** is used more in smoked Salmon as it acts as a highly effective anti-listerial agent, especially when the packing is done in a carbon dioxide atmosphere.
- **Salad dressings**: In salad dressings, the acidity level is reduced to retain the flavor during cold dressing. But this reduction in acidity leads to growth of bacteria. This spoilage is controlled by using Nisin.
- **Alcoholic beverages**: Lactic acid bacteria that are acid tolerant have a tendency to spoil wine and beer. Since yeast does not get affected by Nisin, it is usually added during the fermentation process. **Nisin** is further added to fermenters for preventing contamination in unpasteurized beers for increasing shelf life.

4: Fig: Lactic acid bacteria and its **application**



Lactic acid bacteria:

Lactic acid bacteria refer to a large group of beneficial bacteria that have similar properties and all produce lactic acid as an end product of the fermentation process. They are widespread in nature and are also found in our digestive systems. Although they are best known for their role in the preparation of fermented dairy products, they are also used for pickling of vegetables, baking, and winemaking, curing fish, meats and sausages. Additional characteristic flavours and aromas are often the result of other products of lactic acid bacteria

The Production of Lactic acid:

Lactococcus lactis, originated from the lactic industrial products have been still, extensively studied for their commercial potential, but that strain isolated from plants has been given less attention in sugar mills. *Lactococcus lactis*, like many other lactic acid bacteria, are involved in producing lactic acid.

Application of Lactobacilli in Dairy Industry:

Lactic Acid Bacteria (LAB) especially Lactobacilli are responsible for the formation of the micro flora of most dairy products especially of cheese and fermented milk. Lactobacilli are important for flavor, color and texture of dairy products through acidification due to lactic acid and of the metabolism of milk proteins. The most commonly used species in dairy products are *L.casei, L. helveticus, L. rhamnosus, L. lactis, L. curvatus* and *L. plantarum*. Furthermore, Lactobacilli are incorporated into yogurt, cheese and fermented milk as probiotics due to their

beneficial effect especially on acute and chronic inflammations of the gastrointestinal tract. Addition, due to the production of bacteriocins Lactobacilli also help on the preservation of dairy products.

Application of Lactobacilli on Wine Industry:

Lactobacilli are also applied in wine industry both for grape and fruit wines, such as cider. The organic acids existing in wine which are mainly malic and tartaric acid can be easily metabolized by Lactobacilli. Malic acid is converted to lactic acid and carbon dioxide, this phenomenon is called manolactic fermentation which is extensively used for fruit wines maturation. Though tartaric acid is decomposed into pyruvic and citric acid complete spoilage of the selected food product occurs. So the appropriate choice of the fermenting Lactobacilli is necessary. Usually decomposition of tartrate is observed by *Lactobacillus plantarum* and *Lactobacillus brevis.*

5: Chemical preservation ,organic and inorganic

Chemical preservation

Chemical food preservatives are substances which, under certain conditions, either delay the growth of microorganisms without necessarily destroying them or preventing the growth. It includes some natural food constituents which, when added to foods, it prevent the growth of microorganisms. Sugar is used partly for this purpose in making jams, jellies, and marmalades and in candying fruit. The use of vinegar and salt in pickling and of alcohol in brandying also falls in this category. Some chemicals foreign to foods are added to prevent the

growth of microorganisms. The latter group includes some natural food constituents such as ascorbic acid (vitamin C), which is added to frozen peaches to prevent browning, and a long list of chemical compounds foreign to foods and classified as antioxidants, bleaching agents, acidulates, neutralizers and stabilizers ect.

Organic chemical preservatives

Sodium benzoate and other benzoates are among the principal chemical preservatives. The use of benzoates in certain products in prescribed quantity (usually not exceeding 0.1 percent) is permitted in most countries, some of which require a declaration of its use on the label of the food container. Since free benzoic acid actually is the active agent, benzoates must be used in an acid medium in order to be effective. The ability of cranberries to resist rapid deterioration is attributed to their high benzoic acid content. Benzoic acid is more effective against yeasts than against molds and bacteria. Other organic compounds used as preservatives include vanillic acid esters, propionates, sorbic acid and glycols, ect.

Inorganic chemical preservatives

Sulfur dioxide and sulfites are perhaps the most important inorganic chemical preservatives. Sulfites are more effective against molds than against yeasts and are widely used in the preservation of fruits and vegetables. Sulfur compounds are extensively used in wine making and, as in most other instances when this preservative is used, much care has to be exercised to keep the concentrations low in order to avoid undesirable effects on flavor. Oxidizing agents such as nitrates and nitrites are commonly used in the curing of meats.

Sodium Benzoate & Benzoic Acid:

The two are related because sodium benzoate produces benzoic acid once dissolves in water it has got anti-microbial properties. Most effective on low pH, below 4.5. Sodium Benzoate is used in fruit products, beverages, dressings, salads, & pastries fillings, icing, olives, ect. It works against yeast, molds and some bacteria. Low levels were used to avoid off-flavor and maximum level allowed by Law is 0.1%.

POTASSIUM SORBATE

Sorbic family : Potassium Sorbate, Sodium Sorbate, Calcium Sorbate.

Potasium sorbate will produce Sorbic Acid once its dissolves in water and it is widely used preservative in the world. Effective against yeast, molds and bacteria. Effective up to pH 6.5, maximum level allowable by law is 0.1%. In many food products, Sorbate & Benzoates are used together for greater protection against wider variety of microorganisms. Benzoate effective only below pH 4.5, but Sorbate is effective even with pH 6.5. If preservatives are used in foods, must be declared on the list of ingredients on the label along with short explanation of its use.

Pickled meat

Meat may be preserved by dry curing or with a pickling solution. The ingredients used in curing and pickling are sodium nitrate, sodium nitrite, sodium chloride, sugar and citric acid or vinegar.

Conclusion:

Bio preservation refers to extended storage life and enhanced safety of foods using the natural microflora and (or) their antibacterial products. Lactic acid bacteria have a major potential for use in biopreservation because they are safe to consume and during storage they naturally dominate the microflora of many foods.

Lactic acid bacteria are therefore excellent ambassadors for microbial world. They are not only of major economic significance, but are also of value in maintaining and promoting human health. Nisin is currently recognized as a safe food preservative in approximately 50 countries. Nisaplin is an extremely stable product. Biopresevation reduces the amount of chemical privatives as well as the intensity of heat treatments. Nisin re-mains the most commercially important bacteriocin, although other bacteriocins have been characterized and developed for possible approval and use.

The most common food-borne infections in the European Union (EU) are caused by bacteria, namely *Campylobacter, Salmonella, Listeria*, and viruses. They are reported to affect nearly 4 lacks of European Union citizens each year.

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