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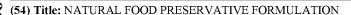
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(57) **Abstract:** A natural food preservative formulation to prevent microbial spoilage and oxidative damage in a food article. The natural preservative includes constituents derived mainly from plants, such as essential oils and organic acids and plant extract. Combination of different plant derived constituents to form different natural preservative formulations are disclosed. Tailor made formulation for specific food material is also disclosed. Methods for preparing the food preservative and application of the food preservative formulation into different food article/products are also disclosed.



NATURAL FOOD PRESERVATIVE FORMULATION

FIELD OF INVENTION

[1] The disclosure relates to a natural food preservative formulation used to prevent microbial spoilage and oxidative damage in a food article. The natural preservative is a formulation which encompasses constituents derived from plants, animals and microbes. The disclosure also provides methods for preparing the formulation and application of said formulation into different food article/products.

BACKGROUND

- [2] Microbial entities such as bacteria, viruses, spores, algae, and fungi often sprout in our food, and they get spoiled because of damage caused to them by such microorganisms. Microorganisms are the common agents of disease. There are two issues with such microbial growth in our food. First, the spoilage, second, the health hazards that come along with such microbial growth.
- There are some visible signs of microbial growth in our food, such as mould formation, discoloration, change in odour and change in taste. Odour can come from two sources: chemicals that are released from the food as the microbes decompose it, or chemicals produced directly by the microbes themselves. Microorganisms such as bacteria and moulds release their enzymes as they grow which eventually leads to structural decay and speeding up oxidative damage. The presence of oxygen enhances the growth of microorganisms, such as moulds and yeasts, and contributes directly to deterioration of fats, vitamins, flavours, and colours within foods through the work of enzymes. Presence of water in food expedites the microbial growth, and microorganisms use the water within the food to fuel their chemical reactions. Many fruits and vegetables are driedto avoid microbial growth. The moisture content in juice, jellies and pudding make them more vulnerable to microbial attack. For this reason, most of the juice we find in the market contains some preservatives.
- [4] An obvious example of natural preservative is the weak organic acids, such as acetic, lactic, benzoic and sorbic acid. These molecules inhibit the outgrowth of both bacterial and

fungal cells. Sorbic acid is also reported to inhibit the germination and outgrowth of bacterial spores (Sofos and Busta, 1981; Blocher and Busta, 1985). Most ofthe time organic acids alter the taste of the food and for some food article it is the distinct feature of that food such as in the case of pickles. Though we as humans have acquired the taste for acidic food but that doesn't mean they don't come with any health problems.

- [5] Most commonly used preservative in the food industry are sodium nitrate, sodium sulfites benzoic acid, butylated hydroxyanisole and sodium benzoate for acidic foods such as jams, salad dressings, Juices, Pickles, Carbonated Drinks. The most fruit based product has Vitamin C in it. Studies have shown when sodium benzoate and ascorbic acid (Vitamin C) is mixed it results in the formation of benzene. Benzene is a known carcinogen (Benzene Production from Decarboxylation of Benzoic Acid in the Presence of Ascorbic Acid and a Transition-Metal Catalyst, Lalita K. Gardner and Glen D. Lawrence, Journal of Agriculture and food chemistry May 1993 volume 41, number 5. McNeal TP et al., Survey of benzene in foods by using headspace concentration techniques and capillary gas chromatography, J AOAC Int. 1993 Nov-Dec;76(6):1213-9.).
- In another study it was shown that sodium benzoate along with artificial food colourings may increase hyperactive behaviour in some children (McCann Donna et al, Food additives and hyperactive behaviour in 3-year-old and 8/9-year-old children in the community: a randomised, double-blinded, placebo-controlled trial, September 6, 2007 DOI:10.1016/S0140-6736(07)61306-3; Jim Stevenson, Dietary influences on cognitive development and behaviour in children, Proceedings of the Nutrition Society (2006), 65, 361–365.).
- [7] Potassium sorbate, another preservativeused extensively in a wide variety of food products such as cheeses, pickles, sauces, soft drinks, and fish products as an antimicrobial agent. Recent studies have shown at excesses concentration potassium sorbate is clearly seen to be genotoxic to the human peripheral blood lymphocytes in vitro (Does potassium sorbate induce genotoxic or mutagenic effects in lymphocytes?, Sevcan Mamura, Deniz Yüzbasıogʻlu, Fatma Ünal, and Serkan Yılmaz; Toxicology in Vitro, 24 (2010) 790–794.).
- [8] In the meat industry traditionally salt is used for curing of meat but in excess, it can affect the flavour. In this day and age synthetic preservatives are also used to reduce the time and to

retain colour and texture. The use of salt is one of the oldest methods of preserving meat since at concentrations greater than 4% in the aqueous phase it inhibits the growth of most spoilage organisms. To inhibit the microbial growth in meat, the salt concentration in meatneeds to be around 17%, at which levels the product would be unpalatable. In most cured meat products the salt concentration is between 2.5 and 5%, and that includes nitrate and nitrite salts. Nitrite inhibits the growth of other organisms. Further Nitrite also reacts with proteins when heated to form compounds (called Perigotype factors) that inhibit the development of spores of Clostridium botulinum, which causes botulism, a severe type of food poisoning.

- [9] Nitrates and nitrites are used as a preservative for meats. It is observed that Nitrites can react with amines in meats to form nitrosamines, a class of potent carcinogens found in cigarette smoke (Karl-Otto Honikel, The use and control of nitrate and nitrite for the processing of meat products, Federal Research Centre for Nutrition and Food, E.C. Baumann Strasse 20, 95326 Kulmbach, Germany, Meat Science 78 (2008) 68–76). Infants are very susceptible to nitrate toxicity as they can develop methemoglobinemia or "blue baby syndrome". The commonly processed meats are luncheon meat, sausages, bacon, pork, ham, chicken, duck, and goose.
- [10] It is known that essential oils derived from some plants, extracts and natural acids have antimicrobial activity but to put them in use as food preservative is not easily done as said. Compared to synthetic preservatives the natural ingredients as preservatives need to be used in huge quantity and this leads to the issue of change in taste and smell. There have been attempts to combine enzymes with natural ingredients as done in the case of US 2003/0072862 A1. In US 2003/0072862 A1 salt of propionic, sorbic, phosphoric, citric, acetic, lactic and benzoic acid is combined with enzymes to enhance the shelf life of bakery products. A preservative composition made from natural ingredients alone, that can preserve the food article from spoilage for a reasonable period time without effecting the taste, colour and smell has not been developed. Natural preservative tailor made for meant, cake, bread and juice is yet to be disclosed.

OBJECTS

[11] An object of the disclosure is to replace synthetic preservatives and pesticides which are harm full for our health used in food article with natural preservatives. The other object of the disclosure is to inhibit microbial growth in edible food material and extend their shelf life with

the use of natural substances as preservatives, more preferably preventing the growth of pathogenic bacteria on food and beverages with the utilization of a formulation made out of natural substance.

[12] Another objective of the disclosure is a method to use a natural preservative to increase the shelf life of food article. Another object of the disclosure is the method to prepare the natural preservatives formulation.

SUMMARY

- [13] A food preservative having an essential oil of a plant, an organic acid or combinations thereof is disclosed. The essential oil of the plant can be cinnamon bark essential oil, clove bud essential oil, celery seed essential oil, cinnamon leaf essential oil, or combinations thereof. The organic acid can be lactic acid, citric acid, ascorbic acid or combinations thereof. A ratio of the essential of the plant to organic acid by weight ranges from about 7:2 to about 1:50.
- [14] The food preservative can further include a plant extract such as a rosemary extract, a nutmeg fruit extract, a cumin seed extract, and an oregano extract. The food preservative can further include a natural flavoring selected from the group consisting of a cassia extract, a cardamom extract, a vanilla extract, and a sugar. The food preservative can further include a bacteriocin.
- [15] The food preservative can further include one or more carrier such as polyoxyethylenesorbitanoleate, gum acacia, Jelucel^R, maltodextrin, wheat fiber, and silica.
- [16] A food preservative for preserving meat includes cinnamon bark essential oil, clove bud essential oil, rosemary extract, oregano extract and citric acid. The meat can be raw meat or processed meat. A food preservative for preserving meat includes; about 10% to about 30% of essential oil of cinnamon bark, about 10% to about 30% of essential oil of clove bud, about 10% to about 30% of rosemary extract, about 5% to about 30% of oregano extract; and, about 10% to about 40% of citric acid.
- [17] A food preservative for preserving meat also includes; about 17% to about 23% of essential oil of cinnamon bark, about 17% to about 23% of essential oil of clove bud, about 17%

to about 23% of rosemary extract, about 17% to about 23% of oregano extract; and, about 10% to about 40% of citric acid.

- [18] The food preservative can include nutmeg fruit extract, when the food preservative is for preserving processed meat. A food preservative for preserving processed meat includes about 5% to about 25% of nutmeg fruit extract, about 10% to about 30% of essential oil of cinnamon bark, about 10% to about 30% of essential oil of clove bud, about 10% to about 30% of rosemary extract, about 5% to about 30% of oregano extract, and about 10% to about 40% of citric acid.
- [19] Some embodiments provide a food preservative for preserving processed meat having; about % to about % of essential oil of cinnamon bark, 13% to about 19% of essential oil of clove bud, 13% to about 19% rosemary extract, 13% to about 19% of oregano extract, 13% to about 19% of nutmeg fruit extract; and 13% to about 19% of citric acid.
- [20] A food preservative for preserving baked or cooked products is disclosed. the food preservative includes cinnamon bark oil, cinnamon leaf oil, clove bud oil, and citric acid. A food preservative for preserving baked or cooked products having; about 0.1% to about 5% of cinnamon bark oil, about 0.1% to about 5% of cinnamon leaf oil andabout 90% to about 99% of lactic acid.
- [21] A food preservative for preserving beverages having cinnamon bark oil, cinnamon leaf oil, lactic acid and citric acid is disclosed. The beverage can be fruit juice, vegetable juice, cold press extracts, carbonated drink, non-carbonated soft drinks, coffee, tea, liquid concentrates, flavored waters, sports drink, energy drinks, liquid herbal extract without fat, fruit-flavored drinks or alcoholic products. A food preservative for preserving beverages having; about 5% to about 25% of cinnamon bark oil, about 5% to about 25% of cinnamon leaf oil, about 50% to about 70% of lactic acid and about 5% to about 25% of citric acid.
- [22] A food preservative for preserving products prepared with raw egg. The food preservative has cinnamon bark oil, rosemary extract and natamycin. A food preservative for preserving products prepared with raw eggwhere the food preservative has; about 1% to about 5% of cinnamon bark oil, about 10% to about 40% of rosemary extract, and about 1% to about 10% of natamycin.

[23] A food preservative for preserving products having a fruit product rich in pectin is disclosed. The food preservative includes clove bud oil, lactic acid, citric acid, cardamom, maltodextrin and silica. The fruit product rich in pectincan be fruit jam, fruit jelly, fruit compote, or fruit conserve. A food preservative for preserving products having a fruit product, the food preservative having; about 1% to about 10% of clove bud oil, about 10% to about 40% of lactic acid, about 10% to about 30% of citric acid, about 1% to about 10% of cardamom, about 10% to about 30% of maltodextrin and about 30% to about 50% of silica.

- [24] A food preservative for preserving a bean containing product is disclosed. The food preservative includes clove bud oil, rosemary extract, cumin seed oil, citric acid, ascorbic acid and lactic acid. The bean containing product can be hummus. A food preservative of for preserving bean containing product, the food preservative having; about 1% to about 20% of clove bud oil, about 0.1% to about 5% of rosemary extract, about 1% to about 10% of cumin seed oil, about 50% to about 70% of citric acid, about 15% to about 30% of ascorbic acid andabout 10% to about 20% of lactic acid.
- [25] A food preservative for preserving dairy or a dairy product is disclosed. The food preservative includes cinnamon bark oil, lactic acid and nisin. A food preservative for preserving dairy or the dairy product, the food preservative having; about 1% to about 10% of cinnamon bark oil, about 20% to about 40% of lactic acid and about 1% to about 10% of nisin.
- [26] A food preservative for preserving a pickle is disclosed. The food preservative includes cinnamon bark oil, lactic acid and nisin. The pickle has one or more of a vegetable and fruit. A food preservative for preserving a pickle, the food preservative having; about 1% to about 10% of cinnamon bark oil, about 20% to about 40% of lactic acid, and about 1% to about 10% of nisin.
- [27] A food preservative for preserving a halwa is disclosed. The food preservative includes cinnamon oil, citric acid, rosemary extract, and clove bud oil. The food preservative can further include sugar, oregano, oregano extract, cardamom extract, vanilla extract, lactic acid, nisin and natamycin. A food preservative for preserving halwa, the food preservative having; about 0.1 to about 2% of cinnamon oil, about 5% to about 10% of citric acid, about 0.05% to about 0.5% of rosemary extract, about 0.1% to about 1% of clove bud oil, about 1% to about 10% of sugar,

about 0.05% to about 0.5% of oregano, about 1% to about 5% of cardamom extract, about 0.1% to about 0.5% of vanilla extract, about 20% to about 60% of lactic acid, about 1% to about 10% of nisin, andabout 3% to about 6% of natamycin.

- [28] A liquid form of food preservative having a carrier such as gum acacia, maltodextrin or silica is disclosed. A powder form of food preservative having a carrier such as polyoxyethylenesorbitanoleateor hydrogenated castor oil is disclosed.
- [29] A method of preventing and inhibiting growth of growth of microbes by adding a food preservative to a food product is disclosed. The microbes that are inhibited can be Bacillus species, Rhizopus species, Penicillium species, Aspergillus species, yeast, Staphylococcus species, Coliforms, Lactobacillus species, Mucorsp, Acetobacter, acetic acid bacteria, Pseudomonas species, salmonella species, Listeria species, Clostridium species, Rhizopus species or Enterococci.
- [30] A method of preserving meat by adding a food preservative to meat is disclosed. A method of preserving beverages by adding a food preservative to a beverage is disclosed. A method of preserving products prepared with raw egg by adding a food preservative to a product having raw eggs is disclosed. A method of preserving food having a fruit product by adding a food preservative to a food having a fruit product is disclosed. A method of preserving bean containing products by adding a food preservative to a food product having a bean product is disclosed. A method of preserving dairy or dairy containing products by adding a food preservative to a food product having a dairy product is disclosed. A method of preserving a pickle by adding a food preservative to a pickle is disclosed. A method of preserving a halwa by adding a food preservative to a halwa is disclosed.

DETAILED DESCRIPTION OF THE INVENTION

[31] The disclosureprovides natural preservative formulations used for food items with its various embodiments, and each embodiment describes a novelfeature. Through these embodiments, a composition, a formulation and product along with a process of production and method of use is disclosed. These embodiments have to be understood in its broadest sense. The illustrations related to said embodiment provide the best mode of formulating the natural

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preservative according to the inventor which can be well understood by a person skilled in the art, and do not intend to narrow the scope of any subject matter claimed.

- [32] The term "food articles" or "food product" is used for an edible substance found naturally or prepared by combining multiple ingredients into a particular process. The process may be heating, freezing, fermentation, curing, or even segregation.
- [33] A formulation made of naturally found substance is disclosedhere, andsaid formulation act as a preservative for food articles. As a preservative the formulation extends the shelf life of the food article, prevent and inhibit microbial growth and prevent oxidative damages. Said formulation shall also be referred as a natural preservative. The natural preservative may also include carrier means and the carrier means will enable the application of preservative to various food articles. Different carrier means may be required according to the food article or the method or preparing such food article.
- [34] The natural preservative is a formulation made of natural constituents such as plant extract, oils and organic acids. Individually the components used for the natural preservative show no or marginal activity towards microbes, but, when combined in a specific formulation the improvement in activity is found to be significantly more than the aggregation of the activity of the components. The natural preservative extends the shelf life of perishable good by restricting microbial growth such as bacteria and moulds. It also curbs oxidative damage and prevents any undesirable enzymatic reaction. The preservative composition preserves the flavour and aroma, and at the same time does not alter the inherent taste and aroma of the product.
- [35] An aspect is a preservative formulation made from one or more plant extract, oil and organic acids. The formulation is a natural preservative capable of preventing and inhibiting microbial growth and oxidative damage in a food article.
- [36] The constituents of the natural preservative are divided into three groups, Group A contains plant oils, Group B contains Organic acids, Group C contains plant extracts. Group-A contains 1) Cinnamon bark oil, 2) Clove bud oil 3) Celery seed oil and 4) Cinnamon leaf oil. Group B contains 1) Lactic Acid, 2) Citric acid, and 3) Ascorbic acid. Group C contains 1) Rosemary extract, 2) Nutmeg extract, 3) Cumin seed extract, Oregano extract.

TABLE 1

GROUP A	GROUP B	GROUP C
Cinnamon Bark Oil,	Lactic Acid,	rosemary extract,
Clove bud oil,	Citric Acid,	Nutmeg fruit extract,
Cinnamon leaf oil.	Ascorbic acid.	Cumin seed extract,
		Oregano extract.

- [37] In one embodiment, natural preservative formulation is made of a combination of one or more plant oils and organic acid with some suitable carrier means. The plant oil and organic acid are present in 7:2 to 1:50 ratio in the natural preservative. In another embodiment the natural preservative formulation also include plant extracts from Group C. The natural preservative formulation made of the combination of one or more plant oils and organic acid are best suited from cake, bread, beverage, fruit jams and dairy products.
- [38] A food preservative having an essential oil of a plant, an organic acid or combinations thereof is disclosed. The essential oil of the plant can be cinnamon bark essential oil, clove bud essential oil, celery seed essential oil, cinnamon leaf essential oil, or combinations thereof. The organic acid can be lactic acid, citric acid, ascorbic acid or combinations thereof. A ratio of the essential of the plant to organic acid by weight ranges from about 7:2 to about 1:50.
- [39] The food preservative can further include a plant extract such as a rosemary extract, a nutmeg fruit extract, a cumin seed extract, and an oregano extract. The food preservative can further include a natural flavoring selected from the group consisting of a cassia extract, a cardamom extract, a vanilla extract, and a sugar. The food preservative can further include a bacteriocin.
- [40] The food preservative can further include one or more carrier such as polyoxyethylenesorbitanoleate, gum acacia, Jelucel^R, maltodextrin, wheat fiber, and silica.

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[41] Said natural preservative is capable of inhibiting the growth of microbes, particularly selected from the group; Bacillus sp., Rhizopus sp., Penicillium sp., Aspergillus sp., Yeast, Staphylococcus sp., Coliforms, Lactobacillus sp, Mucorsp, Acetobacter, Acetic acid bacteria, Pseudomonas sp., Salmonella sp, Listeria sp., Clostridium sp., Rhizopus sp. and Enterococci.

- [42] In one embodiment, the natural preservative formulation is made of cinnamon bark oil, clove bud and citric acid. When citric acid is made part of the formulation, citric acid makes about one fourth (1/4) to about one-half (1/2) of the formulation. Citric acid crystals used have a purity of over 98%. A preferred formulation has cinnamon oil to clove bud oil to citric acid in 1:1:2 to 1:1:4 and more preferably 5:4:10. The formulation is best suited for cake, fresh meat and flour based sweet dish (halwa). The formulation is capable of inhabiting Bacillus sp., Rhizopus sp., Penicillium sp., Aspergillus sp., Yeast, Staphylococcus sp., Coliforms, Lactobacillus sp, Mucorsp, Acetobacter, Acetic acid bacteria, Pseudomonas sp., Salmonella sp, Listeria sp, Clostridium sp, Rhizopus sp. and Enterococci.
- [43] In another embodiment, the formulation of cinnamon bark oil and clove bud oil also includes rosemary. When rosemary extract is made part of the formulation, rosemary extract makes one third (1/3) to two third (2/3) of the formulation. A preferred formulation has cinnamon oil to clove bud oil to rosemary extract in 1:1:1 to 1:1:6 and more preferably 1:1:4. The formulation is best suited for inhabiting Bacillus sp, Rhizopussp, Penicilliumsp, Aspergillus sp, Yeast, Staphylococcus sp., Coliforms, Lactobacillus sp, Mucorsp, Acetobacter, Acetic acid bacteria, Pseudomonas sp., Salmonella sp, Listeria sp., Clostridium sp., Rhizopus sp. and Enterococci in fresh meat, and flour based sweet dish (halwa).
- [44] In another embodiment, the formulation of cinnamon bark oil and clove bud oil also includes oregano. When oregano is made part of the formulation, oregano makes about one-eighth (1/8) to about one-twelfth (1/12) of the formulation. The formulation is best suited for preserving raw meat and carbohydrate rich food.
- [45] In another embodiment, the formulation of cinnamon bark oil and clove bud oil also includes citric acid, rosemary extract, celery seed oil and oregano. In said formulation celery seed oil makes one tenth (1/10) of the formulation, clove bud oil makes one tenth (1/10) of the formulation, rosemary extract makes (2/5) of

the formulation, celery seed oil makes about one tenth (1/10) of the formulation, and oregano makes about one tenth (1/10) of the formulation.

- [46] According to another embodiment, in the formulation of natural preservative, cinnamon bark oil makes about 10-30% of the formulation, clove bud oil makes about 10-30% of the formulation, citric acid makes about 10-40% of the formulation, rosemary extract makes about 10-30% of the formulation, and oregano makes about 5-30% of the formulation. Said natural preservative is capable of inhibiting the growth of microbes, particularly selected from the group; Bacillus sp., Rhizopus sp., Penicillium sp., Aspergillus sp., Yeast, Staphylococcus sp., Coliforms, Lactobacillus sp, Mucorsp, Acetobacter, Acetic acid bacteria, Pseudomonas sp., Salmonella sp, Listeria sp., Clostridium sp., Rhizopus sp. and Enterococci.
- [47] In some embodiments a flavoring ingredent or constituent may be added to the natural preservative. The flavoring constituents are selected from Cassia Extract, Cardamom extract, Vanilla extract, Sugar or a combination there of.
- [48] The natural preservative can also include bacteriocins in the preservative formulation. Bacteriocin is a form of protein that is produced by bacteria. Bacteriocins are ribosomally-synthesized antimicrobial peptides or proteinaceous compounds produced by bacterial strains. They are generally effective in inhibiting the growth of similar or closely related bacterial strains. (Ramu R1, Shirahatti PS, Devi AT, Prasad A, J K, M S L, F Z, B L D, M N NP; Bacteriocins and Their Applications in Food Preservation; Crit Rev Food SciNutr. 2015 Jul 20:0).
- [49] Another aspect is a natural preservative formulationsalong with one or more suitable carrier means. Carrier means includes surfactants, fillers or any other additive that enable the use of the formulation as a preservative. An essential requirement for the natural preservative is that it should be able to spread in the food article uniformly to protect it from microbial attacks. The natural preservative is made in liquid formulation and also in solid powdered formulation, according to the needs. For making a liquid natural preservative product carrier means can be selected but not limited to tween 80, gum acacia, hydrogenated castor oil or a combination thereof. For making a powdered natural preservative product carrier means can be selected but not limited to jelucel, malto dextrin, wheat fibre, silica or a combination thereof. The carrier

means make 25% to 75% of the preservative composition. JELUCEL® (Jelu-Werk Josef Ehrler GmbH & Co. KG) is a powdered cellulose made from natural raw materials and used as a food additive. It is a tasteless, white, fine powder with a bulk density of approximately 140 g/l. It is used in food products to add dietary fiber and as a texturiser, texture stabiliser, humectant, anticaking and anti-sticking agent, and a carrier substance for other additives.

- [50] The constituents used have specific phytochemical feature, as follows:
- [51] Cinnamon bark oil is primarily having linalool (36.0%), limonene (8.3%), α -terpineol (7.8%), terpinen-4-ol (6.4%), γ -terpinene (3.5%), α terpinene (2.3%) and 1, 8-cineole (2.3%) and methyl eugenol (12.8%).
- [52] Rosemary extract is primarily having rosmarinic acid, alpha-pinene, linalool and camphen.
- [53] Clove bud oil primarily ismade of eugenol (87%), eugenyl acetate (8.01%) and β -Caryophyllene (3.56%).
- [54] Celery seed oil is primarily having sedanolide, sedanonic anhydride, d-limonene, selinene and sesquiterpene alcohols. pinene, cymene, caryophyllene, pinene, santalol, dihydrocarvone, apigenin, isoquerticin, sedanolide, sedanonic anhydride, d-limonene, and sesquiterpene alcohols.
- [55] Oregano essential oil is primarily having Carvacrol, Thymol, β -fenchyl alcohol, δ -terpineol, γ -terpinene, α -terpinene, and 1-methyl-3-(1-methylethyl)benzene.
- [56] Nutmeg fruit extract is primarily having pinenes, camphene, p-cymene, sabinene, phellandrene, terpinene, limonene, myrcene, linalool, geraniol, terpineol, myristicin, elemicin, safrol, eugenol and eugenol derivatives.
- [57] Cumin seed extract primarily hasthymoquinone, dithymoquinone, thymohydroquinone and thymol.
- [58] Another aspect is tailor made formulations of natural preservative for different food articles. The constituents of the formulations are derived from at least two groups from Group-A, Group-B and Group-C.

1. MEAT

[59] In one embodiment, a natural preservative formulation suitable as a preservative for meat is made using cinnamon bark oil, clove bud oil, rosemary extract, oregano extract, citric acid. The natural preservative increses the shelflife of the meat product by over 30 days.

[60] In one embodiment the natural preservative formulation for meat product is cinnamon bark oil in the range of 10-30%, clove bud oil in the range of 10-30%, rosemary extract in the range of 10-30%, oregano extract in the range of 5-30%, and citric acid in the range of 10-40%. More specifically, the formulation for raw meat products is cinnamon bark oil in the range of 20±3%, clove bud oil in the range of 20±3%, rosemary extract in the range of 20±3%, oregano extract in the range of 20±3%, and citric acid in the range of 20±3%. The formulation for the active constituents of natural preservative for meat product is provided in the table below.

TABLE 2

INGREDIENTS	PERCENTAGE	SPECIFIC
	RANGE	COMPOSITION
cinnamon bark oil	10-30%	20±3%
clove bud oil	10-30%	20±3%
rosemary extract	10-30%	20±3%
oregano extract,	5-30%	20±3%
citric acid	10-40%	20±3%

[61] In an another embodiment the active constituents in the natural preservatives tailor made for raw meat are: cinnamon bark oil about 1%-10%, more preferably 4%-7%; Citric acid 1%-20%, more preferably 8%-12%; Clove bud oil about 1%-6%, more preferably 4%-7%; Natamycin about 1%-20%, more preferably 7%-12%; Lactic acid 20%-60%, more preferably 38%-42%; Celery seed oil about 1%-10%, more preferably 4%-7%; oregano oil about 0.1%-4%, more preferably 0.5%-2.5%; and rosemary about 1%-4%, more preferably 3%.

[62] In an another embodiment the active constituents in the natural preservatives tailor made for processed meat has nutmeg fruit extract about 5-25% along with cinnamon bark oil, about

10-30%, clove bud oil, 10-30%, rosemary extract, 10-30%, oregano extract, 5-25%, and citric acid. 10-40%. More specifically nutmeg fruit extract about 10±3% along with cinnamon bark oil about 16±3%, clove bud oil 13±3%, rosemary extract 23±3%, oregano extract 10±3%, and citric acid 20±3%.

TABLE 3

	PERCENTAGE	SPECIFIC
INGREDIENTS	RANGE	COMPOSITION
cinnamon bark oil	10-30%	16±3%
clove bud oil	10-30%	13±3%
rosemary extract	10-30%	23±3%
oregano extract	5-25%	10±3%
nutmeg fruit extract	5-25%	10±3%
citric acid	10-40%	20±3%

- [63] In another embodiment the natural preservatives for processed meat is made of constituents selected from Cinnamon Bark Oil about 0.1%-2%, more preferably 0.7%-1.5%; celery seed oil about 1%-2%, more preferably 1.4%-1.6%; nutmeg fruit oil about 1%-6%, more preferably 2%-4%; rosemary extract about 1%-5%, more preferably 2%-3%; lactic Acid about 60%-80%, more preferably 65%-75%, and natamycin about 10%-20%, more preferably 12%-16%.
- [64] The natural preservative for meat may also include carrier means. The carrier means and the active constituents are mixed in a specific ratio. A combination of Tween 80, gum acacia, maltodextrin and hydrogenated castor oil is the preferred excipients.
- [65] In one embodiment the natural preservative for meat is in powdered form and makes use of gum acacia, Maltodextrin and silica as carrier means. The ratio between the active constituent and the carrier mean is 1:9 to 3:2. In another embodiment the natural preservative for meat is in

liquid form and makes use of Tween 80, hydrogenated caster oil. The ratio between the active constituent and the carrier mean is 1:9 to 3:2.A person skilled in the art can use any other suitable carrier means.

- [66] The natural preservative increases the shelf life of the raw meat up to 30 days in comparison to meat product without preservative. In case of processed meat the shelf life is increased to over 3 months. The natural preservative is capable of preserving the natural texture and colour of the meat, protecting it from oxidative damages. After 30 days of storage no colour impartation, no sensory variations is observed. The natural preservative formulation for meat is capable of inhibiting microbial growth in meat during the time of storage. The formulation acts as an enzyme inhibitor to pathogenic microbes and hinders their growth. The formulation inhibits the growth of microbes by interfering with DNA synthesis, cell wall synthesis and cell metabolism of the microbes.
- [67] In one embodiment the natural formulation suited for meat can inhibit and prevent the growth of Staphylococcus sp, Yeast, Coliforms, Pseudomonas sp., Salmonella sp., Listeria sp., Clostridium sp., Lactic acid bacteria., Rhizopus sp., Aspergillus sp., and Enterococci.
- [68] The raw meat as stated above is selected from but not limited to red meat such as beef, veal, lamb, venison and pork, and white meat or light meat such as chicken, turkey, duck, goose and rabbit are there. The natural preservative can be used for fresh or processed meat. Processed meat includes but not limited to bacon, ham, hot dogs, sausages, salami, corned beef, beef jerky, canned meat and meat-based sauces.

2. BEVERAGES

- [69] In another aspect, a natural preservative formulation suitable for beverages is disclosed. In one embodiment the natural preservative for us in beverages is made with cinnamon bark oil, cinnamon leaf oil, lactic acid, and citric acid, the formulation is provided in the table below.
- [70] In one embodiment the formulation for a natural preservative for beverages is cinnamon bark oil about 5-25%, cinnamon leaf oil about 5-25%, Lactic Acid about 50-70% and citric acid about 5-25%. More preferably the formulation is cinnamon bark oil about 12±3%, cinnamon leaf oil about 16±3%, Lactic Acid about 60±5% and citric acid about 10±3%.

TABLE 4

INGREDIENTS	PERCENTAGE RANGE	PRECISE
Cinnamon bark oil	5-25%	12±3%
Cinnamon leaf oil	5-25%	16±3%
Lactic Acid	50-70%	60±5%
Citric Acid	5-25%	10±3%

[71] The natural preservative for beverages may also include a carrier means; the carrier means and the active constituents are mixed in a specific ratio. The cinnamon bark oil, cinnamon leaf oil, lactic acid, and citric acid are the active constituent of the natural preservative for beverages. The active constituents and carrier mean are in 1:10 to 2:3. The preferred carrier means that can be used for natural preservative for beverages are tween 80 and hydrogenated caster oil.

[72] In some embodiment the natural preservative for beverages may also include cassia extract as a flavouring agent. The cassia extract makes 0.2 to 4% of the beverage preservative.

[73] In another embodiment, the active constituents part make up for 20% to 30% natural preservative tailor made for drinks in which citric acid makes up about 5% to 20%, Cinnamon oil makes up about 5% to 15%, Cassia extract makes up about 5% to 20%, Natamycin (95%) makes up about 0.1% to 1%, Nisin makes up about 1 to 5%, lactic acid makes up about 1% to 5%. Excipients make about 70 to 80 % of the composition. Preferred excipients are Tween 80 and hydrolyzed castor oil, but the disclosure is not limited by the excipient used. A person skilled in the art can use other known excipient as desired. Tween 80 is added about 30% to 50% and hydrolyzed castor oil makes about 25% to 35% of the natural preservative.

[74] The natural preservative increases the shelf life of the beverages up to 28 days in comparison to beverages without preservative. The natural preservative is capable of preserving

the original colour of the beverages, protecting it from oxidative damages. After 28 of storage, no visible colour variation or, flavour impartation and sensory variations are observed.

- [75] In the above embodiment of the formulation suited for beverages, the formulation is capable of inhibiting microbial growth in beverages during the time of storage. The formulation acts as an enzyme inhibitor to pathogenic microbes and hinders their growth. The formulation also interferes with DNA synthesis, cell wall synthesis and cell metabolism of the microbes.
- [76] In one embodiment the natural formulation suited for beverages can inhibit and prevent the growth of Yeast, Lactobacillus sp, Acetic acid bacteria, Bacillus sp. The natural preservative makes about 150ppm to 250ppm of the beverage.
- [77] In the natural preservative Citric acid, Cinnamon oil and cassia extract are present in a ratio 10:1:1 to 1:5:5, preferably about 5:2:2 to 2:2:1 and more preferably 2:1:1. In the organic composition, the citric acid makes up about 5% to 50%, Cinnamon oil makes up about 5% to 25%, and Cassia extract makes up about 5% to 25%.
- [78] In one embodiment, a process to increase the shelf life of beverage without the use of synthetic preservatives involves, adding natural preservative into a liquid beverage, and mixing the natural preservative in the liquid beverage to get a true solution. To be more precise, the active component of the natural preservative includes blend of Citric acid, Cinnamon oil and cassia extract, present in a ratio of 10:1:1 to 1:5:5 and the blend are present in the beverage in about 40ppm to 1000ppm, more preferably 120ppm to 180ppm. In a more preferred scenario, the active constituents of natural preservative also include Natamycin, Nisin and Lactic acid. When added to the beverage, Natamycin is about 1ppm to 25ppm, more preferably 1ppm to 5ppm; Nisin is about 4ppm to 100ppm, more preferably 10ppm to 20ppm and lactic acid is about 4ppm to 100ppm, more preferably 10ppm to 20ppm of the liquid beverage.
- [79] Beverages include fruit juice, vegetable juice, cold press extracts, carbonated and non-carbonated soft drinks, frozen ready-to-drink beverages, coffee without cream, tea, as well as liquid concentrates, flavoured waters, sports drink, energy drinks, liquid herbal extract without fat and fruit-flavoured drinks and alcoholic products.

3. BAKERY PRODUCT

[80] Some embodiments provide a natural preservative formulation suitable for bakery productis disclosed, the natural preservative for bakery product capable of increasing the shelf life of bread and cake up to 10 to 15 days. The active constituents of the formulation are made from cinnamon bark oil, cinnamon leaf oil, clove bud oil, and citric acid. The formulation for bread and cake provided in the table below.

[81] In one embodiment, the natural preservative formulation suitable for bakery product is cinnamon bark oil about 0.1-5%, clove bud oil about 0.1-5%, cinnamon leaf oil about 0.1-5% and Lactic Acid about 90-99%. In another embodiment for natural preservative for bread the formulation is cinnamon bark oil about 0.9±0.3%, clove bud oil about 0.3±0.1%, cinnamon leaf oil about 4±3% and Lactic Acid about 94±5%. In another embodiment, natural preservative for cake, the formulation is cinnamon bark oil about 1.5±0.3%, clove bud oil about 0.2±0.1%, cinnamon leaf oil about 0.8±2% and Lactic Acid about 96±3%.

TABLE 5

INGREDIENTS	PERCENTAGE RANGE	BREAD	CAKE
cinnamon bark oil	0.1-5%	0.9±0.3%	1.5±0.3%
clove bud oil	0.1-5%	0.3±0.1%	0.2±0.1%
cinnamon leaf oil	0.1-5%	4±3%	0.8±2%
Lactic Acid	90-99%	94±5%	96±3%

[82] The natural preservative for bakery products may also include carrier means. The carrier means and the active constituents are mixed in a specific ratio. The cinnamon bark oil, cinnamon leaf oil, clove bud oil, and citric acid are the active constituent of the natural preservative for bakery products. The active constituents and carrier mean are in 1:10 to 2:3. The carrier means are selected from a group of Maltodextrin, whet fiber and silica.

[83] According another embodiment in the formulation of natural preservative tailor made for bread, cinnamon oil makes about 0.1%-3%, more preferably 0.5%-0.7% and Clove bud oil

makes 0.1%-3%, more preferably 1%-2%. Encapsulated Cinnamon leaf powder is present about 1%-7%, more preferably 2%-4% of the natural preservative. Sugar makes about 5% to 40%, more preferably 15%-20% of the natural preservative and Lactic acid makes about 10 to 67%, more preferably 20%-40%. Excipients are added to make the mixture into powdered form. Wheat fibre and silica are such suitable excipient, and a person skilled in the art can use other suitable excipients accordingly.

- [84] In yet another embodiment, a formulation for the natural preservative tailor made for Cake is made of cinnamon bark oil about 3%-17%, more preferably 6%-10%; citric acid about 3%-35%, more preferably 13%-20%; clove bud oil about 3%-17%, more preferably about 2%-10%; Lactic acid about 3%-35%, more preferably 13%-20% and Nisin about 5%-17%, more preferably 10%-14% of the natural preservative.
- [85] The natural preservative increases the shelf life of the bread up to 10 to 15 days in comparison to bread without preservative. The natural preservative is capable of resisting the baking temperature and making it easier to use it before cooking. Some embodiments natural preservative for bread, the cinnamon leaf oil may be encapsulated so that it releases the oil during baking. Only after 10 to 15 days of storage, flavour impartation or structural damage is observed.
- [86] In the above embodiment of the formulation suited for bakery products, the formulation is capable of preventing microbial growth in bread during the time of storage. The formulation also capable of interferes with cell wall synthesis and cell metabolism of the microbes, so not only is the formulation capable of preventing microbial growth it can inhibit a microbial growth.
- [87] In one embodiment the natural formulation suited for bakery products can inhibit and prevent the growth of Bacillus sp, Rhizopussp and Penicillium sp. and Aspergillus sp.
- [88] In another embodiment, a method to preserve bakery product from spoilage and to increase its shelf life is provided. The method includes adding the natural preservative into the flour, followed by mixing the flour with necessary ingredients and yeast, keeping the mixture for fermentation and, cooking the mixture after fermentation. Sugar and Lactic acid facilitate smooth fermentation in the presence of the cinnamon oil and Clove bud oil. Encapsulated cinnamon leafs

extract do not interfere with the fermentation process in case of bread. The cinnamon leave powder is released from encapsulation during the cooking process and then onwards it prevents microbial growth in the bread.

- [89] Adding the natural preservative before cooking or baking enables a uniform distribution of preservative and a longer shelf life. The conventional method of preservation includes painting with antimicrobial preservatives on the surface of the bread. Such methods cannot hold microbial growth for a longer durationa slightest wear and tear makes the bread susceptible to moulding.
- [90] Bread is not to be limited to the traditionally baked loaf bread. Here bread includes any food usually baked/cooked and leavened food made of a mixture whose basic constituent is flour, such as biscuits, zucchini Bread, naan, baguette, challah, pita, brioche, ciabatta, bagel, tortilla, matzo, paratha, roti and Kifli. The flour can be from any raw grains or dried roots, preferably with moderate to high protein content. The bread can be steamed, baked or made in a pan.
- [91] Unlike Bread, Cake is much sweeter, and the flour used for cake has low protein content compared to flour used for bread. The protein is supplemented in cake through the use of eggs. The addition of eggs makes the final product susceptible to different pathogens and accordingly the natural preservative composition is to be modified.

4. RAW EGG BASED

- [92] Some embodiments provide a natural preservative formulation suitable for raw egg based products is made from cinnamon bark oil, and rosemary extract. In some embodiment natural preservative tailor made for raw egg based food product also includes Natamycin along with cinnamon bark oil and rosemary extract.
- [93] In one embodiment the formulation for natural preservative for raw egg based products is Cinnamon bark oil about 1-15%, Rosemary extract about 40-80%, and Natamycin in the range 1-20%. More precisely Cinnamon bark oil about 12±3%, Rosemary extract about 76±3%, and Natamycin in the range 18±3%. The active constituents in the natural preservatives are shown in the table below.

TABLE 6

INGREDIENTS	PERCENTAGE RANGE	PRECISE
Cinnamon bark oil	1-5%	12±3%
Rosemary extract	10-40%	16±3%
Natamycin	1-10%	5±3%

- [94] In another embodiment the formulation of natural preservative for raw egg based product is cinnamon about 1%-10%, more preferably 5%-8%; rosemary about 75%-90%, more preferably 80%-85%, and Natamycin 10%-25%, more preferably 15%-20%.
- [95] In one embodiment the active constituents are blended with suitable emulsifier, surfactants or fragrance ingredient and so on to form a liquid product. Tween 80 is used as an excipient. A person skilled in the art can use any other suitable excipient. In 1:10 to 2:3 ratios the active constituents are blended with the excipient.
- [96] The natural preservative increases the shelf life of the raw egg based products up to 6 months in comparison to raw egg based product without preservative. The addition of natural preservative does not alter the texture. After six months of storage, no significant change in flavour or texture was observed.
- [97] In the above embodiment of the formulation suited for raw egg based products, the formulation is capable of reducing oxidative rancidity and preventing microbial growth during the time of storage. The formulation also interferes with cell wall synthesis; inhibit the enzymatic activity of microbes and cellular metabolism of the microbes.
- [98] In one embodiment the natural formulation suited for the raw egg based products can inhibit and prevent the growth of Bacillus sp., Yeast, Aspergillus sp., Penicilliumsp, Staphylococcus sp., Coliforms.

[99] Raw egg based products for which said preservative can be used are, but not limiting, for Mayonnaise, Caesar salad dressing, Hollandaise sauce and eggnog also ice cream, smoothies and milkshakes.

5. BEAN DIP

[100] Some embodiments provide a natural preservative formulation suitable for bean dip is disclosed, and the formulation is made of Citric acid from group B, Clove bud oil from Group A and rosemary extract from Group C. In a more preferred embodiment Cumin seed oil and Ascorbic acid is also made part of the natural preservative for bean dip. In some embodiment, the natural preservative formulation tailored made for bean dip also includes lactic acid, natamycin and nisin along with citric acid, rosemary, clove bud, cumin, and ascorbic acid.

[101] In one embodiment the formulation for natural preservative for been dip is in one embodiment the formulation for natural preservative for bean dip is Clove bud oil about 1-20%, Rosemary extract about 0.1-5%, Cumin seed oil about 1-10%, Citric acid about 50-70%, Ascorbic acid about 15-30% and Lactic acid 10-20%. More preferably the formulation to be Clove bud oil about 10±3%, Rosemary extract about 2±0.5%, Cumin seed oil about 6±1%, Citric acid about 57±5%, Ascorbic acid about 20±0.5% and Lactic acid 15±3%. The formulation for natural preservative for bean dip is provided in the table below.

TABLE 7

INGREDIENTS	PERCENTAGE RANGE	PRECISE
Clove bud oil	1-20%	10±3%
Rosemary extract	0.1-5%	2±0.5%
Cumin seed oil	1-10%	6±1%
Citric acid	50-70%	57±5%
Ascorbic acid	15-30%	20±0.5%

Lactic acid	10-20%	15±3%
Lactic acid		

[102] The natural preservative for bean dip can also have a carrier mean such as Maltodextrin. The ratio between active constituents of natural preservative for bean dip to carrier mean is about 3:7-1:1.

[103] In yet another embodiment the active constituents in the natural preservatives for bean dip are: citric acid about 10%-30%, more preferably 20%-25%; rosemary extract about 0.1%-2%, more preferably 0.1%-0.6%; clove bud oil about 1%-6%, more preferably 2%-4%; cumin seed oil about 1%-5%, more preferably 1%-3%; Ascorbic acid about 0.1%-2%, more preferably 0.1%-1%; Lactic acid about 15%-30%, more preferably 20%-25%; Natamycin about 10%-20%, more preferably 12%-16% and Nisin about 10%-30%, more preferably 15%-20%.

[104] In one embodiment the natural preservative is a solid powdered product. The active constituents are blended with suitable carrier means to form a solid powdered product. A person skilled in the art can use any suitable excipients, for the present embodiment Maltodextrin used. The active constituents are blended with the excipient in a 3:1 to 3:2 ratios.

[105] The natural preservative increases the shelf life of the bean dip up to 10 to 12 days in comparison to bean dip without preservative. The addition of natural preservative does not result in any form of colour impartation, no impact on texture preserve the flavour. After 10 to 12 days of storage, not much alteration in aroma or structural damage was observed.

[106] In the above embodiment of the formulation suited for bean dip, the formulation is capable of preventing microbial growth during the time of storage. The formulation interferes with the microbial cell wall synthesis; inhibit the enzymatic activity of microbes and cellular metabolism of the microbes.

[107] In one embodiment, the natural formulation suited for bean dip can inhibit and prevent the growth of Bacillus sp., yeast, Aspergillus sp., Penicilliumsp, Staphylococcus sp., and Coliforms.

[108] Bean dip includes any form of paste or mash made from cooked beans or seed. Beans may be selected from but not limited to Black turtle, mustard, pinto bean, white beans, chickpeas, soya bean, pigeon peas, sesame seeds, and kidney beans. Hummus is one such bean dip.

6.FRUIT JAM

[109] Some embodiments provide a natural preservative formulation suitable for fruit jam rich in pectin is disclosed. The formulation is made of cinnamon bark oil and clove bud oil of Group A and citric acid from Group B. In some embodiment lactic acid, and natamycin is also made part of natural preservative for fruit jam along with cinnamon bark oil, clove bud oil and citric acid.

[110] In one embodiment the formulation for natural preservative for fruit jam is Clove bud oil about 1-10%, Lactic acid about 10-40%, Citric acid 10-30%, Cardamom about 1-10%, Maltodextrin about 10-30% and Silica about 30-50%. More preferably the formulation is Clove bud oil about 5±2%, Lactic acid about 35±3%, Citric acid 25±5%, Cardamom about 3±2%, Maltodextrin about 10±5% and Silica about 20±5%. The formulations are provided in the table below.

TABLE 8

INGREDIENTS	PERCENTAGE RANGE	PRECISE
Clove bud oil	1-10%	5±2%
Lactic acid	10-40%	35±3%
Citric acid	10-30%	25±5%
Cardamom	1-10%	3±2%
Maltodextrin	10-30%	10±5%
Silica	30-50%	20±5%

[111] In another embodiment the active constituents in the natural preservatives are: citric acid about 20%-40%, more preferably 25%-30%; clove bud oil about 1%-10%, more preferably 5%-10%; cardamom extract about 1%-10%, more preferably 1%-5%; lactic acid about 25%-45%, more preferably 35%-40% and Natamycin about 30%-40%, more preferably 25%-30%.

- [112] In one embodiment the natural preservative tailor made for fruit jam is a solid powdered product. The active constituents are blended with suitable carrier means to form a solid powdered product. Maltodextrin and Silica combo is used as an excipient for natural preservative; a person skilled in the art can use any suitable excipient. The active constituents are blended with an excipient in a 7:3 to 1:1 ratios.
- [113] The natural preservative increases the shelf life of the Fruit jam up to 6 months in comparison to Fruit jam without preservative. The addition of natural preservative does not result in any form of colour impartation or affect the flavour. The preservative is stable at different pH. After six months of storage, no alteration in aroma or taste was observed.
- [114] In the above embodiment of the formulation suited for Fruit jam, the formulation is capable of preventing microbial growth during the time of storage. The formulation interferes with the microbial cell wall synthesis and also inhibits the enzymatic activity of microbes.
- [115] In one embodiment the natural formulation suited for Fruit jam can inhibit and prevent the growth of Yeast, Lactobacillus sp, Acetic acid bacteria, and Bacillus sp.
- [116] Fruit jam includes fruit product rich in pectin but not limited to Jam, Jelly, Compote, and Conserve made from fruits and other plant products.

7. DAIRY PRODUCTS

- [117] Some embodiments provide a natural preservative formulation suitable for milk based sweet products is disclosed. The formulation is made from Cinnamon Bark Oil. In yet another embodiment, Lactic Acid and Nisin is also made part of natural preservative for milk based sweet products.
- [118] In one embodiment the formulation of the active constituents is Cinnamon bark oil about 1-10%, Lactic acid about 20-40% and Nisin about 1-10%. More preferred formulation is

Cinnamon bark oil about 5±3%, Lactic acid about 30±5% and Nisin about 5±3%. The formulations are presented in the table below.

TABLE 9

INGREDIENTS	PERCENTAGE	PRECISE
MOREDIENTS	RANGE	TRECISE
Cinnamon bark oil	1-10%	5±3%
Lactic acid	20-40%	30±5%
Nisin	1-10%	5±3%

[119] In another embodiment the active constituents in the natural preservatives are; Cinnamon Bark Oil about 1%-10%, more preferably 3%-7%, sugar about 1%-10%, more preferably 3%-7%, Lactic Acid about 60%-40%, more preferably 60%-50% and Nisin about 20%-40%, more preferably 25%-35%.

[120] In one embodiment the natural preservative tailor made for dairy products is a solid powdered product where the active constituents are blended with suitable carrier means to form a solid powdered product. Jelucel, Maltodextrin and Silica combo are the preferred excipient. A person skilled in the art can use any other suitable excipient. The active constituents are blended with the excipient in a 9:4 to 6:7 ratios.

[121] Milk based sweet products include but not limited to; thickened milk dessert, condensed dairy products, ice cream, yoghurt and Khoa.

8. HALWA

[122] Some embodiments provide a natural preservative formulation suitable for Halwais disclosed. Halwa is a sweet dish made from rice, soy, semolina flour or a combination thereof.

[123] The formulation is made from cinnamon bark oil, clove bud oil, rosemary extract, oregano extract, lactic acid, and citric acid. In some embodiment, nisin and natamycinare also made part of natural preservative for halwa.

[124] In one embodiment active constituents in the natural preservatives for Halwa are: cinnamon oil about 0.1 to 2%; citric acid about 5%-10%; rosemary extract about 0.05%-.5%, clove bud oil about 0.1%-1%, sugar about 1%-10%, oregano about 0.05%-0.5%, cardamom extract about 1%-5%, vanilla extract about 0.1%-0.5%, lactic acid about 20%-60%, nisin about 1%-10% and, natamycin about 3%-6%.

[125] In another embodiment the natural preservative tailor made for Halwa is a liquid product. The active constituents are blended with suitable carrier means such as emulsifier, surfactants or fragrance ingredient and so on to form a liquid product. Excipients preferred are tween 80. A person skilled in the art can use any suitable excipient. The active constituents are blended with the excipient in 4:1 to 1:1 ratios.

[126] The natural preservative prevents and inhibits microbial growth in Halwa. Preservative gives a shelf life of about 6 to 7 day for Halwa.

9. PICKLE

[127] A natural preservative for pickle, the preservative extend the shelf life of the pickle by up to 90 days. The active constituents of the natural preservative are made of cinnamon bark oil, lactic acid and Nisin.

[128] In one embodiment the formulation for pickle is Cinnamon bark oil 2-15%, clove bud oil 1-20%, Lactic acid 30-70%, citric acid 15-45% and rosemary 1-30%. A more preferred formulation is Cinnamon bark oil $7\pm2\%$, clove bud oil $5\pm1.5\%$, Lactic acid $52\pm5\%$, citric acid $26\pm3\%$ and rosemary $10\pm3\%$. The formulations for pickle are provided in the table below.

TABLE 10

INGREDIENTS	PERCENTAGE RANGE	PRECISE
Cinnamon bark oil	1-10%	5±3%
Lactic acid	20-40%	30±5%
Nisin	1-10%	5±3%

[129] The natural preservative for pickle may also have a carrier mean such as Tween 80, Hydroginated castor oil and glysrol. The ration between the active constituents of natural preservative for pickle and carrier mean is 1:4-2:3.

- [130] For liquid form of food preservative having a carrier such as gum acacia, maltodextrin or silica is disclosed. A powder form of food preservative having a carrier such as polyoxyethylenesorbitanoleate or hydrogenated castor oil is disclosed.
- [131] A method of preventing or inhibiting growth of growth of microbes by adding a food preservative to a food product is disclosed. The microbes that are inhibited are Bacillus species, Rhizopus species, Penicillium species, Aspergillus species, yeast, Staphylococcus species, Coliforms, Lactobacillus species, Mucorsp, Acetobacter, acetic acid bacteria, Pseudomonas species, salmonella species, Listeria species, Clostridium species, Rhizopus species or Enterococci.
- [132] In another embodiment, in to a mechanical stirrer cinnamon bark oil 35g, clove bud oil 35g, rosemary extract 35g, oregano extract 35g, and citric acid 40g is taken along with the excipients Polyoxyethylenesorbitanoleate350g, gum acacia 50g, and hydrogenated castor oil 420g is to be taken and mixed well to obtain 1000g of homogenised liquid blend of natural preservative for raw meat.
- [133] For 100g of fresh meat about 0.3g of natural preservative is taken, the preservative is poured on to the meat and it is applied all over the surface of the meat to cover it up. This way the meat is marinated with the natural preservative. The marinated meat is kept in standard cold storage. The meat is tested after 30days, there is no spoilage is observed, no variation in colour is observed and on preparation no change in taste is observed.
- [134] In a ribbon mixture about cinnamon bark oil 4g, clove bud oil 1.5g, encapsulated cinnamon leaf oil 20g, lactic acid 400g, is taken along with the excipients sugar 120g, wheat fibre 29g, and silica 425.5g are taken and mixed till a homogenised powdered mixture is not obtained. About 1000g of powder natural preservative is obtained.
- [135] About 2g of powder natural preservative for bread is mixed with 1000g of bread dough that is about 0.2% preservative in the dough. The preservative is added before the baking and it

is mixed well to spread out evenly in the dough. Finally the bread is baked and kept for observation. The bread is tested after 15days, there is no spoilage observed. Also, there is any change in the texture or taste of the bread even after 15 days.

- [136] In a ribbon mixture about cinnamon bark oil 5g, clove bud oil 1g, cinnamon leaf oil 3g, lactic acid 350g, is taken along with the excipients sugar 115g, maltodextrin 136g, wheat fibre 40g, and silica 350g are taken and mixed till a homogenised powdered mixture is obtained. About 1000g of powder natural preservative is obtained.
- [137] The natural preservative makes about 0.2% of the cake batter, about 2g of powder natural preservative for cake is mixed along with sugar and fat in to the cake batter. The final weight of the cake batter is about 1000g. Finally the cake is baked and kept for observation. After 15 day the cake is tested, there is not spoilage observed for about 15 days. Also, there is no change in the taste of the cake even after 15 days.
- [138] In a mechanical stirrer, cinnamon bark oil 60g, encapsulated cinnamon leaf oil 80g, lactic acid 300g, citric acid 50g, cassia extract 3g mixed along with excipients sugar 200g, Polyoxyethylenesorbitanoleate18.5g and hydrogenated castor oil 122g are taken and mixed well to obtain a homogenised liquid blend. About 1000g of liquid natural preservative is obtained. In case there are any solid partials left the preservative can be passed through a ball mill.
- [139] To fresh fruit juice about 50ppm natural preservative for beverages is added and mixed well. The Juice is kept for observation, after 28 day the juice is tested. There is no sign of spoilage even after 28 days and no alteration in smell or taste is observed when tested after 28 days.
- [140] In a mechanical stirrer cinnamon bark oil 20g, rosemary extract 200g, natamycin 80gm, and Polyoxyethylenesorbitanoleate 700g are taken and mixed well till homogenised mixture is not obtained. A 1000g homogenised liquid preservative mixture is obtained.
- [141] In to freshly prepared 100g mayonnaise about 0.2g of liquid preservative is added and mixed well. The mayonnaise is kept for observation in standard storage for 6 months. After 6 months there is not spoilage observed, no alteration in taste or texture is observed.

[142] In a ribbon mixture clove bud oil 30g, lactic acid 250g, citric acid 193g, cardamom extract 7g, natamycin 70g, maltodextrin 200g, and silica 250g is taken and mixed well. About 1000g of uniformly mixed powder preservative formulation for fruit jam is obtained.

- [143] In to freshly prepared 100g fruit jam about 0.2g of liquid preservative is added and mixed well. The jam is kept for observation in standard storage for 6 months. After 6 months the jam sample is tested there is no spoilage observed, no alteration in taste, colour or texture is observed.
- [144] In a ribbon mixture clove bud oil 20g, Lactic acid 150g, citric acid 190g, ascorbic acid 50g, cumin seed extract 18.5g, rosemary extract 1.5g, along with natamycin 150g and nisin 150g, and maltodextrin 270g are taken and mixed till a uniformly blend powdered formulation is not obtained. About 1000g of natural preservative for been dip is obtained.
- [145] In to freshly prepared 100g bean dip about 0.02g of liquid preservative is added and mixed well. The bean dip is kept for observation in standard storage for 12 days. After 12 days the bean dip sample is tested, there is no spoilage observed, no alteration in taste, colour or texture is observed.
- [146] In a ribbon mixture cinnamon bark oil 30g, lactic acid 240g, nisin 50g, sugar 30g, maltodextrin 20g, and silicon 45g are taken together and mixed well till a uniformly blended mixture is not formed. About 1000g natural preservative for dairy product is obtained.
- [147] In to freshly prepared 100g condensed milk about 0.2g of the preservative is added and mixed well. The condensed milk is kept for observation in standard storage for 30 days. After 30 days the product is tested and there is no spoilage observed and no alteration in taste, colour or texture is observed.
- [148] In a mechanical stirrer, cinnamon bark oil 15g, clove bud oil 10g, lactic acid 100g, citric acid 50g, rosemary extract 18g, Polyoxyethylenesorbitanoleate 667g, silica 20g, and glysrol 120g is to be taken and mixed well to get a homogenised solution. About 1000g liquid preservative for pickle will be obtained.
- [149] In to freshly prepared 100g raw mango pickle about 0.2g of the preservative is added and mixed well. The pickle is kept for observation in standard storage for 1 year. After a year the

product is tested and there is no spoilage observed and no alteration in taste or texture is observed.

[150] A method of inhibiting growth of microbes by adding the natural food preservative to a food product infested with microbes such as Bacillus species, Rhizopus species, Penicillium species, Aspergillus species, yeast, Staphylococcus species, Coliforms, Lactobacillus species, Mucorsp, Acetobacter, acetic acid bacteria, Pseudomonas species, salmonella species, Listeria species, Clostridium species, Rhizopus species and Enterococci.

[151] In another embodiment a method of preventing growth of microbes by adding the natural food preservative to a food product. The microbes that are prevented grouth on a food article are selected from a group consisting of Bacillus species, Rhizopus species, Penicillium species, Aspergillus species, yeast, Staphylococcus species, Coliforms, Lactobacillus species, Mucorsp, Acetobacter, acetic acid bacteria, Pseudomonas species, salmonella species, Listeria species, Clostridium species, Rhizopus species and Enterococci.

[152] It will be readily understood by the skilled artisan that numerous alterations may be made to the examples and instructions given herein. These and other objects and features of present invention will be made apparent from the following examples. The following examples as described are not intended to be construed as limiting the scope of the present invention.

EXAMPLES

EXAMPLE 1A: METHOD TO MAKE A FORMULATION OF THE NATURAL PRESERVATIVE FOR BEVERAGES.

[153] In a blender, 6g of Cinnamon bark oil and 0.3g of Cassia bark oil were added, and the mix was blended well. Citric acid, 5g, was dissolved in 30g HR-40 (hydrogenated castor oil) and kept at 50°C. The resultant Citric acid dissolved in HR 40 was added into the blender in which Cinnamon bark oil and Cassia bark oil were mixed. The cinnamon bark oil, cassia bark oil and citric acid were in the ratio between 5:2:2 to 2:2:1. Into the mix, 45g tween 80 was added and dissolved it to form a uniform blend. Tween 80 and HR40 were used as surfactants, the quantity of which can be varied according to the requirement of a beverage, and a person skilled in the art

can use other surfactants according to their needs. The mix then transferred to a homogenizer and homogenised to obtain uniformly blended homogenised liquid product.

[154] The homogenised liquid mixture obtained was the natural preservative suited for beverages. Adding 0.075% of the natural preservative in a fruit juice can protect the juice from spoilage for over 28days.

EXAMPLE 1B: METHOD TO MAKE A NATURAL PRESERVATIVE FORMULATION FOR CAKE.

[155] In a blender, 0.5g of cinnamon bark oil, 0.3 cinnamon leaf oil, 0.2g of clove bud oil and 35g lactic acid were added, the mixture was blended well. In another vessel 13.5g Maltodextrin, 4g of Wheat fibre and 35 grams of Silica was blended with 11.5g sugar. The solid constituents of the mixture were mixed well and powdered in a pulveriser. The cinnamon bark oil and clove bud oil mix were taken into an agitating vessel and, into the agitating vessel the powdered solid constituents mixture was added at a steady rate. The mixture in the vessel agitated continuously to avoid any lumps. The mixture was again pulverised to get a fine powdered product. The final powdered product was the natural preservative for Cake.

[156] The natural preservative can be added at 2500ppm by weight into a cake batter before baking. The preservative will protect the cake from spoilage for over 15 days.

EXAMPLE 1C: METHOD TO MAKE A NATURAL PRESERVATIVE FORMULATION FOR CHICKEN.

[157] Cinnamon Bark Oil 3g, rosemary extract 3g, clove bud oil 3gand Oregano extract 3g were all taken in a blender and mixed well. In a different vessel, citric acid crystals 3.5g were mixed with 15g of gum acacia, 30g of Maltodextrin and 38g of silica. The solid mixture was made into a fine powder in a pulveriser. The liquid constituent mixed in the blender was taken into an agitating vessel, and the powdered solid constituent mixture was added at a steady rate into the agitating vessel. The mixture in the vessel was agitated continuously to avoid any lumps. The mixture was again pulverised to get a fine powder product. The final powdered product was the natural preservative for raw chicken.

EXAMPLE 1D: METHOD TO MAKE A NATURAL PRESERVATIVE FORMULATION FOR BREAD.

[158] Cinnamon Bark Oil 0.4g and clove bud oil 0.15g were taken into a blender along with 40g lactic acid and mixed well. Sugar about 12g along with 2.9g wheat fibre and 42.55g silica was mixed well in a vessel and powdered well in a pulveriser. The Cinnamon Bark and clove bud oil were taken into an agitating vessel, and the finely powdered mixture of sugar, silica and wheat fibre was added at a steady rate into the vessel. The mixture was agitated well to make dry powder. The mixture was again pulverised to form a fine powder. Into the finely powdered mixture, 2g of encapsulated cinnamon leaf powder was mixed to get the final formulation.

EXAMPLE 1E: METHOD TO MAKE A NATURAL PRESERVATIVE FORMULATION FOR HUMMUS.

[159] Citric acid 25g was mixed with 8.5g ascorbic acid crystals in a vessel and mixed. The mixture was pulverised to get a fine powder. In a different agitating vessel 1 gram of rosemary extract, 5g of clove bud extract,6.3g of Lactic acid and 2.5g of cumin seed extract was taken. Into the agitating vessel, the powdered citric acid and the ascorbic acid mixture was added along with maltodextrin. The mixture in the vessel was continuously agitated to avoid the formation of any lump. maltodextrin was added to the mixture till a powdered formulation was obtained. About 40 to 58g maltodextrin was used in the present illustration. The final product was again subjected to pulverisation to obtain homogenised fine powdered product.

EXAMPLE 2: SHELF LIFE STUDY CONDUCTED FOR FOOD PRODUCT HAVING THE NATURAL PRESERVATIVE.

[160] Sensory analysis was done in two parts. Firstly, sensory analysis was done to determine the effect of added natural preservatives on colour, taste, aroma and overall acceptability of raw meat as compared to control sample. Here, a control sample is the one in which no preservative has been added. For this analysis, difference control test was employed on a scale of 0-4 with 0 corresponding to no difference from control to 4 which corresponds to a huge difference. Secondly, the sensory analysis was done daily for all the samples to determine any fungal spoilage visually. Also, the sample was also looked for a change in smell and taste to determine its sensory shelf-life.

[161] A specific weight of the solid sample was homogenised aseptically in nine volumes of sterile appropriate diluents to get a homogenous suspension of microorganisms. The liquid sample was directly used as homogeneous suspension of microorganisms. The suspension of microorganisms so obtained was diluted serially (10 times, 100 times, 1000 times, and so on). Here 10⁻¹, 10⁻², 10⁻³, etc. were called dilutions. Their reciprocals (10¹, 10², 10³, and so on) were called dilution factors. A definite volume of the suspension of microorganisms forms each dilution was inoculated onto agar plates and spread properly, to space the individual microbial cells wide apart and isolate them from each other. Culture media used for this evaluation were Nutrient Agar (NA), Plate Count Agar (PCA), Sabouraud Dextrose agar (SDA), Potato Dextrose agar (PDA) and Rose Bengal Chloramphenicol Agar (RBA).

[162] Incubate the bacterial agar plates at 37°C for 24h and fungal agar plates at 27°C for 48-72h. The total plate count (TPC) in the sample was calculated by multiplying the number of CFUs (Colony-Forming Unit) with the respective dilution factors.

[163] Colonies from the primary plates were aseptically picked with a sterile wire loop and transferred on to freshly prepared sterile Nutrient Agar plate (bacteria) SDA (fungi), with streaking technique such that discrete colonies appear at the end of streaked lines after incubation. The subculture plates were incubated at 37°C for 24 to 48h for bacteria and 27°C for 48 to 72 h for fungi. Discrete colonies from the subculture plates were aseptically transferred and streaked on slants and incubated for another 24 h at 37°C for bacteria and 27°C for 48 to 72h for fungi. The identification of Bacteria and Fungi was based on the morphological (Gram staining & Motility) and biochemical characteristics.

[164] A challenge test was prepared. "Challenge test" is a microbiological evaluation of a preservative ability to kill or prevent the growth of microorganisms over a relatively extended period of a particular product. In this test, the product was evaluated for the presence of pathogens after manufacturing. The product was separated out into five different containers. Each container was inoculated with a different microorganism at a relatively high concentration, and the initial "challenge" concentration was found (10⁶ to 10⁷cfu/g of product). The product was incubated at approximately room temperature for an extended period (usually at least seven days). The product was re-evaluated for the presence of the microorganisms that were inoculated into it.

[165] The identification of Bacteria and Fungi was based on the morphological (Gram staining, Motility) and biochemical characteristics.

[166] Characterization and identification of the colony isolates were achieved by initial morphological examination of the colonies in the plate (macroscopically) for colonial appearance, size, elevation, form, edge, consistency, colour, odour, opacity, haemolysis and pigmentation and the results were recorded.

[167] Gram's staining from the colonies provided a preliminary identification of the pathogenic bacteria. Fungal moulds and yeasts were identified by performing Lactophenol cotton blue staining.

[168] Biochemical characterisation of the bacteria was done by performing specific tests such as catalase, oxidase, TSI, Indole, Methyl red, VogesProskauer and citrate tests, carbohydrate fermentation tests, nitrate tests, growth at 42°C, coagulase, DNAasetests, mannitol fermentation, O/F tests and sensitivity to Novobiocin, bacitracin and furazolidone. Identification of yeast was made using Corn meal agar morphology.

[169] TBC: Total bacterial count.

[170] TFC: Total fungal count.

[171] TNTC: Too Numerous To Count.

[172] NPB: Natural preservative for beverages.

EXAMPLE 2A: STUDY ON THE SHELF LIFE OF BEVERAGE USING THE NATURAL PRESERVATIVE FOR BEVERAGES (NPB).

[173] Natural preservative for beverages (NPB) utilized for this illustration was prepared as per example 1A. The NPB is mixed with fruit juice at different dosages, i.e. at 0.05%, 0.075% to 0.05% of the juice. The juice samples are tested for various parameters as explained in Example 2, and the data are provided in tables below.

[174] Organisms isolated & its percentage.

Table 11

Sample name	Organisms isolated	Percentage (%)
Fruit Juice	Lactic acid bacteria	20 to 25
	Acetic acid bacteria	10 to 15
	Bacillus sp.	30 to 35 %
	Yeast	5 to 10 %

[175] The fruit juice samples were evaluated for differences in sensory parameters as compared to control and results are shown in Table 2. It was seen from the results of the sensory analysis that there was no difference of added formulations on colour and texture of the fruit juice (as indicated by value 0 which corresponds to no difference from control). Further, sensory results of taste and aroma showed that the added preservative had no significant effect on these parameters at a concentration level of 0.1%.

[176] Effect of adding Natural Preservatives on Sensory Properties of Fruit juice.

Table 12

Sample Name	Colour	Taste	Aroma	Overall acceptability
Fruit juice Control	0	0	0	Acceptable
Fruit juice + NPB at 0.05%	0	0	0	Acceptable
Fruit juice + NPB at 0.075%	0	0	0	Acceptable
Fruit juice + NPB at 0.05%	0	0	0	Acceptable

[177] The scale used: 0 - no difference, 1 - a Very slight difference, 2 - a Moderate difference, 3 - Large difference, 4 - huge difference.

[178] Decrease in the microbial count in Fruit juice sample during challenge study.

Table 13

Sample	1 St	day	7 th	7 th day 14 th day		21 st	day	28 th day		
name	TBC	TFC	TBC	TFC	TBC	TFC	TBC	TFC	TBC	TFC
control	TNTC	TNTC	TNTC	TNTC	TNTC	TNTC	TNTC	TNTC	TNTC	TNTC
NPB at 0.05%	230	00	90	00	15	00	05	00	07	00
NPB at 0.075%	60	00	30	00	09	00	03	00	00	00
NPB at 0.1%	20	00	00	00	00	00	00	00	00	00

[179] From the above results, it was apparent that NPB at 0.075% was working against microbes present in fruit juice even on the 28th day. In challenge study, 1 to 3 log reductions in bacteria from the initial level should occur in one to two weeks with no further increase in microbes after that at 28 days, for yeast and mould no increase from the Inoculum level was permitted at all sampling intervals for three-month shelf life products.

EXAMPLE 2B: SHELF LIFE STUDY FOR BREAD USING THE NATURAL PRESERVATIVE FOR BAKED (BREAD) (NPBB).

[180] Natural preservative for baked (bread) NPBB used for this illustration was prepared as per example 1D. The NPBB is mixed with the dough at different dosages, i.e. at 0.25%, 0.1% by weight of the dough. The final baked bread samples are tested for various parameters as explained in Example 2, and the data are provided in tables below.

[181] The test groups:

- [182] The natural preservative formulation from example 1C was employed at dosage 2500ppm and 1000ppm by weight of the dough.
- [183] One loaf of bread was prepared without any preservatives.
- [184] Effect of Adding Natural Preservatives on Sensory Properties of Bread.

Table 14

	applied					acceptability
Breads Without preservative	-	0	0	0	0	Acceptable
Bread with natural preservative	0.1%	0	0	0	0	Acceptable
Bread with natural preservative	0.25%	0	0	0	0	Acceptable

[185] The scale used: 0 - no difference, 1 - a Very slight difference, 2 - a Moderate difference, 3 - a Large difference, 4 - huge difference.

[186] A total of four isolates were identified as Bacillus sp., Rhizopus sp., Penicillium sp. and Mucor sp. The isolates were subjected to grow on selective agar media. Isolates when Gram stained, found rod shaped, long and positive in Gram reaction those all are typical characteristics of Bacillus sp.

[187] Identification of Spoilage By Microbes From Bread Samples.

Table 15

	Mic	robes identified
	Bacteria	Yeast/mould
Bread Without preservative	Bacillus sp.	Rhizopussp, Penicillium sp., Mucor sp.
Bread with synthetic preservative	Bacillus sp.	Penicillium sp.
Bread with natural preservative	Bacillus sp.	Rhizopus sp.

[188] Effect of Adding Natural Preservatives on Shelf Life Of Bread.

Table 16

Sample name	Dosage applied	Visible mould growth	Shelf life
Without preservative	-	Visible mould growth on 4th day	3 days
With synthetic preservative	0.1%	Visible mould growth on 7th day	6 days
Bread with natural	0.25%	Visible mould growth on 11th day	10 days

preservative			
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[189] Microbiological Parameters Total Plate Count (cfu/gm)).

Table 17

Table 17												
	1 st I	Day	3 rd Day		5 th]	Day	7 th Day		11 th Day			
Sample name	TFC	TBC	TFC	TBC	TFC	TBC	TFC	TBC	TFC	TBC		
Without preservative	00	00	00	00	TNT C	TNTC	-	-	-	-		
With natural Preservative 0.1%	00	00	00	00	00	00	37	00	139	11		
With natural Preservative 0.25%	00	00	00	00	00	00	00	00	11	9		

[190] The prepared Bread samples (Control sample and preservative added samples) were analysed for microbiological parameters and appearance of mould growth, results provided in Table 7. Microorganisms isolated and identified from the samples are presented in Table 5. After adding natural preservative in bread, the shelf life increases as compared to control sample. Results are given in Table 6.

[191] From analytical, sensory, and shelf life study natural preservative blend was a proven product for preserving bread. It was seen from the results of the Microbiological analysis that there was an increase in the shelf of Bread as compared to control sample.

EXAMPLE 2C: SHELF LIFE STUDY FOR BREAD USING NATURAL PRESERVATIVE FOR BAKED (CAKE).

[192] To study the effect of natural preservatives from example, 1B on the shelf-life extension of the range of cakes (Tea, Marble, Sponge, Carrot and Banana cake) is provided.

[193] The essential ingredients of all the cake are flour, sugar, margarine, butter, eggs and flavour. In addition to these ingredients, fresh carrot and banana were added in carrot and banana cake. All the ingredients were mixed in a mechanical mixer. The batter was then filled in round tin moulds and kept for baking. After baking, the cake was allowed to cool and then packed.

- [194] Tea cake (with synthetic preservative)
- [195] Tea cake (without preservative)
- [196] Tea cake (with natural preservative)
- [197] Fruit cake (with synthetic preservative)
- [198] Fruit cake (without preservative)
- [199] Fruit cake (with natural preservative)
- [200] The natural preservative formulation isemployed at a dosage 2500ppm by weight of the batter.
- [201] The synthetic preservative (Potassium sorbate) isemployed at a dosage 1500ppm by weight of the batter.
- [202] A mould of cake was prepared without any preservative.
- [203] The prepared tea cake and fruit cake samples were evaluated for differences in sensory parameters as compared to control and results are shown in Table 8. It was seen from the results of the sensory analysis that there was no difference of added formulations on the colour of the tea cake and fruit cake. Further, sensory results of taste and aroma showed that the added preservative had no significant effect on these parameters at a concentration level of 0.25%.
- [204] Further, all the cake samples were evaluated daily until spoiled for change in colour, the appearance of any visible growth and off odour. It was found that the control cake had visible dark green growth on the 3rd day of storage day. Also, the sample started to give off-odor from 4th day onwards. However, the samples with added natural preservatives showed neither any visible growth nor any off-odour till 9th day of storage.
- [205] Effect of Adding Natural Preservatives on Sensory Properties of Cake.

Table 18

Sample Name	Dosage applied	Colour	Taste	Aroma	Texture	Over all acceptability
Tea cake without preservative	0	0	0	0	0	Acceptable

Tea cake with Synthetic pre- servative	0.15%	0	0	0	0	Acceptable
Tea cake with natural preservative	0.25%	0	0	0	0	Acceptable
Fruit cake without preservative	-	0	0	0	0	Acceptable
Fruit cake with synthetic preservative	0.15%	0	0	0	0	Acceptable
Fruit cake with natural preservative	0.25%	0	0	0	0	Acceptable

[206] The scale used: 0 - no difference, 1 - a Very slight difference, 2 - a Moderate difference, 3 - a Large difference, 4 - huge difference.

[207] It was observed earlier that shelf-life in tea cake was limited to 3-4 days as after that microbial growth occurs in tea cake (as seen from table 9). To study the effect of added formulations on microbial load in black tea cake, Total plate count (bacteria) and fungal count for tea cake were evaluated daily until samples spoiled using plate method. The results of the bacterial and fungal count are shown in table 9a and 9b. It can be seen from the table 10 that no bacterial spoilage was observed in cake samples during 15 days storage. However, fungal decay started in control cake on the 3rd day of storage as seen from table 9a and 9b.

[208] Table 9a and Table 9b show the effect of different levels of preservatives on the shelf life of cake samples at different time intervals.

[209] Microbiological Parameters (Total Plate Count (cfu/gm)) in Tea cake.

Table 19a

	2nd Day 6th D		Day 9th I		Day 11th		Day 13th		Day	
Sample name	TFC	TBC	TFC	TBC	TFC	TBC	TFC	TBC	TFC	TBC

Control	26	00	>250	TNTC						
natural pre- servative 0.15%	00	00	00	00	06	00	24	09	71	00
natural preservative 0.25%	00	00	00	00	00	00	52	00	60	11

[210] Microbiological Parameters (Total Plate Count (cfu/gm)) in fruit cake.

Table 9b

Sample name	2nd	Day	6th Day		9th D	ay	11th	Day	13th Day	
	TFC	TBC	TFC	TBC	TFC		TFC	TBC	TFC	TBC
Control	00	00	36	28	TNTC	43	TNTC	TNTC	TNTC	TNTC
natural preservative @ 0.15%	00	00	00	04	80	16	130	TNTC	220	TNTC
natural preservative @ 0.25%	00	00	00	00	00	76	14	27	120	54

[211] Visible fungal growth study on the cake.

Table 20

Sample name	Dosage applied	Visible mould growth	Shelf life
	r	Геа Cake	
Without preservative	-	Visible mould growth on 3rd day	4days
With natural preservative	0.15%	Visible mould growth on 12th day	11days
With natural preservative	0.25%	Visible mould growth on 16th day	15days
	I	Fruit cake	
Without preservative	-	Visible mould growth on 3rd day	4 Days
With natural preservative	0.15%	Visible mould growth on 10th day	9days
With natural preservative	0.25%	Visible mould growth on 12th day	11days

[212] Results showed that the addition of natural preservative at levels of 0.25% controls the microbial growth in the cake sample without effecting the flavor of the cake.

EXAMPLE 2D: SHELF LIFE STUDY FOR BREAD USING THE NATURAL PRESERVATIVE FOR RAW MEAT (NPRM).

- [213] About 100 grams of meat samples were collected in clean, dry and sterile polythene bags and transported to the laboratory for microbiological analysis within one hour.
- [214] The samples were aseptically cut into thin smaller pieces using a sterile knife. The analytical portions were placed in separate sterile plastic bags to which 250 ml of buffered peptone water was added. The bags were shaken vigorously, and the sample rinsate was collected.
- [215] Applied two different dosages (0.3%, and 0.5%) of preservative from example 1C in raw meat and Shelf life study carried out at different time intervals.
- [216] Initial Total Plate Count (cfu/g).

Table 21

14010 21								
Sample name	TBC	TFC						
Sample 1	$>10^{5}$	10^{3}						
Sample 2	>10 ⁶	10^{3}						
Sample 3	>10 ⁵	10^{3}						

[217] Organisms isolated & its percentage.

Table 22

	1 4010 22	
Sample name	Organisms isolated	Percentage
	Staphylococcus sp.	10 to 15 %
Sample 1	E.Coli	20 to 25 %
Sample 1	Yeast	30 to 35 %
	Bacillus sp.	5 to 10 %
	Staphylococcus sp.	15 to 20 %
Sample 2	E.coli	20 to 25 %
Sample 2	Yeast	30 to 40 %
	Bacillus sp	10 to 15 %
	Staphylococcus sp.	30 to 40 %
Sample 3	Yeast	20 to 25 %
_	E.coli	10 to 15 %

[218] Effect of Adding Natural Preservatives on Sensory Properties of meat (after cooking).

[219] The cooked meat samples were evaluated for differences in sensory parameters as compared to control and results are shown in Table 13. It was seen from the results of the sensory analysis that there was no difference of added formulations on colour and texture of the meat before and after cooking (as indicated by value 0 which corresponds to no difference from control). Further, sensory results of taste and aroma showed that the added preservative had no significant effect on these parameters at a concentration level of 0.3%.

[220] Effect of Adding Natural Preservatives on Sensory Properties of meat (after cooking).

Table 23

Sample Name	Colour	Taste	Aroma	Texture	Overall acceptability						
Meat Control	0	0	0	0	Acceptable						
Meat + NPRM at 0.3%	0	0	0	0	Acceptable						
Meat + NPRM at 0.5%	0	1	0	0	Acceptable						

[221] The scale used: 0 - no difference, 1 - a Very slight difference, 2 - a Moderate difference, 3 –Large difference, 4 - huge difference.

[222] After adding natural preservative in meat, the shelf life increases as compared to control sample; Day 1, 7 and 14.

Table 24a

Sample	1st day		7th day		14th day	
name	TBC	TFC	TBC	TFC	TBC	TFC
Sample	TNTC	TNTC	TNTC	TNTC	TNTC	TNTC
1(control)						
0.3%	260	00	140	00	120	00
0.5%	220	00	70	00	47	00
Sample	TNTC	TNTC	TNTC	TNTC	TNTC	TNTC
2(control)						
0.3%	290	00	200	00	170	00
0.570	200		200		170	
0.5%	160	00	80	00	65	00
0.570	100	00	00	00	03	

Sample 3(control)	TNTC	TNTC	TNTC	TNTC	TNTC	TNTC
0.3%	190	00	120	00	100	00
0.5%	130	00	69	00	40	00

[223] After adding natural preservative in meat, the shelf life increases as compared to control sample; Day 21 and 28.

Table 24b

		21st	28th day			
Samplename	TBC	TFC	TBC	TFC		
Sample1(control)	TNTC	TNTC	TNTC	TNTC		
0.3%	250	00	350	00		
0.5%	90	00	140	00		
Sample2(control)	TNTC	TNTC	TNTC	TNTC		
0.3%	>230	00	>250	00		
0.5%	130	00	190	00		
Sample3(control)	TNTC	TNTC	TNTC	TNTC		
0.3%	170	00	>250	00		
0.5%	140	00	>250	00		

[224] It was seen from the results of the Microbiological analysis that there was an increase in the shelf of Meat as compared to control sample. Additions of our preservative blends extend the shelf life of meat at chilled condition up to 15 days. During the procedure, meat was vulnerable to cross contamination from water, utensils and the environment. These conditions limit the shelf life of raw meat.

EXAMPLE 2E: NATURAL PRESERVATIVE FOR TESTING SHELF LIFE OF HUMMUS (NPH)

[225] To study Microbial spoilage mechanisms in Hummus sample.

[226] Hummus is considered as one of the most popular traditional foods in Middle East countries. It is prepared from dried chickpeas (Cicer arietinum L.) and tahini (an oily viscous fluid obtained by milling de-hulled roasted white sesame seeds). Hummus is made from 20-25% of boiled chickpea, lemon juice or citric acid, garlic and salt. The processing steps for hummus preparation include chickpeas soaking in water for overnight, then boiling, followed by blending with tahini and other ingredients to obtain the underlying smooth Chickpea mix.

- [227] The natural preservative formulation isemployed at two dosages viz. 3000ppm and 5000ppm by weight of the Final product.
- [228] The synthetic preservative (potassium sorbate) was used at one dosage 2000ppm by weight of the Final product.
- [229] Hummus prepared without any preservatives.
- [230] The prepared Hummus samples evaluated for differences in sensory parameters as compared to control and results shown in Table 15. From the results of the sensory analysis showed that there was no difference of added formulations on colour and texture of the Hummus. Further, sensory results of taste and aroma showed that the added preservative had no significant effect on these parameters at a concentration level of 0.5%.
- [231] Effect of Adding Natural Preservatives on Sensory Properties Of Hummus.

Table 25

	Sample Name	colour	Taste	Aroma	Texture	Overall acceptability
Hummus without preservative	-	0	0	0	0	Acceptable
Hummus with natural preservative	0.3%	0	0	0	0	Acceptable
Hummus with natural preservative	0.5%	0	0	0	0	Acceptable

- [232] The scale used: 0 no difference, 1 a Very slight difference, 2 a Moderate difference, 3 a Large difference, 4 huge difference.
- [233] Effect of Natural Preservatives on the Shelf life of Hummus.

[234] Table 16 shows the effect of different levels of preservatives on the shelf life of hummus at various time intervals. Results demonstrated that the addition of our natural preservative at levels of 0.3% and 0.5% control the microbial growth in the hummus sample.

[235] Microbiological Parameters (Total Plate Count (cfu/gm)).

Table 26

Sample	7 th day		9 th day		day	11 th	14 th day		
name	TBC	TFC	TBC	TFC	TBC	TFC	TBC	TFC	
Control	LINIC	INIC	TNTC	TNTC	INIC	INIC	TNTC	TNTC	
0.3%	00	00	06	00	17	07	84	21	
0.5%	00	00	00	00	12	11	56	18	

[236] Additions of natural preservative blends extend the shelf life of hummus at refrigerator temperature. Lactobacillus sp. and yeasts were the predominant microorganisms that caused spoilage in Hummus. During the preparation procedure, hummus is vulnerable to cross contamination from ingredients, utensils and the environment. Except for boiling of the chickpeas, there is no heat treatment during preparation. These conditions limit the shelf life of hummus.

EXAMPLE 3: STUDY TO DETERMINE SYNERGY FOR THE FORMULATIONS: MIC AND ZOI ARE TESTED FOR INDIVIDUAL COMPONENTS FROM GROUP A AND THEIR VARIOUS FORMULATIONS.

[237] Antimicrobial efficacy study, cinnamon bark oil, citric acid, rosemary extract and clove bud oil, against ten well-known pathogens responsible for food spoilage.

[238] Cinnamon bark oil, citric acid, and clove bud oil were taken for antimicrobial study. 100mg oil was dissolved in 1ml of 60% DMSO and kept it as a stock solution.

[239] The antimicrobials present in the plant extract were allowed to diffuse out into the medium and interact in a plate freshly seeded with the test organisms. The resulting zone of inhibition is uniformly circular as there is a confluent lawn of growth. The diameter of ZOI can be measured in millimetres.

[240] Petri plates containing 20ml Muller Hinton medium were seeded with a 24hr culture of the bacterial strain. 5mm diameter wells were cut, and 50µl, 100µl & 150µl of the plant extract, negative control and positive control were added. The plates were then incubated at 37°C for 24h. The antibacterial activity was assayed by measuring the diameter of the inhibition zone formed around the well.

- [241] Antibacterial activity was further characterised by determining whether bacteriostatic or bactericidal. The test was performed by swabbing of the growth inhibition zone. The swab was streaked onto Nutrient agar plates and incubated aerobically at 37°C for 24hrs. The presence of growth in nutrient agar plate was interrupted as an inhibitory activity i.e. bacteriostatic while no growth was interrupted as bactericidal.
- [242] MIC -Agar dilution method assays:
- [243] Evaluation of the antimicrobial activity of plants extracts was conducted according to the agar dilution method.
- [244] Inoculation preparation:
- [245] At least four well-isolated colonies of the same type from a culture agar plate were selected and touched the top of the colony with a loop and transferred to a tube containing 4 ml of a suitable broth such as nutrient broth. The suspension was incubated at 37°C, and the size was adjusted to the 0.5 McFarland standard turbidity (1.5x 10° cfu/ml).
- [246] Preparation of antimicrobial plates:
- [247] The diluted extracts were added to the melted and cooled medium in a ratio of 1 part extract sample agent to 9 parts medium (2 ml of plant extract to 18 ml of Mueller Hinton agar for each petri dishes plate) with most susceptibility test.
- [248] Inoculation of test organisms:
- [249] Fill each well of multiple-inoculator with inoculums test organisms and dip the tip of multiple-inoculator on Mueller Hinton Agar plates and incubate at 37°C for 24 hr. At least three repetitions were run for each assay.

[250] Minimum inhibitory concentration (MIC) and Minimum bactericidal concentration (MBC):

- [251] The MIC value of the extract was determined as the lowest concentration that completely inhibited bacterial growth after 48 hr of incubation at 37°C.
- [252] For the determination of MBC, a portion of liquid (5 µl) from each plate well that exhibited no growth were taken and then incubating at 37°C for 24 hr. The lowest concentration that revealed no visible bacterial growth after sub-culturing was taken as MBC. Positive and negative cultures were also prepared.
- [253] The antimicrobial activity of cinnamon bark oil, citric acid, and clove bud oil, against microbes, was investigated by well dilution test method. The results of inhibition zone were recorded in Table 17a and 17b and MIC values in Table 18a and 18b. Cinnamon bark oil showed inhibitory activity against all the tested organisms. Citric acid also showed slight inhibitory activity.

[254] Antimicrobial Activity measured as ZOI (mm).

Table 27a

	Organisms	Cinnamon bark oil		Clove bud oil		Citric acid		Rosemary	
		100 ul/ml	150 ul/ml	100 ul/ml	150 ul/ml	500 ul/ml	1000 ul/ml	500 ul/ml	1000 ul/ml
1	Staphylococcus aureus	20	26	26	31	9	22	9	12
2	Streptococcus pyogens	17	24	10	14	12	24	7	14
3	Pseudomonas aeruginosa	15	23	nil	nil	nil	12	nil	nil
4	Klebsiella pneumoniae	14	23	nil	10	nil	nil	nil	nil
5	Proteus	9	14	nil	10	-	-	nil	nil
6	Enterobacter	16	24	9	14	-	16	nil	6
7	Enterococcus	25	41	11	16	-	-	nil	nil
8	E.Coli	19	21	10	15	-	-	nil	nil
9	Lactobacillus sp.	23	37	10	16	-	1	nil	nil

10	Candida albicans	27	49	26	>30	23	35	nil	nil
1	aioicans			l		l			

[255] ZOI for the formulation along with expected ZOI for the formulation of cinnamon bark oil, clove bud oil and citric acid.

Table 27b

		ric acid 100ul/ml					
		1:1	1:2	5:4:	:10	1:1:4	
	Organisms	Expected value	Actual value	Expected	Actual value	Expected value	Actual
1	Staphylococcus aureus	12.4	30	11.56	32	8.879	27
2	Streptococcus pyogens	7.95	26	7.72	28	6.105	24
3	Pseudomonas aeruginosa	3.75	17	3.9	20	2.505	14
4	Klebsiella pneumoniae	3.5	19	3.64	22	2.338	16
5	Proteus	2.25	14	2.34	15	1.503	9
6	Enterobacter	6.25	21	6.05	25	4.175	17
7	Enterococcus	9	27	8.81	11	6.012	21
8	E.Coli	7.25	22	7.04	25	4.843	19
9	Lactobacillussp	8.25	27	8.08	31	5.511	22
1 0	Candida albicans	15.55	32	14.78	34	11.91	28

[256] ZOI for the formulation along with expected ZOI for the formulation of cinnamon bark oil, clove bud oil and citric acid.

Table 27c

	Organisms	cinnamon bark oil: clove bud oil: rosemary 100ul/ml						
		1:1:1		1:1:4		1:1:6		
		Expected value	Actual value	Expected	Actual value	Expected	Actual value	
1	Staphylococcus aureus	16	25	18	32	9	32	

2	Streptococcus	9.5	15	11	18	6.	20
3	Pseudomonas aeruginosa	5	14	5	10	2.	8
4	Klebsiella pneumoniae	4.7	10	5	9	2	7
5	Proteus	3	14	6	12	1	6
6	Enterobacter	8.3	21	8.	13	4	13
7	Enterococcus	12	27	12	19	6	19
8	E.Coli	9.7	22	9	15	5	15
9	Lactobacillussp .	11	27	11	18	5	18
1	Candida	17.7	28	17	29	12	28
0	albicans						

[257] Antimicrobial Activity measured as MIC (mg/ml).

Table 28a

Name of organisms	Name of sample MIC (ul/ml)						
	Cinnamon bark oil	clove bud oil	Citric acid	Rosemary			
Staphylococcus aureus	75	75	200	250			
Streptococcus pyogens	75	>200	125	125			
Pseudomonas aeruginosa	70	>500	>250	1000			
Klebsiella pneumoniae	70	>200	>250	-			
Proteus	170	>500	>250	-			
Enterobacter	85	250	>250	-			
Enterococcus	25	250	>250	-			
E.Coli	75	>200	>250	250			
Lactobacillus sp.	50	>200	-	-			
Candida albicans	10	50	50	50			

[258] Antimicrobial Activity measured as MIC (mg/ml) for the formulation and expected value.

Table 28b

Name of	Name of sample MIC (ul/ml)					
	cinnamon bark	oil: clove bud oil: citric acid 100ul/ml				
organisms	1:1:2	5:4:10	1:1:4			

	Expecte	Actual	Expected	Actual	Expected	Actual
	d value	value	value	value	value	value
Staphylococcus aureus	109.1	66	111.76	60	128.57	70
Streptococcus pyogens	116.5	50	114	45	119.20	54
Pseudomonas aeruginosa	164.7	75	159.09	70	185.84	75
Klebsiella pneumoniae	146.6	55	144.56	52	170.04	60
Proteus	251.9	112	245.44	102	251.23	120
Enterobacter	168.3	100	165.47	97	188.88	111
Enterococcus	76.92	55	74.22	51	100	64
E.Coli	151.9	95	150	87	174.75	97
Lactobacillus sp.	160	110	158.33	100	240	116
Candida albicans	25	46	24.358	40	30	55

[259] Antimicrobial Activity measured as MIC (mg/ml) for the formulation and expected value.

Table 28c

	Name of sample MIC (ul/ml)								
Name of	cinnamon bark oil: clove bud oil: rosemary 100ul/ml								
organisms	1:1:	1	1:1	.:4	1:1:6				
	Expected	Actual	Expected		Expected	Actual			
	value	value	value	value	value	value			
Staphylococcus aureus	134	65	140	50	157	60			
Streptococcus pyogens	102	55	119	50	120	55			
Pseudomonas aeruginosa	155	75	295	100	358	160			
Klebsiellapneum oniae	164	90	311	125	414	200			
Proteus	303	120	761	150	1014	355			
Enterobacter	190	100	380	150	507	230			
Enterococcus	68	30	136	55	182	100			
E.Coli	140	70.	174	70	189	105			

Lactobacillussp.	150	105	240	145	320	225
Candida	21	15	30	20	22	25
albicans	<u> </u>	13	30	20	33	23

[260] In Table 17b the activity of formulation made with cinnamon bark oil, clove bud oil and citric acid are recorded. The recorded values from the experimentation were also compared with the expected results in table 17b. The expected results were calculated from the reading recorded in table 17a. From the recorded data from table 18b, it is evident that the formulation has better activity in comparison to the expected calculated values. The results from table 18b signify synergetic effect of the formulation. The formulation of cinnamon bark oil, clove bud oil and citric acid at 5:4:10 ratios has the best ZOI readings.

[261] In Table 18b the MIC value of various formulation of cinnamon bark oil, clove bud oil and citric acid are recorded. The experimental values were compared against expected results. The expected results were calculated from the reading recorded in table 18a. The formulation has better activity in comparison to the expected calculated values; this signifies synergetic effect for the formulation. The formulation of cinnamon bark oil, clove bud oil and citric acid at 5:4:10 ratios has the best ZOI readings.

[262] The ZOI and MIC values from microbes treated with three different formulations of the cinnamon bark oil, clove bud oil and rosemary are recorded in Table 17c and 18c. The ZOI and MIC values are compared against the expected values of such formulation. The expected value was calculated by the individual ZOI and MIC provided in Table 17a and 18a. From the data recorded in Table 18c and 17c, it is evident that the formulation has efficacy of clinical significance, this is in light of the expected ZOI and MIC values provided.

EXAMPLE 4 METHOD OF PREPARATION AND APPLICATION OF DIFFERENT PRESERVATIVE FORMULATIONS

EXAMPLE 4A PRESERVATIVE FOR RAW MEAT

[263] In a mechanical stirrer cinnamon bark oil 35g, clove bud oil 35g, rosemary extract 35g, oregano extract 35g, and citric acid 40g was taken along with the excipients Polyoxyethylenesorbitanoleate350g, gum acacia 50g, and hydrogenated castor oil 420g was

taken and mixed well to obtain 1000g of homogenised liquid blend of natural preservative for raw meat.

Method of use

[264] For 100g of fresh meat about 0.3g of natural preservative was taken, the preservative was poured on to the meat and it was applied all over the surface of the meat to cover it up. This way the meat was marinated with the natural preservative. The marinated meat was kept in standard cold storage. The meat was tested after 30days, there was no spoilage was observed, no variation in colour was observed and on preparation no change in taste was observed.

EXAMPLE 4B PRESERVATIVE FOR BREAD

[265] In a ribbon mixture about cinnamon bark oil 4g, clove bud oil 1.5g, encapsulated cinnamon leaf oil 20g, lactic acid 400g, was taken along with the excipients sugar 120g, wheat fibre 29g, and silica 425.5g were taken and mixed till a homogenised powdered mixture was not obtained. About 1000g of powder natural preservative was obtained.

Method of use

[266] About 2g of powder natural preservative for bread is mixed with 1000g of bread dough that is about 0.2% preservative in the dough. The preservative is added before the baking and it is mixed well to spread out evenly in the dough. Finally the bread was baked and kept for observation. The bread was tested after 15days, there was no spoilage observed. Also, there was any change in the texture or taste of the bread even after 15 days.

EXAMPLE 4C PRESERVATIVE FOR CAKE

[267] In a ribbon mixture about cinnamon bark oil 5g, clove bud oil 1g, cinnamon leaf oil 3g, lactic acid 350g, was taken along with the excipients sugar 115g, maltodextrin 136g, wheat fibre 40g, and silica 350g were taken and mixed till a homogenised powdered mixture was not obtained. About 1000g of powder natural preservative was obtained.

Method of use

[268] The natural preservative makes about 0.2% of the cake batter, about 2g of powder natural preservative for cake is mixed along with sugar and fat in to the cake batter. The final weight of the cake batter is about 1000g. Finally the cake was baked and kept for observation. After 15 day the cake was tested, there was not spoilage observed for about 15 days. Also, there was no change in the taste of the cake even after 15 days.

EXAMPLE 4D PRESERVATIVE FOR BEVERAGE

[269] In a mechanical stirrer, cinnamon bark oil 60g, encapsulated cinnamon leaf oil 80g, lactic acid 300g, citric acid 50g, cassia extract 3g mixed along with excipients sugar 200g, Polyoxyethylenesorbitanoleate18.5g and hydrogenated castor oil 122g were taken and mixed well to obtain a homogenised liquid blend. About 1000g of liquid natural preservative was obtained. In case there are any solid partials left the preservative can be passed through a ball mill.

Method of use:

[270] To fresh fruit juice about 50ppm natural preservative for beverages was added and mixed well. The Juice was kept for observation, after 28 day the juice was tested. There was no sign of spoilage even after 28 days and no alteration in smell or taste was observed when tested after 28 days.

EXAMPLE 4E PRESERVATIVE FOR RAW EGG BASED PRODUCTS

[271] In a mechanical stirrer cinnamon bark oil 20g, rosemary extract 200g, natamycin 80gm, and Polyoxyethylenesorbitanoleate 700g were taken and mixed well till homogenised mixture was not obtained. A 1000g homogenised liquid preservative mixture is obtained.

Method of use:

[272] In a freshly prepared 100g mayonnaise about 0.2g of liquid preservative was added and mixed well. The mayonnaise was kept for observation in standard storage for 6 months. After 6 months there was not spoilage observed, no alteration in taste or texture was observed.

EXAMPLE 4F PRESERVATIVE FOR JAM

In a ribbon mixture clove bud oil 30g, lactic acid 250g, citric acid 193g, cardamom extract 7g, natamycin 70g, maltodextrin 200g, and silica 250g was taken and mixed well. About 1000g of uniformly mixed powder preservative formulation for fruit jam was obtained.

Method of use:

[273] In to freshly prepared 100g fruit jam about 0.2g of liquid preservative was added and mixed well. The jam was kept for observation in standard storage for 6 months. After 6 months the jam sample was tested there was no spoilage observed, no alteration in taste, colour or texture was observed.

EXAMPLE 4G PRESERVATIVE FOR BEEN DIP

[274] In a ribbon mixture clove bud oil 20g, Lactic acid 150g, citric acid 190g, ascorbic acid 50g, cumin seed extract 18.5g, rosemary extract 1.5g, along with natamycin 150g and nisin 150g, and maltodextrin 270g are taken and mixed till a uniformly blend powdered formulation is not obtained. About 1000g of natural preservative for been dip was obtained.

Method of use:

[275] In to freshly prepared 100g bean dip about 0.02g of liquid preservative was added and mixed well. The bean dip was kept for observation in standard storage for 12 days. After 12 days the bean dip sample was tested, there was no spoilage observed, no alteration in taste, colour or texture was observed.

EXAMPLE 4H PRESERVATIVE FOR DAIRY PRODUCT

[276] In a ribbon mixture cinnamon bark oil 30g, lactic acid 240g, nisin 50g, sugar 30g, maltodextrin 20g, and silicon 45g were taken together and mixed well till a uniformly blended mixture is not formed. About 1000g natural preservative for dairy product was obtained.

Method of use:

[277] In to freshly prepared 100g condensed milk about 0.2g of the preservative was added and mixed well. The condensed milk was kept for observation in standard storage for 30 days. After

30 days the product was tested and there was no spoilage observed and no alteration in taste, colour or texture was observed.

EXAMPLE 4I PRESERVATIVE FOR DAIRY PRODUCT

[278] In a mechanical stirrer, cinnamon bark oil 15g, clove bud oil 10g, lactic acid 100g, citric acid 50g, rosemary extract 18g, Polyoxyethylenesorbitanoleate 667g, silica 20g, and glysrol 120g were taken and mixed well to get a homogenised solution. About 1000g liquid preservative for pickle was obtained.

Method of use:

[279] In to freshly prepared 100g raw mango pickle about 0.2g of the preservative was added and mixed well. The pickle was kept for observation in standard storage for 1 year. After a year the product was tested and there was no spoilage observed and no alteration in taste or texture was observed.

I Claim;

1. A food preservative selected from the group comprising of an essential oil of a plant, an organic acid and combinations thereof.

- 2. The food preservative of claim 1, wherein the essential oil of the plant is selected from the group consisting of cinnamon bark essential oil, clove bud essential oil, celery seed essential oil, cinnamon leaf essential oil, and combinations thereof.
- 3. The food preservative of claim 1, wherein the organic acid is selected from the group consisting of lactic acid, citric acid, ascorbic acid and combinations thereof.
- 4. The food preservative of claim 1, wherein a weight ratio of the essential of the plant to the weight ratio of organic acid ranges from about 7:2 to about 1:50.
- 5. The food preservative of claim 1 further comprises a plant extract selected from the group consisting of a rosemary extract, a nutmeg fruit extract, a cumin seed extract, and an oregano extract.
- 6. The food preservative of claim 1 further comprises a natural flavouring selected from the group consisting of a cassia extract, a cardamom extract, a vanilla extract, and a sugar.
- 7. The food preservative of claim 1 further comprises a bacteriocin.
- 8. The food preservative of claim 1 further comprises one or more carrier selected from the group consisting of polyoxyethylenesorbitanoleate, gum acacia, Jelucel^R, maltodextrin, wheat fiber, and silica.
- 9. The food preservative of claim 1 for preserving meat selected from the group consisting of raw meat and processed meat, whereinthe food preservative comprises cinnamon bark essential oil, clove bud essential oil, rosemary extract, oregano extract and citric acid.
- 10. The food preservative of claim 9 for preserving meat comprises: about 10% to about 30% of essential oil of cinnamon bark; about 10% to about 30% of essential oil of clove bud; about 10% to about 30% of rosemary extract;

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about 5% to about 30% of oregano extract; and, about 10% to about 40% of citric acid.
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11. The food preservative of claim 9 for preserving meat comprises:

about 17% to about 23% of essential oil of cinnamon bark;

about 17% to about 23% of essential oil of clove bud;

about 17% to about 23% of rosemary extract;

about 17% to about 23% of oregano extract; and,

about 17% to about 23% of citric acid.

- 12. The food preservative of claim 9 for preserving meat further comprises nutmeg fruit extract.
- 13. The food preservative of claim 12 comprises:

about 10% to about 30% of essential oil of cinnamon bark;

about 10% to about 30% of essential oil of clove bud;

about 10% to about 30% of rosemary extract;

about 5% to about 30% of oregano extract;

about 5% to about 25% of nutmeg fruit extract; and

about 10% to about 40% of citric acid.

14. The food preservative of claim 12 comprises:

about 13% to about 19% of essential oil of cinnamon bark;

about 10% to about 16% of essential oil of clove bud;

about 20% to about 26% rosemary extract;

about 7% to about 13% of oregano extract;

about 7% to about 13% of nutmeg fruit extract; and

about 17% to about 23% of citric acid.

15. The food preservative of claim 9, wherein the raw meat is selected from the group consisting of red meat and white meat, wherein the red meat is selected from the group consisting of beef, veal, lamb, venison and pork, and wherein the white meat is selected from the group consisting of chicken, turkey, duck, goose and rabbit.

16. The food preservative of claim 9, further comprises a carrier selected from the group consisting of polyoxyethylenesorbitanoleate, gum acacia, maltodextrin, silica and hydrogenated castor oil and combinations thereof.

- 17. The food preservative of claim 1 for preserving baked product, the food preservative comprises cinnamon bark oil, cinnamon leaf oil, clove bud oil, and citric acid.
- 18. The food preservative of claim 17 for preserving baked or cooked product comprises:

about 0.1% to about 5% of cinnamon bark oil;

about 0.1% to about 5% of clove bud oil;

about 0.1% to about 5% of cinnamon leaf oil; and

about 90% to about 99% of lactic acid.

19. The food preservative of claim 17, wherein baked or cooked product is bread and said preservative comprises:

about 0.6% to about 1.2% of cinnamon bark oil;

about 0.2% to about 0.4% of clove bud oil;

about 1% to about 7% of cinnamon leaf oil; and

about 89% to about 99% of lactic acid.

20. The food preservative of claim 17, wherein baked or cooked product is cake and said preservative comprises:

about 1.2% to about 1.8% of cinnamon bark oil;

about 0.1% to about 0.3% of clove bud oil;

about 0.6% to about 1% of cinnamon leaf oil; and

about 93% to about 99% of lactic acid.

- 21. The food preservative of claim 1 for preserving beverages, the food preservative comprises cinnamon bark oil, cinnamon leaf oil, lactic acid and citric acid.
- 22. The food preservative of claim 21 for preserving beverages comprises:

about 5% to about 25% of cinnamon bark oil;

about 5% to about 25% of cinnamon leaf oil;

about 50% to about 70% of lactic acid; and,

about 5% to about 25% of citric acid.

23. The food preservative of claim 21 for preserving beverages comprises:

about 9% to about 15% of cinnamon bark oil;

about 13% to about 19% of cinnamon leaf oil;

about 55% to about 65% of lactic acid; and,

about 7% to about 13% of citric acid.

24. The food preservative of claim 1 for preserving products prepared with raw egg, the food preservative comprises cinnamon bark oil, rosemary extract and natamycin.

25. The food preservative of claim 24 for preserving products prepared with raw egg, the food preservative comprises:

about 1% to about 5% of cinnamon bark oil;

about 10% to about 40% of rosemary extract; and,

about 1% to about 10% of natamycin.

26. The food preservative of claim 1 for preserving products having a fruit product, selected from the group consisting of fruit jam, fruit jelly, fruit compote, and fruit conserve, wherein the food preservative comprises clove bud oil, lactic acid, citric acid, cardamom, maltodextrin and silica.

27. The food preservative of claim 26 for preserving products having a fruit product rich in pectin, the food preservative comprises:

about 1% to about 10% of clove bud oil;

about 10% to about 40% of lactic acid;

about 10% to about 30% of citric acid;

about 1% to about 10% of cardamom;

about 10% to about 30% of maltodextrin; and

about 30% to about 50% of silica.

28. The food preservative of claim 26 for preserving products having a fruit product rich in pectin, the food preservative comprises:

about 3% to about 7% of clove bud oil;

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about 32% to about 38% of lactic acid;
about 20% to about 30% of citric acid;
about 1% to about 5% of cardamom;
about 5% to about 15% of maltodextrin; and
about 15% to about 25% of silica.
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29. The food preservative of claim 1 for preserving a bean containing product, the food preservative comprises clove bud oil, rosemary extract, cumin seed oil, citric acid, ascorbic acid and lactic acid, wherein the bean containing such product is hummus.

30. The food preservative of claim 29 for preserving the bean containing product, the food preservative comprises:

about 1% to about 20% of clove bud oil; about 0.1% to about 5% of rosemary extract; about 1% to about 10% of cumin seed oil; about 50% to about 70% of citric acid; about 15% to about 30% of ascorbic acid; and about 10% to about 20% of lactic acid.

31. The food preservative of claim 29 for preserving the bean containing product, the food preservative comprises:

about 7% to about 13% of clove bud oil; about 1.5% to about 2.5% of rosemary extract; about 5% to about 7% of cumin seed oil; about 52% to about 62% of citric acid; about 15% to about 25% of ascorbic acid; and about 12% to about 17% of lactic acid.

- 32. A food preservative of claim 1 for preserving dairy or a dairy product, selected from the group consisting of thickened milk dessert, condensed dairy products, ice cream, yoghurt and Khoa wherein the food preservative comprises cinnamon bark oil, lactic acid and nisin.
- 33. The food preservative of claim 32 for preserving the dairy or the dairy product, the food preservative comprises:

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about 1% to about 10% of cinnamon bark oil; about 20% to about 40% of lactic acid; and about 1% to about 10% of nisin.
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34. The food preservative of claim 32 for preserving the dairy or the dairy product, the food preservative comprises:

about 2% to about 8% of cinnamon bark oil; about 25% to about 35% of lactic acid; and about 2% to about 7% of nisin.

- 35. The food preservative of claim 32 further comprises a carrier selected from the group consisting of jelucel, matodextrin and silica.
- 36. The food preservative of claim 1 for preserving a pickle, the food preservative comprises cinnamon bark oil, lactic acid and nisin, wherein the pickle has one or more of a vegetable and fruit.
- 37. The food preservative of claim 36 for preserving the pickle, the food preservative comprises: about 2% to about 15% of cinnamon bark oil;

about 1% to about 20% of Clove Bud; about 30% to about 70% of Lactic acid; about 15% to about 45% of Citric acid; and about 1% to about 30% of rosemary extract.

38. The food preservative of claim 36 for preserving the pickle, the food preservative comprises:

about 5% to about 9% of cinnamon bark oil;

about 3.5% to about 6.5% of Clove Bud;

about 47% to about 57% of Lactic acid;

about 23% to about 29% of Citric acid; and

about 7% to about 13% of rosemary extract.

39. The food preservative of claim 1 for preserving a halwa, the food preservative is selected from the group consisting of cinnamon oil, citric acid, rosemary extract, and clove bud oil and

further include sugar, oregano, oregano extract, cardamom extract, vanilla extract, lactic acid, nisin and natamycin.

40. The food preservative of claim 39 for preserving the halwa, the food preservative comprises:

about 0.1 to about 2% of cinnamon oil;

about 5% to about 10% of citric acid;

about 0.05% to about 0.5% of rosemary extract;

about 0.1% to about 1% of clove bud oil;

about 1% to about 10% of sugar;

about 0.05% to about 0.5% of oregano;

about 1% to about 5% of cardamom extract;

about 0.1% to about 0.5% of vanilla extract;

about 20% to about 60% of lactic acid;

about 1% to about 10% of nisin; and

about 3% to about 6% of natamycin.

- 41. A liquid form of the food preservative of claim 1 comprises a carrier selected from the group consisting of gum acacia, Maltodextrin and silica.
- 42. A powder form of the food preservative of claim 1 comprises a carrier selected from the group consisting of polyoxyethylenesorbitanoleate and hydrogenated castor oil.
- 43. A method of preventing growth of microbes by adding the food preservative of claim 1 to a food product, wherein the microbes are selected from the group consisting of Bacillus species, Rhizopus species, Penicillium species, Aspergillus species, yeast, Staphylococcus species, Coliforms, Lactobacillus species, Mucorsp, Acetobacter, acetic acid bacteria, Pseudomonas species, salmonella species, Listeria species, Clostridium species, Rhizopus species and Enterococci.
- 44. A method of inhibiting growth of microbes by adding the food preservative of claim 1 to a food product, wherein the microbes are selected from the group consisting of Bacillus species, Rhizopus species, Penicillium species, Aspergillus species, yeast, Staphylococcus species, Coliforms, Lactobacillus species, Mucorsp, Acetobacter, acetic acid bacteria, Pseudomonas

species, salmonella species, Listeria species, Clostridium species, Rhizopus species and Enterococci.

- 45. A method of preserving meat by marinating the meat with food preservative of claim 1.
- 46. A method of preserving beverages by adding the food preservative of claim 1 to a beverage.
- 47. A method of preserving products prepared with raw egg by adding the food preservative to a product having raw eggs.
- 48. A method of preserving food having fruit product by adding the food of claim 1 to a food having a fruit product.
- 49. A method of preserving bean containing products by adding the food preservative to a food product having a bean product.
- 50. A method of preserving dairy containing products by adding the food preservative to a food product having a dairy product.
- 51. A method of preservingpickle comprises adding the food preservative to pickle.
- 52. A method of preserving halwa comprises adding the food preservative to halwa.