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Conventional Food Preservation Methods and their Biochemical Response

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Abstract

Preservatives prolong the shelf life of the foods. Low temperature, reduced water activity, food acidification, fermentation, addition of preservatives and modified atmospheric packaging are different methods of food preservation. Rapid cooling crystallizes the lipids of cell membrane system and cause the leakage of ions and cell components. Reduced water activity and low pH were respectively harming the microbial metabolism and cell components. Bacteriocins interact with lipid II through cell wall components and hampering the cell wall formation. Glycolysis is affecting by benzoate and nitrites are producing lethal nitric oxide and peroxy nitrites. Sulphites reacting with disulphide bonds (S-S), thiol (S-H) groups of microbial proteins / enzymes and causing several conformational changes in proteins. High CO_2 also affecting the microbial metabolism.

Introduction

Food preservation is a phenomenon of preventing the undesirable changes in foods during storage and transportation. All foods deteriorate in quality following harvest, slaughter or manufacture, in a manner that is dependent on food type and composition, formulation (of manufactured foods) and storage conditions (Gould et al 2000). Natural or synthetic preservatives added to foods to prolong the shelf life of food. The importance of natural preservative compounds is increasing due to the more extensive use than synthetic compounds. Low temperature, reduced water activity, food acidification, fermentation, addition of preservatives and modified atmospheric packaging are different methods of food preservation and their mechanism of inhibiting the microorganisms discussed in this mini project.

Low temperature storage is widely used for minimizing the disease development. It protects the food samples from physical, chemical change (Yang and Liu, 2019), reducing the chilling injury and ion leakage (Jin et al 2015). The quantity of E.coli bacteria was been reduced in milk without significantly affecting pH or color properties (Gurol et al 2012). Water is a most important factor governing microbial spoilage of foods. Heating, freeze drying, freeze concentration, and osmotic concentration methods are used to reduce water content of foods. Measured water activity values have correlation well with their potential growth and metabolic activity (Chirife et al 1996).

Many foods have been traditionally processing by the acidification which imparts special flavor for the consumer needs. Acidic foods are the foods that containing acidic ingredients for producing a final equilibrium pH of 4.6 or below. Acidification is a one of the limiting factors for the growth of pathogenic microorganisms. It's a prominent way of controlling microorganism when is resistance to pasteurization and cooking (cals.cornell.edu).

Low temperature

Freezing halts the activities of spoilage microorganisms in and on foods and can preserve some microorganisms for long periods of time (Archer, 2004). Preservation of food by freezing is based on the retardation of microbial growth to the point where decomposition due to microbial action does not occur (Gunderson and Peterson, 1977). Freezing causes the apparent death of 10 – 60% of microbial population, a percentage that gradually increases during frozen storage (Moharram and Rofael, 1993). Generally, microbiological spoilage could cease at temperatures between -9°C and -12°C but non-sporulating rods, spherical bacteria are resistant and no impact could be on bacterial spores.

Biochemical response

Rapid cooling from room temperature (RT) to -150^{o}C significantly crystallizes the lipids of cell membrane system **(**Fennema and Powrie, 1964) which leads to leakage of K^+ ions, galactosidase, low mw solutes, amino acids, RNA and strands of DNA (ss / ds). Alternately, increased extracellular solutes and its conversion to ice dehydrate the cells based on osmosis principle (Rahman and Velez-Ruiz, 2007).

Reducing water activity (a_w**)**

The water activity (a_w) of a food is the ratio between the vapor pressure of the food itself, when in a completely undisturbed balance with the surrounding air media, and the vapor pressure of distilled water under identical conditions (fda.gov). Low–water activity (a_w) foods and food ingredients are either naturally low in moisture or they are produced from high a_w foods that are deliberately dried. Simulation of drying process could be achieved by the addition of sugar, salt to the foods. The a_w of 0.62 is the minimum requirement for microbial growth. Molds, yeast and bacterial exhibit the growth by increasing the a_w value. Exceptionally xerophilic bacteria grow at a_w 0.62 and water activity value of 0.86 is most important because of its limiting the growth of *Staphylococcus aureus* (Roos, 2011). Many methods such as heating, freeze drying, freeze concentration, and osmotic concentration are used to reduce water activity of foods. Halophiles propagation occurs between 0.80 – 0.75 a_w value. Most bacilli, cocci and lactobacilli propagate between 0.95 – 0.91 a_w value. Most of yeast and molds grow at an a_w value of respectively 0.86 – 0.80 and 0.91 – 0.88 (Erkmen and Faruk, 2016).

Biochemical response

Reduced water activity has sublethal and lethal injuries on microorganisms. It prevents the vegetative growth of microbes, spore germination and toxin production in molds, bacteria. Exposure of *E. coli* O175:H7 to combined cold and water activity stresses resulted in adaptive weakness, disruption of energy generation processes, apparent DNA damage, the down regulation of molecular chaperones and proteins associated with responses to oxidative damage (King et al 2016). Microorganisms cannot grow and divide when desiccated, but can survive for certain periods of time, depending on their features. Desiccation, thermal treatments strictly control the energy metabolism, rate of replication, and protein synthesis in the *Salmonella enterica* serovar Typhimurium strain. Evidently these stress events increased the levels of ribosomal proteins (Maserati et al 2018). Desiccation stress elevates the gene expression of a DNA polymerase V subunit UmuC, which participate in DNA damage control and translesion DNA synthesis (Reuven et al 1999). The genes responsible for adaptive conditions to desiccation were upregulated and their upregulation is increasing in high fat and protein content of the diet (Crucello et al 2019).

Dehydration is oxidizing the yeast cells (Pereira, Panek and Eleutherio, 2003). In the *S. cerevisiae,* dehydration decreased the levels of plasma membrane protein- Agt1 or α-glucoside transporter (Agt1) which transports a wide range of α-glucosides such as maltose, sucrose, trehalose, isomaltose and palatinose (Kulikova-Borovikova et al 2018). Sodium chloride mediated osmosis significantly reduced a_w value and reduced the radial growth rates in *Paecilomyces niveus, Penicillium brevicompactum, Penicillium expansum* and *Penicillium roqueforti* and also delayed conidial germination in *P. expansum* (Van Long et al 2017).

Acidification

Acidification is a process of acidifying the food products to a final pH (4.6 or below) for the production of acidified food products (Ekanayake and John Kester, 2013). This natural phenomenon imparts organic acids into the food. Acid in the food slow down or prevent the disease or spoilage causing organism’s growth and prolong the shelf life. Foods without adequate acidity may allow the growth of micro- organisms and foodborne illness (Rushing and Curtis, 1993). Adding organic acids and other acids could also create an artificial acidification to the foods by adjusting the pH, improving the flavor (Booth and Stratford, 2003), and controlling *L. monocytogenes* in the small-scale cheese production (Brown et al 2018). Combination of acidification, chemical preservatives and citric acid significantly inhibiting the total viable count, yeasts, and molds (Musyoka et al 2018).

Biochemical response

Food acidification increases the internal pH of MO's as part of regulatory mechanisms. Bacterial species increase the internal pH in response to external acidification (Shioi et al 1980; Matin et al 1982; Krulwich et al 2011). Low pH damages the essential components of the cell. Acidification of foods promotes the acidification of periplasm which is an insignificant barrier for proton movement and also possesses limited buffering capacity. Non-ionized molecules cross the inner membrane and dissociates in the cytoplasm. Protons enter into the cytoplasm using the protein channels buried in the cell membrane and damage the membrane (Deamer, 1987; Swanson & Simons, 2009). High acid stress (acidification) fails the inducible mechanisms of survival and causing the bacterial viability very low (Richard & Foster, 2004; Lund et al 2014).

Fermentation

Fermentation is a technology, most ancient and economic method of food preparation in which the growth and metabolic activities of microorganisms are used to preserve foods (Wilburn and Ryan 2017). The positive effects of fermentation exerted by a combination of the live microorganisms present in the fermented food and bioactive components released into the foods as by-products of fermentation (Mathur, Beresford and Cotter, 2020). Microorganisms break down fermentable substrates into organic acids and alcohol and bacteriocins (Kim et al 2016). Fermentation increases the shelf life, organoleptic properties, digestibility of carbohydrates, proteins and bioavailability of vitamins and minerals (Altay et al 2013; Hwang et al 2017). The organisms such as *Saccharomyces cerevisiae,* Lactic acid bacteria (LAB) are been found crucial for the taste, texture and smell of fermented products. The baker's yeast is an indispensable for baking bread and brewing beer and LABs were the key bacteria to ferment cheeses, yoghurts and vegetables (Gaenzle, 2015; Rebbeca et al 2017; Wuyts et al 2020).

Varieties of microorganisms are affecting the sensory changes by using the food as carbon and energy source. Hence a spoiling food had a succession of different populations that may increase in number or down based on nutrients availability. Lactic acid bacteria and molds secrete chemicals that inhibit the competitors (Gram et al 2002). Lactic acid bacteria (LAB) produce an array of active antimicrobial substances like organic acids, hydrogen peroxide, bacteriocins etc (Todorov et al 2012). Bacteriocins are ribosomally-synthesized secreted anti-microbial peptides capable of inhibiting both food spoilage/pathogenic bacteria from both Gram-negative and Gram-positive group (Suganthi and Mohanasrinivasan, 2015). Nisin and AMP producing *Lactococcus lactis* are FDA approved safe bacteriocins. Nisin is found to inhibit the germination of *C. botulinum* spores in cheese and spreads among other foods (Mazzotta et al 1997).

Nisin antimicrobial action

Nisin does not require the membrane receptor on its target cells. Nisin interact with cell wall components such as s teichoic acids, teichuronic acids and lipoteichoic acids, acidic polysaccharides or phospholipids by hydrophobic or electrostatic interactions. Subsequently nisin interact with lipid II which is essential for bacterial cell wall biosynthesis and prevents peptidoglycan monomer to incorporate into the growing peptidoglycan network. Broadly nisin interfere the growth of peptidoglycan network (Kramer et al 2008; Punyauppa-path et al 2015).

Addition of preservatives

Benzoic acid

Berries and some fruits contain high levels of secondary metabolite, benzoic acid. Benzoic acid and its salt are widely used as preservatives in foods, beverages, toothpastes, mouthwashes, dentifrices, cosmetics and pharmaceuticals. Benzoic inhibits the growth of yeast, mold and bacteria by perturbing the cell membranes (Hazan et al 2004). Benzoate enters into the yeast cells which shift the internal pH and cause to restrict the cell growth by accumulating the hexose monophosphates levels and preventing glycolysis and ATP formation (Krebs et al 1983).

Nitrites

Nitrite is a food additive in meat, fish and cheese products. It is an effective growth inhibitor for *Clostridium botulinum* (Dahle, 1979). Nitrate has the same function as nitrite which forms reactive nitrite on long periods of time. Nitrates enter into the cell through facilitated protein transporters across the bacterial membrane (Goddard et al 2017). Bacteria excrete nitrite into the cytoplasm equivalent to the nitrate levels. Nitrites further degrade to nitric oxide ($NO^{\cdot}$) which converts to peroxynitrite ($ONOO^{-}$) by hydrogen peroxide in the bacteria and also deoxymyoglobin of the meat performs nitric oxide formation (jia et al 2009; Gladwin and Kim-Shapiro, 2008). Bacterial phagosomes convert nitric oxide to peroxynitrite (ONOO–) further kills the bacteria (Majou and Christieans, 2018).

Sulphites

Sulphites are a group of chemical compounds apply to the foods as sulphur dioxide gas, hydrogen sulphites, metabisulphites and salt forms with potassium, calcium or sodium. These sulfite forms are use as additives in beverages such as beer, wine, juices, dried fruits, processed fish, seafood, meats and some canned goods. Sulphites prevent the oxidation and bacterial growth is also found in some fermented foods as metabolites of yeast (Carocho et al 2014; Irwin et al 2017). Sulphites effect on microorganisms is dose dependent and may control the growth or completely kill the organism. Sulphites passively pass through the bacterial cell membrane and react with disulphide bonds (S-S), thiol (S-H) groups of microbial proteins / enzymes and causing several conformational changes in proteins. Moreover, sulphites degrade the essential components of cofactors, coenzymes and prosthetic groups. This causes global inactivation of respiratory enzymes and normal metabolism with partial or complete loss of activity (Schimz, 1980; Simon and Kroneck, 2013; D'Amore et al 2020).

CO_2 enriched modified atmospheric packaging

Surrounding a food with CO_2 enriched modified atmospheric packaging reducing the rate of microbial spoilage. High CO_2 levels inhibiting the gram negative psychrotrophs largely and least on gram positive psychrotrophs. Displacement of O_2, lowering the pH of the food medium by forming carbonic acid and direct effects on microbial metabolism are different methods of microbial control mechanisms by CO_2 enriched modified atmospheric packaging (Devlieghere and Debevere 2000; Hotchkiss, Werner and Lee, 2006).

Conclusion

Food preservation prevents the undesirable changes in stored foods and extends the shelf life. All of the different food preservation methods invented have the capabilities to control or kill the pathogenic organism complete. Preservation methods were affecting the microbial cell components, cell wall formation, transport system, ionic equilibrium, microbial metabolism and tertiary structure of proteins. Effectively all of the discussed preservation methods have significant impact in controlling bacteria, molds and yeast type of microorganisms.

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