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REPORT

Preservation: A Preventive Measure of Food Spoilage

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Food preservation is an important aspect in protecting food and food products from spoilage for domestic use as well as for food manufacturers.

Preservative means a substance which when added to food is capable of inhibiting, retarding or arresting the process of fermentation, acidification or other decomposition of food.

In most cases microorganisms use our food supply as a source of nutrients for their growth. By multiplying their number they deteriorate our food and render it unfit for human consumption. The interaction between microorganisms, plants and animals is natural and constant.

Microorganisms have an absolute demand for water because without water (moisture) no growth can occur. The water requirement is best expressed in terms of available water or water activity also. Moisture content of food is of great importance. Water occurs in foods in two forms: as bound water and free water as available.

Bound water includes water molecules chemically bonded whereas free water is not physically linked to the food matrix and is easily lost on evaporation or drying. Since most foods are a heterogeneous mixture of substances, they may contain varying proportions of the two types of moistures.

The regulation of water as an ingredient of food was the earliest form of food preservation. The availability of water for microbiological growth and biochemical reaction can be controlled by dehydration, freezing or addition of salt and sugar and chemical preservatives. Water activity is a measure of the availability of liquid water and is defined as the ratio of the equilibrium vapour pressure of the sample (P) to the equilibrium vapour pressure of pure water (P_0). At the same temperature, $a_w = P/P_0$. Equilibrium relative humidity (ERH) refers to the atmosphere surrounding the food and is equal to $100 \times a_w$ where a_w refers to the activity of water in solid or liquid foods. Dried foods having $a_w < 0.6$ are not generally at risk of spoilage by the growth of microorganisms.

Approximate threshhold values for microbiological growth in food are shown in Table-1.

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TABLE-1

Species	a_w
Bacteria	0.91
Yeast	0.85
Molds and fungi	0.75
Xerophilic molds and fungi	0.65
Osmophilic yeast	0.60

Spores have much greater tolerance to low a_w.

Growth Curve of Microbial Cultures: Whenever microorganisms are added to a food and conditions are favourable, the organisms will begin to multiply and will pass through a succession of phases. When counts of organisms are made periodically and the results are plotted with logarithms of numbers of organisms per mL as ordinates and time units as abscissae, a growth curve is obtained, as shown in Figure. 1. This curve is ordinarily divided into two phases as indicated in the figure: (1) the initial lag phase (A to B), during which there is no growth or even a decline in numbers; (2) the phase of positive acceleration (B to C), during which the rate of growth is continuously increasing; (3) the logarithmic or exponential phase of growth (C to D), during which the rate of multiplication is most rapid and is constant; (4) the phase of negative acceleration (D to E), during which the rate of multiplication is decreasing; (5) the maximum stationary phase (E to F), where numbers remain constant, (6) the accelerated death phase (F to G); and (7) the death phase or phase of decline (G to H), during which numbers decrease. With many bacteria (or other microorganisms) the numbers do not decrease at a fixed rate to zero, as indicated by the unbroken line in the figure, but taper off vary gradually as low numbers are approached, as shown by the broken line, and a few viable cells remain for some time.

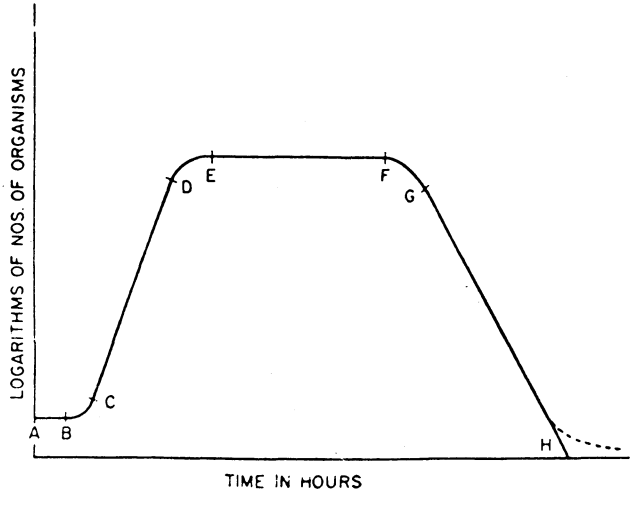

Fig. 1.

Applications to Food Preservation: Especially important in food preservation, (*i.e.*, preservation of spoilage) is the lengthening, as much as possible, of the lag phase and the phase of positive acceleration, often combined and called the lag phase. This can be accomplished in different ways.

In accomplishing the preservation of food by various methods, the following principles are involved:

(1) Prevention or delay of microbiological decomposition:
   (a) By keeping out microorganisms (asepsis)
   (b) Removal of microorganisms by filtration.
   (c) By hindering the growth and activity of microorganisms by low temperature.
   (d) By drying. This includes the tying up of water by solute.
   (e) Anaerobic conditions in sealed and evacuated container.
   (f) Using chemical preservatives.
   (g) By killing the microorganisms, *i.e.*, by heating or radiation.
   (h) Irradiation.

(2) Prevention or delay of self-decomposition of the food:
   (a) By destruction or inactivation of food enzymes that is by blanching.
   (b) Prevention of oxidation by means of antioxidants
   (c) Prevention of damage due to insects, etc.

Food additive is a substance or mixture of substances other than basic food stuff which is present in food. Those food additives which are specifically added to prevent the deterioration or decomposition of food have been referred to as chemical preservatives. These deteriorations may be caused by microorganisms, food enzymes or purely chemical reactions. The inhibition of the growth or chemical deterioration is one of the main purposes of the use of chemical preservatives. Preservatives may inhibit microorganisms by interfering with their cell membranes, enzymatic activities (enzymatic browning) or their genetic mechanisms.

Preservatives added to inhibit or to kill microorganisms may be classified on various other factors such as chemical composition, mode of action, specificity, effectiveness and legality. Preservatives like sugar are effective because of their physical action; sodium metabisulphite, benzoate etc. because of their chemical action; and sodium chloride because of a combination of these effects.

Antioxidants are added to oil and fats to delay the onset of oxidative rancidity, which is the chief cause of spoilage due to rancidity and liberation of free fatty acids. It is noted that antioxidants exhibit synergism, being more potent as mixture than when used individually. Antioxidants are phenolic compounds. Naturally occurring constituents of food such as tocopherols also have antioxidant properties. The definitions and list of permitted antioxidants with their permitted limits and other provisions under PFA Rules (1955) are shown in Table-2.

TABLE-2
PREVENTION OF FOOD ADULTERATION RULES

Definition of anti-oxidants: 'Anti-oxidant' means a substance which when added to food retards or prevents oxidative deterioration of food and does not include sugar, cereal oils, flours, herbs and spices.

Restriction on use of anti-oxidants: No anti-oxidant other than lecithin, ascorbic acid and tocopherol shall be added to any food:

Provided that the following anti-oxidants, not exceeding in concentration mentioned against each, may be added to edible oils and fats except ghee and butter, namely:

Antioxidant	%
1. Ethyl gallate	
2. Propyl gallate	
3. Octyl gallate or mixture thereof	0.01
4. Dodecyl gallate	
5. Ascorbyl palmitate	0.02
6. Butylated hydroxanisole (BHA)	0.02
7. Citric acid	
8. Tartaric acid	
9. Gallic acid	0.01
10. Resin guiaice	0.05
11. Tertiary butyl hydroquinone (TBHQ)	0.02

Provided that dry mixes of Rassgollas and Vadas may contain butylated hydroxyanisole (BHA) not exceeding 0.02 per cent calculated on the basis of fat content;

Provided further that anti-oxidants permitted in Rule 59 may be used in permitted flavouring agents in concentration not exceeding 0.01 per cent;

Provided further that wherever butylated hydroxyanisole (BHA) is used in conjunction with the antioxidants mentioned as item Nos. 1 to 4 of the preceding provision, the quantity of the mixture shall not exceed the limit of 0.02%;

Provided also that ghee and butter may contain butylated hydroxyanisole (BHA) in a concentration not exceeding 0.02 per cent;

Provided also that fat spread may contain butylated hydroxyanisole (BHA) or tertiary butyl hydroquinone (TBHQ) in a concentration not exceeding 0.02 per cent by weight on fat basis;

Provided further that ready-to-eat dry breakfast cereals may contain butylated hydroxyanisole (BHA) not exceeding 0.005% (50 ppm);

Provided also that in ready-to-drink infant milk substitute, lecithin and ascorbyl palmitate may be used up to a maximum limit of 0.5 g/100 mL and 1 mg/100 mL respectively.

Use of Anti-oxidants in Vitamin D preparations:

Vitamin D preparations may contain anti-oxidants prescribed in Rule 59 not exceeding 0.08%.

Each organism has its own optimal a_w and its own range of a_w for growth for a given set of environmental conditions. Factors affecting the moisture requirement of organisms are (a) the nutritive properties of the substrate, (b) its pH, (c) its content of inhibitory substances, (d) availability of free oxygen and (e) temperature. The range of a_w, permitting growth, is narrowed still more if two or more conditions are not favourable. The more unfavourable the a_w of the

substrate the greater is the delay in initiation of growth of micro-organisms. This is an important aspect of food preservation. In general, bacteria require more moisture than yeasts and yeasts more than molds.

Preservatives are divided into the following categories:

1. Class-I preservatives.
2. Class-II preservatives which are chemical compounds.

The list of class-I and class-II preservatives as per Rules laid down in PFA Act 1954 is given in Table-3.

TABLE-3
PREVENTION OF FOOD ADULTERATION RULES, 1955

Rule 55: Definition of Preservative

"Preservative" means a substance which when added to food, is capable of inhibiting, retarding or arresting the process of fermentation, acidification or other decomposition of food.

Classification of Preservatives.

Preservatives shall be divided into the following classes:

(i) Class-I preservatives shall be:

(a) Common salt
(b) Sugar
(c) Dextrose
(d) Glucose (Syrup)
(e) Spices
(f) Vinegar or acetic acid
(g) Honey
(h) Edible vegetable oils

Addition of Class-I preservatives in any food is not restricted, unless otherwise provided in the rules:

Provided that the article of food to which a Class-I preservative has been added conforms to the specifications laid down in Appendix 'B'.

(ii) Class-II preservatives shall be:

(a) Benzoic acid including salt thereof,
(b) Sulphurous acid including salts thereof,
(c) Nitrates or nitrites of sodium or potassium in respect of food like ham, pickle, meat,
(d) Sorbic acid including its sodium, potassium and calcium salts (proportionates of calcium or sodium, lactric acid, and acid calcium phosphate),
(e) Nicin
(f) Sodium and calcium propionate,
(g) Methyl or propyl *para*-hydroxy-benzoate
(h) Propionic acid, including esters or salts thereof,
(i) Sodium diacetate, and
(j) Sodium, potassium and calcium salts of lactic acid.

Use of more than one Class-II preservative prohibited

No person shall use in or upon a food more than one Class-II preservative:

Provided that where in column (2) of the table given below Rule 55, the use of more than one preservative has been allowed in the alternative, those preservatives may, notwithstanding anything contained in Rule 55, be used in combination with one or more alternatives, provided the quantity of each preservative so used does not exceed such number of parts out of those specified for that preservative in column (3) of the aforesaid table as may be worked out on the basis of the proportion in which such preservatives are combined.

Illustration: In the group of foods specified in item 6 of the table given below Rule 55, sulpur dioxide or benzoic acid can be added in the proportion of 40 parts per million or 200 parts per

million respectively. If both preservatives are used in combination and the proportion of sulphur dioxide is 20 parts per million, the proportion of benzoic acid shall not exceed the proportion of 100 parts per million.

Use of Class-II preservatives restricted

The use of class-II preservatives shall be restricted to the following group of foods in connection not exceeding the proportions given below against each.

	Article of food	Preservative	Parts per million
1.	Sausages and sausage meat containing raw meat, cereals and condiments	Sulphur dioxide	450
2.	Fruit, fruit pulp or juice (not dried) for conversion into jam or crystallised glace or cured fruit or other products:		
	(a) Cherries	Sulphur dioxide	2,000
	(b) Strawberries and raspberries	—do—	2,000
	(c) Other fruits	—do—	1,000
3.	Fruit juice concentrate	—do—	1,500
4.	Dried fruits:		
	(a) Apricots, peaches, apples, pears and other fruits	—do—	2,000
	(b) Raisins and sultanas	—do—	750
5.	Other non-alcoholic wines, squashes, crushes, fruit syrups, cordials, fruit juices and barley water (to be used after dilution).	—do— or Benzoic acid	350 600
6.	Jam, marmalade, preserved canned cherry and fruit jelly	Sulphur dioxide or Benzoic acid	40 200
7.	Crystallised glace or cured fruit (including candied peel)	Sulphur dioxide	150
8.	Fruit and fruit pulp not otherwise specified in the schedule	Sulphur dioxide	350
9.	Plantation white sugar, cube sugar, dextrose, gur or jaggery, misri	Sulphur dioxide	70
9. A	Khandsari (sulphur) and Bura	—do—	150
9. B	Refined sugar	—do—	40
10.	Corn flour and such like starches	—do—	100
11.	Corn syrup	—do—	450
11. A	Canned Rassogolla (the cans shall be internally lacquered with sulphur dioxide resistant lacquer)	—do—	100
12.	Gelatin	—do—	1,000
13.	Beer	—do—	70
14.	Cider	—do—	200

	Article of food	Preservative	Parts per million
15.	Alcoholic wines	Sulphur dioxide	450
16.	Sweetened mineral water (and sweetened ready-to-serve beverages)	Sulphur dioxide or Benzoic acid	70 120
17.	Brewed ginger beer	Benzoic acid	120
18.	Coffee extract	—do—	450
19.	Pickles and chutneys made from fruit or vegetables	Benzoic acid or Sulphur dioxide	250 100
20.	Tomato and other sauces	Benzoic acid	750
21.	Cooked pickled meat including ham and bacon	Sodium or potassium nitrite (calculated as sodium nitrate) or Commercial saltpetre (calculated as sodium nitrite)	200 500
22.	Danish tinned caviar	Benzoic acid	50
23.	Dehydrated vegetables	Sulphur dioxide	2,000
24.	Tomato puree and paste	Benzoic acid	750
25.	Syrups and sharbats	Sulphur dioxide or benzoic acid	350
26.	Dried ginger	Sulphur dioxide	2,000
27.	Hard boiled sugar confectionery	Sulphur dioxide	350
28.	Cheese or processed cheese	Sorbic acid including its sodium, potassium and calcium salts (calculated as sorbic acid) or Nicin (calculated as sorbic acid)	1,000 —do—
29.	Flour confectionery	Sorbic acid or its sodium salt	1,500
30.	Smoked fish (in wrappers)	Sorbic acid	(Only wrappers may be impregnated with sorbic acid).
31.	Dry mixes of rasogollas	Sulphur dioxide	100
32.	(a) Soups (other than canned) (b) Dried soups (c) Dehydrated soup mix when packed in containers other than cans	—do— —do— —do—	150 1,500 1,500
33.	Fruits and vegetables, flakes, powder, figs	—do—	600
34.	Flour for baked food	Sodium diacetate or propionate or Methyl propyl hydroxy benzoate	2,500 3,200

	Article of food	Preservative	Parts per million
34.	Flour for baked food	Sodium diacetate or propionate or	2,500
		Methyl propyl hydroxy benzoate	3,200
35.	Preserved chapaties	Sorbic acid	1,500
36.	Paneer or channa	Sorbic acid and its sodium, potassium or calcium salts (calculated as sorbic acid) or	2,000
		Propionic acid and its sodium or potassium salts (calculated as propionic acid)	2000
37.	Fat spread	Sorbic acid and its sodium, potassium and calcium salts (calculated as sorbic acid) or	1,000
		Benzoic acid and its sodium and potassium salts (calculated as benzoic acid) or both	1,000
38.	Jams, jellies, marmalades, preserved, crytallised, glazed or candid fruits including candid peels, fruit bars	Sorbic acid and calcium /sodium/potassium salts (calculated as sorbic acid)	500
39.	Fruit juice concentrates with preservatives for conversion in juices, nectars for ready-to-serve beverages in bottles/pouches selling through dispenser	—do—	100
40.	Fruit juices (tin, bottles or puoches)	—do—	200
41.	Nectars, ready-to-serve beverages in bottles, pouches or selling through dispensers		50

Rule 55-A: Use of Class-II preservatives in mixed foods

In a mixture of two or more foods or groups of foods mentioned against each item in the table under Rule 55, the use of Class-II preservative or preservatives shall be restricted to the limit up to which the use of such preservative or preservatives is permitted for the foods or groups of foods contained in such mixture.

Illustration: In the foods specified in Item 23 of the table given below Rule 55, sulphur dioxide can be added to dehydrated vegetables in the proportion of 2,000 parts per million. If this food is mixed with the food specified in Item 24 given in the said table, that is to say, tomato puree and paste, where benzoic acid is permitted to an extent of 250 ppm, then in the mixture containing equal parts of these two foods, the proportion of sulphur dioxide and benzoic acid shall be 1,000 ppm and 125 ppm respectively.

Rule 55-B: Restriction on use of nitrate and nitrite
No nitrate or nitrite shall be added to any infant food.

Rule 55-C: Use of natamycin for surface treatment of cheese (hard)
Natamycin may be used for surface treatment of cheese (hard) under label declaration as specified in clause (9) of sub-rule (ZZZ) of rule 42 subject to the following conditions, namely:
(i) Mximum level of application of natamycin shall not exceed 2 mg/dm^3.
(ii) The penetration depth of natamycin in cheese (hard) shall not exceed 2 mm.
(iii) The maximum residue level of natamycin in the finished cheese (hard) shall not exceed 1 mg/dm^3.
Container of food, which contains preservative not to be marked pure.

Foods for human consumption can be divided into the following main categories from plants and animals origins:

Plant origin:

1. Cereals and cereal products
2. Sugar and sugar products
3. Vegetable and vegetable products
4. Fruits and fruit products
5. Spices and its products
6. Edible oils.

Animal origin:

1. Meat and meat products
2. Poultry and eggs
3. Fish and other sea foods
4. Milk products

Most of these kinds of foods are readily decomposed by microorganisms unless special methods and preservatives are used for their preservation.

The addition of preservatives to food is controlled by Food Regulation Act (PFA Act 1954).

Inorganic Preservatives

Sulphur dioxide: Sulphur dioxide may be used in the form of gas, in solution as sulphurous acid or as the sulphites of sodium or potassium but for the regulations it is calculated as sulphur dioxide (SO_2). Sulphurous acid inhibits the growth of molds, yeasts and acrobic bacteria and also prevents the enzymatic browning of foods. SO_2 can be present in free or bound sulphur dioxide depending upon its dissociation in the solution. In aqueous solution, sodium metabisulphite forms sulphurous acid, the acid microbial compound. The effectiveness of sulphurous acid is enhanced at low pH values. In addition to the antimicrobial action of sulphites, theyare also used to prevent enzymatic and non-enzymatic changes and decolourization in some food containing natural colours.

Sodium and potassium nitrates and nitrites have been allowed in the preparation of meat and meat products. In recognition of the concern regarding the potential to form nitrosamine and to interfere with infant metabolism, the regulation prohibits the presene of added nitrates and nitrites in all foods specially

used by infants and children. Nitrites have a more potential preservation action than nitrates, their most important function being the inhibition of growth of *Clostridium botulinum* in crude foods. They also contribute to the colour of the product. The pink colour of meat preparation is due to the formation of myoglobin to form nitrosohaemoglobin. Nitrites and nitrates are used in curing solutions and curing mixtures of meats. Nitrites decompose to nitric acid which forms nitrosomyoglobin when it reacts with heme pigments in meats and thereby forms a stable red colour. Nitrates probably only react with secondary and tertiary amines to form nitrosamines which are known to be carcinogenic. Recent work has emphasized the inhibitory properties of nitrites towards *Clostridium botulinum* in meat products, particularly in canned meat products.

Organic Preservatives

Benzoic Acid: Benzoic acid is generally used as a preservative in the form of its calcium, sodium, potassium salts but for the purpose of regulation it is calculated as benzoic acid. Benzoic acid is only about half as effective as sulphurous acid. Benzoic acid retards the growth of yeast and molds, the effective agent being the undissociated acid. It is effective between pH 2.5 to 4.0 and diminished at pH value above 5.0.

Sorbic Acid: Sorbic acid is usually incorporated as a salt and is more effective against yeasts and molds than bacteria. Sorbic acid has been identified as the cause of the development of off-flavour in cheese due to decarboxylation by molds.

Chemical preservatives may inhibit the action of micro-organisms by interfering with their cell membranes, their enzymatic activities or genetic mechanisms. Other preservatives may be used as anti-oxidant to hinder the oxidation of unsaturated oils and fats, as neutralizers of acidity, as stabilizers to prevent physical change.

Sugar and Salt: These compounds tend to tie up moisture and thus have an adverse effect on micro-organisms. Sodium chloride is used in brine and curing solution and applied directly to the food.

Salt has been reported to have the following effects: (1) It causes high osmotic pressure and hence plasmolysis of cell. (2) It dehydrates food by drawing out and tying up moisture as it dehydrates microbial cell. (3) It ionizes to yield chlorine ions which are harmful to organisms. (4) It interferes with the action of proteolytic enzymes. The effectiveness of NaCl varies directly with its concentration and temperature. Sugars such as glucose and sucrose owe their effectiveness as preservatives to their ability to make water unavailable to micro-organisms and their osmotic effect.

The storage of foods in the frozen condition has been an important preservation method for centuries. Under the usual condition of storage of frozen foods microbial growth is prevented entirely and the action of food enzymes is greatly retarded. The effective and quick freezing method rapidly slows down the chemical deterioration and enzymatic reactions and stops microbial growth.

Low temperatures are used to retard chemical reactions and action of food enzymes and to slow down or stop growth and activity of microorganisms in food.

The lower the temperature, the slower will be chemical reaction, enzymatic action and microbiological growth and a low enough temperature will prevent the growth of any microorganisms. Each microorganism present has an optimal or best temperature below which it cannot multiply. As the temperature drops from this optimal temperature towards the minimal, the rate of growth of the organisms decreases and is slowest at the minimal temperature.

Drying usually is accomplished by the removal of water. Any method that reduces the amount of available moisture that is lowering the contents of water molecules in a food is a form of drying. Apart from mechanical drying, sugar, salt etc. are added to reduce the amount of available moisture. Moisture may be removed from food by sun-drying, and mechanical drying also.

The killing of micro-organisms by heat is supposed to be due to denaturation of the proteins and especially to the interaction of enzymes required for metabolism. The killing of organisms and their spores varies with the kind of organisms. Pasteurisation is a heat treatment that kills part of pathogenic bacteria but not all the microorganisms present. When milk is heated up to 150°C by use of steam injection followed by "flash evaporation" of condensed steam and rapid cooling, it is called sterilised, *i.e.*, free from all bacteria. This type of milk is called UHT (Ultrahigh Temperature) milk. It increases the self life of milk without refrigeration for a certain period.

Canning is defined as the preservation of food in sealed containers and usually implies heat treatment as the principal factor in the prevention of spoilage. Some inert gases like nitrogens also used in packed food products to make oxygen unavailable for microbiological spoilage.

Food preservation is the most important and absolutely essential aspect in food industries. By preservation seasonal foods are made available in off-seasons to the consumers. More and more modern technologies are being developed to prevent food spoilage. This review has covered the main principles and chemistry of food preservations and PFA Rules.

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