PHARMACEUTICALS FOR TREATING OR PREVENTING ORAL DISEASES

PRIOR RELATED APPLICATIONS

[0001] This application claims priority to CN_____, filed _____ and incorporated by reference in its entirety.

FEDERALLY SPONSORED RESEARCH STATEMENT

[0002] Not applicable.

REFERENCE TO MICROFICHE APPENDIX

[0003] Not applicable.

FIELD OF THE INVENTATION

[0004] The present invention relates to pharmaceutical, oral or antibacterial products that are manufactured using 5-hydroxymorusin as an active ingredient, the method of preparing 5-hydroxymorusin and its use in manufacturing pharmaceutical, oral or antibacterial products for use in treating or preventing oral disease or as an antibacterial.

BACKGROUND OF THE INVENTION

[0005] Common human oral diseases include caries and periodontal disease. Caries is a chronic bacterial disease occurring in hard dental tissues, manifesting as color, shape, and qualitative changes in the dental material. Clinical symptoms of periodontal disease are gum bleeding, purulence, tooth mobility, absorption of alveolar bone and formation of periodontal pockets. Caries and periodontal disease are also one of the primary causes for the tooth loss of adults. The pathogeny of both caries and

periodontal disease is now considered to be endogenous infections resulted from an imbalance in the normal oral flora caused initially by plaque and resulting in the build up of specific pathogenic microorganisms.

[0006] Bacteria are necessary for the occurrence of caries, and *Streptococcus mutans*, *Actinomyces naeslundi*, *Actinomyces viscosus* and the like break up carbohydrates to yield acids, causing tooth inorganic material to demineralize and form caries cavities. *Streptococcus mutans* and *Streptococcus sanguinis* are also the pioneer bacteria in the formation of dental plaques, forming dental plaques together with *Actinomyces naeslundi, Actinomyces viscosus*, *Porphyromonas gingivalis, Actinobacillus actinomycetemcomitans and Fusobacterium nucleatum*. The toxins and other detrimental substances produced by the bacteria in the dental plaques may cause the inflammatory reactions of the host, lead to the increase in vascular permeability and the spread of inflammation, destroy the gum, dental bone and alveolar bone, and cause gingivitis and periodontal diseases.

[0007] Therefore, inhibiting oral pathogenic microorganisms is an important approach to prevent caries, gingivitis and periodontal diseases. However, products for treating caries and periodontal disease are very limited at present. On one hand, pharmaceuticals can be used to eliminate or inhibit the plaque bacteria, but the long-term use of broad-spectrum antibacterial agents will result in the selection for drug-resistant strains and other side effects. For example, the chemical plaque-controlling agent Chlorhexidine used both at home and abroad in recent years is a diguanido-hexane having obvious antibacterial function, but the long-term use of gargle containing 0.12-0.20% Chlorhexidine will cause stained tooth and formation of brown tongue coat at the back of tongue. On the other hand, Traditional Chinese Medicines can be used instead of pharmaceuticals, such as gargling “heat-clearing” and “detoxifying” extracts like honeysuckle, etc., as well as external-application methods, such as applying Bingpeng San (borneol and borax powder) or Asarum on the diseased part. These traditional remedies are inconvenient, however, and there is no conclusive evidence proving their efficacy. Nevertheless, if the gingivitis is not treated timely or properly, the inflammation may

spread to the periodontal apex to cause the inflammation of periodontal apex tissues, and further affecting the alveolar bone or the neighboring tissues.

[0008] What is needed in the art is research and development of new medicines from natural plants for treating common oral diseases such as caries and periodontal diseases, and to develop oral pharmaceuticals or oral care products for inhibiting primary oral pathogenic microorganisms and dental plaques, gingivitis, and for countering breath odor.

[0009] Research on natural plants that can inihibit oral microorganisms has occurred thourghout the world. Research has identified plants of the anti-caries effects, such as *Anogeissus Wall.* and *Vernonia esculenta*, *Garcinia morella Desv.* (gamboges) of Africa and the Middle East; blood grass roots and cocoa fruit of Latin American countries; *Areca catectu L.* (betel nut) of Southeast Asia; and the Eucalyptus leaves of Australia. There has been no report in the prior art, however, on oral products containing hydroxy-morusin as an active ingredient.

DESCRIPTION OF INVENTION

[0010] The present invention provides a pharmaceutical or oral care product for treating or preventing oral diseases that is prepared with purified 5-hydroxymorusin as an active ingredient, the preparing method thereof and the use thereof in manufacturing medicaments or oral care products for treating or preventing oral diseases.

[0011] By "purified 5-hydroxymorusin" what is meant is the pharmaceutical, oral or antibacterial product is made using a 5-hydroxymorusin that is at least 90% pure. Thus, the definition excludes prior art teas, or other traditional herbal preparations of the originating plant and has sufficient purity so as to be pharmaceutically acceptable. However, even though a product may be made with at least 90% pure 5-hydroxymorusin, or preferably at least 95% pure 5-hydroxymorusin for pharmaceutical applications, the final concentration in the ultimate product may of course be quite low, as

in mouth washes or antiseptic sprays. In other embodiments, such as tablets, the final content may be quite high.

[0012] More particularly, the invention relates to an oral product, containing purified 5-hydroxymorusin together with pharmaceutically or orally acceptable carriers or excipients, and optionally including additional medical adjuvants.

[0013] The oral product includes, but not limited to, oral pharmaceuticals and oral care products such as mouth wash, rinses, toothpaste, oral care gel strips, dental gums, tablest, losenges, and the like.

[0014] Further, although the inventors have an interest in oral uses for purified 5-hydroxymorusin, it's activity against bacteria can be employed in a variety of bactericidal or bacteriostatic uses, such as in cleansers, disinfectants, soaps, and the like, or as an antibacterial additive for tissue paper, plastics, coatings, and the like.

[0015] The present invention also provides a method for preparing the purifiying 5-hydroxymorusin. *Cortex Mori* root and/or bark are harvested, extracted with ethanol, then with ethyl acetate, and then subjected to silica gel chromatography and elution with petroleum ether: acetone in 8:2 (v/v). The purified 5-hydroxymorusin is then formulated to make pharmaceutical, oral or antibacterial products.

[0016] The inventor of the present invention finds for the first time that 5-hydroxymorusin has certain inhibiting effect on oral pathogenic microorganisms. The purified compound may be applied in anti-caries and breath odor-inhibiting oral pharmaceuticals or oral care products as an effective ingredient, and may also be used in oral pharmaceuticals or oral care products for countering gingivitis and dental plaques, removing oral sensitivity, countering tartar and whitening the teeth.

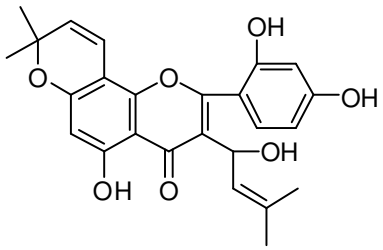
[0017] The invention will be illustrated in the following non-limiting examples.

EXAMPLE 1 PREPARATION OF HYDROXYL-MORUSIN

[0018] 50kg *Cortex Mori* (White Mulberry root-bark) is extracted with 500kg 90% ethanol under reflux for 3 times, 3h each time, and the extract is recovered to dryness to obtain 10.5kg extractum. The extractum is dissolved and suspended with 8kg hot water and 4kg 90% ethanol, and extracted with 50kg ethyl acetate for three times, recovered to dryness, obtaining 4kg ethyl acetate extractum. The resultant extractum is dissolved with acetone and adsorbed on 6kg silica gel, evaporated to dryness at room temperature, and subjected to a silica gel column chromatography (30 meshes, 30kg), eluted with petroleum ether: acetone (v/v, 8:2). Every 50 liters is taken as a fraction, obtaining 27 fractions (Fr.1~Fr.27) in all. The fraction Fr.5 is subjected to a silica gel column chromatography, and eluted with petroleum chloroform: methanol (150:1) to obtain the compound 5-hydroxymorusin (2.78mg).

[0019] Auto SPEC type 3000mass spectrometer was used for mass spectrometry (MS) analysis. Bruker DRX-500 superconductive NMR spectrometer was used to determine the nuclear magnetic resonance spectrometry (^{1}H NMR and ^{13}C NMR), with TMS as the internal standard. The silica gel for column chromatography and thin layer column chromatography was a product of Qingdao Meigao Group Co. Ltd.

[0020] 5-hydroxymorusin is also known as “artonin E” or 5'-hydroxymorusin or 4H,8H-Benzo(1,2-b:3,4-b')dipyran-4-one, 5-hydroxy-8,8-dimethyl-3-(3-methyl-2-butenyl)-2-(2,4,5-trihydroxyphenyl)-; CAS 129683-93-8, and has the following structure:



Properties: light-yellow powder
Molecular formula: $C_{25}H_{24}O_7$
Molecular weight: 436
^{1}H-NMR($CDCl_3$, 500 MHz) δ: 7.65(1H, d, H-6'), 6.77(1H, d, J = 10.0Hz, H-14), 6.56(1H, d, J = 8.5 Hz, H-5'), 6.43(1H, s, H-6), 6.26(1H, s, H-3'), 6.24(1H, s, H-9), 5.61(1H, d, J =

10.5 Hz, H-10), 5.42(1H, d, J = 9.5 Hz, H-15), 1.97(3H, s, H-12),1.70(3H, s, H-13), 1.48(3H, s, H-17), 1.47(3H, s, H-18).
^{13}C-NMR($CDCl_3$, 100MHz) δ: 161.4(s, C-2), 108.8(s, C-3), 178.2(s, C-4), 105.1(s, C-4a), 163.0(s, C-5), 99.7(d, C-6), 157.8(s, C-7), 101.0(s, C-8), 150.8(s, C-8a), 69.2(t, C-9), 124.7(d, C-10), 138.4(s, C-11), 25.6(q, C-12), 18.3(q, C-13), 114.5(d, C-14), 127.2(d, C-15), 77.6(s, C-16), 27.8(s, C-17), 27.8(s, C-18), 107.1(s, C-1'), 155.4(s, C-2'), 104.2(d, C-3'), 158.6(s, C-4'), 110.0(d, C-5'), 120.8(s, C-6').

[0021] 5-hydroxymorusin prepared by this method may have a very high purity. In certain examples, the purity of the 5-hydroxymorusin prepared by this method is higher than 90%, 95%, 98%, 99% or 99.9% as measured by common techniques, such as HPLC. In certain other examples, the concentration of the 5-hydroxymorusin prepared by this method is at least 50% and preferably at least 75%, 80%, or 85%.

[0022] In the examples above, we have used *Cortex Mori* (White Mulberry root-bark) as the plant source of the 5-hydroxymorusin compound. However, it will be understood by those of ordinary skill in the art that other plants may also be applicable, such as those of the genus *Morus*, such as *M. sinensis*, *M. alba, M. latifolia*, *M. serrata*, *M. tiliaefolia, M. bombycis, M. rotundiloba* and *M. laevigata*, provided they contain adequate levels of the desired ingredient.

[0023] In addition to natural sources, those skilled in the art may synthesize the active ingredient easily by chemical methods without creative work, to substitute the extract or purified substance used in the present invention. Therefore, those skilled in the art should understand that, in addition to extraction or purification from plants, the goals of the present invention may also be achieved through chemical synthesis. Other obvious modifications of the compound disclosed in the present invention will also be included within the protection scope of the present invention.

EXAMPLE 2 ANTIBACTERIAL EFFECT

[0024] The various bacterial species cultivated for antibacterial tests are listed in Table 1.

Table 1: Related oral pathogenic microorganisms

Strains	ATCC number	Gram properties	Culture medium
Streptococcus mutans **(S. m)**	25175	G(+)	TSB
Porphyromonas gingivalis **(P. g)**	33277	G(-)	FTM / RCM (3:1)
Actinomyces viscosus **(A.v)**	27044	G(+)	TSB
TSB = tryptone soya broth, FTM = fluid thioglycollate medium, RCM = reinforced clostridial medium			

[0025] A single colony was picked from the Trypticase Digested Soybean peptone agar blood plate (TSA5B) of ordinarily stored bacteria species and used to innoculate corresponding broth culture medium, cultivated in a microaerobic environment (P.g needs anaerobic cultivation) with 95% air, 5% CO_2 at 37.0 ˚C ± 1.0 ˚C, wherein S.m is cultivated for 18-24 h, and the remaining two species are cultivated for 40-48 h. Then corresponding broth cultivate medium was used to adjust the turbidity of the bacteria suspension to $0.5^{\#}$ McFarland standard, equivalent to 1.0×10^8 CFU/mL.

[0026] Bacteria were diluted through a series of 2-fold dilutions, and the anti-bacterial pharmaceutical was quantitatively inoculated with tested bacteria, and observed after incubated at 37˚C for 18-24 h, wherein the lowest pharmaceutical concentration that inhibited bacterial growth visible to the naked eye is the minimum inhibitive concentration (MIC) of the tested pharmaceuticals active against the tested bacteria. The operating steps were:

[0027] a. Preparation of the stock solution of antibacterial pharmaceutical: Preparing 1% stock solution of various antibacterial pharmaceuticals with 100% pure ethanol as solvent. The prepared stock solution was filtered to eliminate bacteria, and was distributed into small portions to be used.

[0028] b. Range of measured concentration: the present experiment used 250 ppm as the ceiling measured concentration of the anti-bacteria pharmaceutical.

[0029] c. Measuring method: micro-dilution method.

[0030] Firstly, 100 μL broth medium was added to each well of the 96-well plate, and 100 μL sterile anti-bacteria pharmaceutical solution which was diluted 10-fold (1000mg/l) was added to each hole of the first column, then each hole of the first column is blowed repeatedly for 7-8 times using multi-channel micropipet, and 100μL sample is transferred to the second column after sufficiently mixing the pharmaceutical with TSB. After similarly being blowed for 7-8 times, 100μL sample is transferred to the third column, and analogously to the last column. Thus the concentration of the pharmaceutical is 2-fold gradiently diluted, from 500 mg/L of the first column to 0.24 mg/L of the last column (the 12th column).

[0031] The bacteria to be tested and standard bacteria were prepared as above. The bacteria solution is diluted with broth medium to a bacteria content of about 10^6 CFU/mL. Then 100μL was inoculated into each well. Thus the final diluted concentration of the anti-bacteria pharmaceutical in each line are 250, 125, 62.5…0.12 mg/L, and the final inoculation quantity is about 5×10^7 CFU/mL or 5×10^6 bacteria in each well. The 96-well plate was placed on a microshaker and shaken for 1 min to mix the solutions in each well. The microwell plate was covered and sealed with adhesive paper to reduce the evaporation during the incubation process and placed in a humid box, cultivated in microaerobic environment with 95% air, 5% CO_2 (or in anaerobic environment with 90% N_2, 5% H_2, 5% CO_2) at 37.0 ˚C ± 1.0 ˚C for 18-24h. The 96-well plate is placed under an enzyme-labeling instrument, and by comparing the growing characteristics of the tested bacteria with the standard bacteria, the lowest anti-bacteria pharmaceutical concentration contained in the hole without bacteria was taken as MIC. The results are shown in Table 2.

Table 2: Effect of 5-hydroxymorusin on oral pathogenic microorganisms

	A. v ATCC 27044	**S. m ATCC 25175**	**P. g ATCC 33277**
Negative control (DMSO)	-	-	-
Positive control (Triclosan)	3.9	3.9	7.8
5-hydroxymorusin	7.8	31.25	62.5

EXAMPLE 3: ANTI-INFLAMMATORY EFFECT

[0032] Anti-inflammatory activity was assessed using the KB cell (the epidermic cancer cell in human oral cavity). During the experiment, the KB cell inoculated in the cultivating plate was treated with or without (control) compound 5-hydroxymorusin. After the treatment, the supernatant of the cultivating medium was collected and stored in -80°C refrigerator. Inflammation was assessed by measuring several inflammation markers: Prostaglandin E2 (PGE_2) in the supernatant was detected by Enzyme-linked immunsorbent assay, and Luminex multifunctional liquid-phase chip analysis system was used for the detection of granulocyte monocyte colony stimulating factor (GM-CSF), tumor necrosis factor-alpha (TNF-α), and interleukins 1-beta and 6 (IL-1β and IL-6).

[0033] Prostaglandin E2 (PGE_2) result showed that the 50% inhibitory concentration of 5-hydroxymorusin to the growth of oral KB cell is 0.02 ppm, equivalent to that of the positive control Triclosan. The result indicated that 5-hydroxymorusin had significant anti-inflammatory activity.

[0034] Luminex multifunctional liquid-phase chip analyses of GM-CSF, TNF-α, IL-1 and IL-6: The 96-well plate was enclosed with enclosing liquid for 30min. Fluorescent microspheres were added into the 96-well plate after dilution. The standard and the samples to be measured were added, and incubated overnight at 4˚C. The next day, the supernatant was discarded and 50 μl GM-CSF, TNF-α, IL-1β and IL-6 antibodies are added to each well. After being rinsed for 4 times, the 96-well plate was placed in a shaking table under ambient temperature to shade from light for 1h; rinsed 4 times, and added PE labeled with streptavidin and shaded from light for 15 min at room temperature, and then rinsed for 4 times. The microspheres were suspended in the rinsing buffer, and the 96-well plate placed immediately on Luminex (Luminex Corporation, Austin, Tx.) for analysis.

[0035] The compound 5-hydroxymorusin has a very strong anti-inflammatory ability. The specified results are shown in Table 3.

Table 3: the anti-inflammatory effects of 5-hydroxymorusin (the larger the value, the stronger the anti-inflammatory ability)

Extract/compound	GM-CSF	IL-1β	IL-6	TNF-α
Compound 5-hydroxymorusin	11%	21%	52%	89%
Triclosan (Positive control)	5%	7%	31%	81%

[0036] The above experimental results indicate that 5-hydroxymorusin has excellent inhibitory effect on oral pathogenic microorganisms (*Actinomyces viscosus, Actinobacillus actinomycetemcomitans, Streptococcus mutans*, *Porphyromonas gingivalis*), has significant anti-inflammatory activity to oral KB cells, and also has significant inhibitory effect on inflammatory factors, and therefore should be very useful in the formulation of oral care products and pharmaceuticals for the treatment of bacterial and inflammatory diseases.

EXAMPLE 4: 5-HYDROXYMORUSIN USES

[0037] 5-hydroxymorusin may be used directly, or may be employed in oral pharmaceuticals or oral care products such as toothpaste, mouthwash, paste, tablets, thin film, chewing gum and the like as the effective ingredient. It may be in an effective amount together with other ingredients such as humectant, abrasive, surfactant, pharmaceutically acceptable carrier and/or additive, colorants, stabilizers, flavorants, sweeteners, and it may include other pharmecurtical agents such as antibacterials.

[0038] The term “effective amount” indicated herein refers to an amount sufficient to produce a positive effect. Such an effective amount will vary in particular embodiments, for example, a gum may contain less active ingredient that a mouthwash since it may be used several times a day. In addition, there may be multiple effective dosages or an effective dosage range. In some examples, 5-hydroxymorusin may be mixed with other ingredients in an amount of 0.0005-20% (all % are mass percentage unless other wise indicated). In some other embodiments, the content of 5-hydroxymorusin is 0.001-10%. In some preferred examples, the content of 5-hydroxymorusin is 0.025-5%. In some other examples, the content of 5-hydroxymorusin is 0.05-0.5%.

[0039] Preferably, when used in a tablet formulation, the content of 5-hydroxymorusin is 4%; in nasal sprays, the content of 5-hydroxymorusin is 4%; in dropping pills, the content of 5-hydroxymorusin is 10%; in toothpastes, the content of 5-hydroxymorusin is 0.5%; in mouthwash, the content of hydroxyl-morusin is 0.5%, and in oral pastes, the content of 5-hydroxymorusin is 0.01%. Other effective amounts may also be applicable in the present invention.

[0040] The composition of formulation according to the present invention may also contain cationic surfactants and/or non-ionic surfactants. Cationic surfactants include, but not limited to, cetyltrimethylammonium chloride, di(isobutyl-phenoxy-ethoxy-ethyl)-di(xylene-benzyl) ammonium bromide. Non-ionic surfactants include Poloxamer, polyethoxy ether, and ethoxy fatty acid and the like. Poloxamer is a block copolymer of polyoxyethylene and polyoxypropylene, having industrial applications, such as that produced by BASF under the trade name “Pluronic”. Polyethoxy ether includes polyoxyethylene sorbitol fatty acid ester (a typical monoester of polyethoxy alcohol), which is produced by ICL company with the trade name Tween. Other non-ionic surfactants include polyoxyethylene alkylphenol, polyoxyethylene alcohols, fatty acids, polyoxyethylene esters, polyoxyethylene alkyl amine glyceride, polyglyceride, tetritol ester, pentitol ester, hexitol ester, dehydrated tritol ester and polyoxyalkylamine polyalkyl ester. Typically, the mass content of surfactant in the formulation is 0.001%-3.0%. Divalent metal ions, such as zinc, copper, selenium, calcium or magnesium, may be included in the formulation. They will be in the form of soluble inorganic salts such as zinc chloride or the form of organic/inorganic compounds. The amount of divalent metal ions added into the mixture will be 0.001% - 3.0%. The formulation may contain oligosaccharides, which will be in the water-soluble state, and may convert into soluble salt, if added, in an amount of at least 0.01%.

[0041] The present invention may include various commonly used oral pharmaceuticals or oral hygiene products, such as mouthwash, oral spray, toothpaste, gels, oral mucosa plasters, oral pellicle, instant tablet and film, chewing gum and the like products for mammals, especially human and animals, to inhibit plaque and prevent dental caries and periodontal disease.

[0042] Other additives may be used azs appropotiate for the embodiment—mouthwash may contain acidic substances; mouthwash may also contain desensitizing substance such as potassium nitrate; toothpaste may contain abrasive such as sodium carbonate, calcium phosphate, alumina and silica; solubilizer such as PEG, glycerol, ethanol; other taste masking agents such as xylitol; thickener such as carrageenan; and surfactant such as SLS, etc. may all be used.

[0043] Waxes such as beeswax may be used in toothpaste or oral mucosa plaster; thickener and pellicle-forming agents such as carrageenan, CMC, HPMC, xanthan gum and the like may also be added.

[0044] Final products may includes essences such as mint, spearmint, eucalyptus leaf oil, menthol, carvone, wintergreen, cloves, cinnamon, lemon, grapefruit, orange and some essences decomposed and digested from yeast and protein.

[0045] In tablets and pellicles, inert excipients may be used for the formation of the products. Adjuvants such as colorant, taste masking agent and the like are mainly included to improve the taste and appearance.

[0046] The composition of the formulation related with the application of the present invention in tablets may contain filler, adhesive, lubricant, disintegrant. Fillers include, but not limited to, starch, lactose, microcrystalline cellulose and the like. Adhesives include, but not limited to, starch slurry, cellulose derivatives, polyvidone, gelatin and the like. Lubricants include, but not limited to, magnesium stearate, micropowder silica gel, talcum powder, polyethylene glycols and the like. Disintegrants include, but not limited to, sodium carboxymethyl starch, low substituted hydroxylpropyl cellulose, cross-linked polyvidone, cross-linked sodium carboxymethyl cellulose and the like. All these ingredients are pharmaceutically acceptable adjuvants recorded in the Chinese Pharmacopoeia and the Pharmacopoeia of many countries such as United Kingdom and United States.

[0047] Matrix such as polyethylene glycol and the like may be used in the pastes of the present invention. Polyethylene glycol in solid may be combined with liquid

forms to adjust the consistency. Ingredients such as magnesium stearate may also be added to adjust the consistency.

EXAMPLE 5: SPECIFIC FORMULATIONS

[0048] Specific formulations are presented below:

5-hydroxymorusin toothpaste formulation 1:

Components	Content (mass percentage, %)
5-hydroxymorusin	0.0005 – 20
Abrasive	10-30
Humectant	50-69
Water, Flavor, Flavoring	proper amount

5-hydroxymorusin toothpaste formulation 2:

Components	Content (mass percentage, %)
5-hydroxymorusin	5
Silica	30
Glycerol	5
Sorbitol	50
Water, Flavor, Flavoring	proper amount

5-hydroxymorusin toothpaste formulation 3:

Components	Content (mass percentage, %)
5-hydroxymorusin	0.001
Silica	10
Sorbitol	69
Water, Flavor, Flavoring	proper amount

5-hydroxymorusin toothpaste formulation 4:

Components	Content (mass percentage, %)
5-hydroxymorusin	10
Abrasive	25
Humectant	65

5-hydroxymorusin toothpaste formulation 5:

Components	Content (mass percentage, %)
5-hydroxymorusin	0.0005
Silica	10
Sorbitol	70
Water, Flavor, Flavoring	proper amount

5-hydroxymorusin toothpaste formulation 6:

Components	Content (mass percentage, %)

5-hydroxymorusin	20
Abrasive	25
Humectant	55

5-hydroxymorusin toothpaste formulation 7:

Components	**Content (mass percentage, %)**
5-hydroxymorusin	0.025
Silica	20
Glycerol	5
Sorbitol	60
Sodium fluoride	0.221
Sodium saccharin	0.3
Water	7.954
Polyethylene glycol	3
Sodium dodecyl sulfate	2
Essence	1.5

5-hydroxymorusin toothpaste formulation 8:

Components	**Content (mass percentage, %)**
5-hydroxymorusin	0.3
Silica	20
Glycerol	5
Sorbitol	60
Sodium fluoride	0.221
Sodium saccharin	0.3
Water	7.679
Polyethylene glycol	3
Sodium dodecyl sulfate	2
Essence	1.5

5-hydroxymorusin toothpaste formulation 9:

Components	**Content (mass percentage, %)**
5-hydroxymorusin	0.5
Silica	20
Glycerol	5
Sorbitol	60
Sodium fluoride	0.221
Sodium saccharin	0.3
Water	7.479
Polyethylene glycol	3
Sodium dodecyl sulfate	2
Essence	1.5

5-hydroxymorusin mouthwash formulation 1:

Components	**Content (mass percentage, %)**
5-hydroxymorusin	0.0005 – 20
Water	70-94
other additives	proper amount

5-hydroxymorusin mouthwash formulation 2:

Components	Content (mass percentage, %)
5-hydroxymorusin	5
Water	80
Pluronics	3.00
other additives	proper amount

5-hydroxymorusin mouthwash formulation 3:

Components	Content (mass percentage, %)
5-hydroxymorusin	0.001
Water	94
Alcohol	5.00
other additives	proper amount

5-hydroxymorusin mouthwash formulation 4:

Components	Content (mass percentage, %)
5-hydroxymorusin	0.5
Water	90
Pluronics	3.00
Alcohol	3.00
other additives	proper amount

5-hydroxymorusin mouthwash formulation 5:

Components	Content (mass percentage, %)
5-hydroxymorusin	0.025
Water	85
Pluronics	5.00
Alcohol	3.00
other additives	proper amount

5-hydroxymorusin mouthwash formulation 6:

Components	Content (mass percentage, %)
5-hydroxymorusin	0.05
Water	91.7
Pluronics	3.00
Alcohol	5.00
Essence	0.25

5-hydroxymorusin mouthwash formulation 7:

Components	Content (mass percentage, %)
5-hydroxymorusin	0.0005
Water	94
Alcohol	5.00
other additives	proper amount

5-hydroxymorusin mouthwash formulation 8:

Components	Content (mass percentage, %)

5-hydroxymorusin	20
Water	70
Pluronics	3.00
Alcohol	3.00
other additives	proper amount

5-hydroxymorusin paste formulation 1:

Components	**Content (mass percentage, %)**
5-hydroxymorusin	10
Polyethylene glycol	90

5-hydroxymorusin Paste formulation 2:

Components	**Content (mass percentage, %)**
5-hydroxymorusin	0.001
Polyethylene glycol	80
other additives	proper amount

5-hydroxymorusin paste formulation 3:

Components	**Content (mass percentage, %)**
5-hydroxymorusin	0.01
Polyethylene glycol	90
other additives	proper amount

5-hydroxymorusin paste formulation 4:

Components	**Content (mass percentage, %)**
5-hydroxymorusin	5
Polyethylene glycol	95

5-hydroxymorusin paste formulation 5:

Components	**Content (mass percentage, %)**
5-hydroxymorusin	0.025
Polyethylene glycol	99.975

5-hydroxymorusin paste formulation 6:

Components	**Content (mass percentage, %)**
5-hydroxymorusin	20
Polyethylene glycol	80

5-hydroxymorusin paste formulation 7:

Components	**Content (mass percentage, %)**
5-hydroxymorusin	0.0005
Polyethylene glycol	99
other additives	proper amount

[0049] 5-hydroxymorusin Tablets—5-hydroxymorusin 10mg, lactose 180mg, starch 55mg, magnesium stearate 5mg. Active ingredient, lactose and starch are mixed and wetted uniformly with water. The wetted mixture is subjected to sieve and dry, subjected to sieve again, added in magnesium stearate. The mixture is then is compressed into tablets, weighing 250 mg each, with a final content of the active ingredient content of 10 mg per tablet.

5-hydroxymorusin nasal spray:

5-hydroxymorusin	80 mg
sodium chloride	8 mg
EDTA	1 mg
Sodium phosphate buffer (pH6.5)	10 mg
Polyethoxy ether	10 mg
redistilled water	to 2ml

[0050] Preparing method: One ingredient at a time is added into proper volume of redistilled water under stirring until completely dissolved, then another ingredient is added. After adding water to 2ml, the solution is filtered on a sterile filter, filled into bottles and separated according to suitable dosage.

5-hydroxymorusin dropping pills:

5-hydroxymorusin	1g
polyethylene glycol 6000	9g

[0051] Preparing method: preparation of molten liquid of 5-hydroxymorusin and polyethylene glycol 6000: 5-hydroxymorusin is weighed according to the prescribed amount above, to which a proper amount of dehydrated ethanol t is added. After being dissolved by tepefaction, the resultant substances are added into prescribed amount of polyethylene glycol molten liquid (incubated in 60˚C water bath).The mixture is stirred to mix uniformly until ethanol is completely evaporated. The resultant is allowed to stay in 60˚C water bath to incubate for 30 min. After bubbles being completely removed, the uniformly mixed molten liquid above in which bubbles are completely removed is transferred into a storing tank. Under a condition that the temperature is maintained at 80-85˚C, the molten liquid is dropped by controlling the dropping speed, the melton liquid is dropped dropwise into a condensing liquid under the controlling of the dropping speed. After the condensation being completed, the condensing liquid is decanted and the drop

pills are collected. Condensing liquid on the pills is trickled and removed with filter paper. Then, the pills are placed in a silica gel desiccator or dried naturally.

[0052] The above detailed description of the present invention sets forth a lot of specified details, the object thereof is to give clear and integrated explanation of the invention to facilitate the public to read and understand. However, what will be clear to one skilled in the art is that in some cases, the present invention may also be practiced without these specified details, or by making some non-essential modification or substitution to these specified details. These variations should be all considered to fall into the sprite and scope of the present invention. The scope of the present invention is determined by the plain language of the appended claims, other than limited by the specific examples provided in the detailed description.

CLAIMS:

1. A pharmaceutical composition, comprising purified 5-hydroxymorusin in an amount effective for treating or preventing bacterial or inflammatory disease, and a pharmaceutically acceptable carrier.
2. The pharmaceutical composition of claim 1, wherein the final content of the 5-hydroxymorusin is at least 0.05%.
3. The pharmaceutical composition of claim 1, wherein the final content of the -hydroxymorusin compound is 0.1-50%.
4. An antibacterial composition, comprising purified 5-hydroxymorusin in an amount effective for killing bacteria or preventing the growth of bacteria, together with a carrier.
5. The antibacterial composition of claim 4, wherein the final content of the 5-hydroxymorusin compound is at least 0.0005%.
6. The antibacterial composition of claim 4, wherein the final content of the 5-hydroxymorusin compound is 0.001-10%.
7. An oral care product, comprising purified 5-hydroxymorusin in an amount effective for oral care, and an orally acceptable carrier.
8. The oral care product of claim 5, wherein the final content of the 5-hydroxymorusin compound is at least 0.0005%.
9. A use of purified 5-hydroxymorusin for in manufacturing a medicament for treating or preventing oral diseases.
10. A use of purified 5-hydroxymorusin compound in manufacturing oral care products.
11. A method of purifying 5-hydroxymorusin, comprising extracting a plant containing 5-hydroxymorusin with an organic solvent; and isolating 5-hydroxymorusin by chromatography to produce 5-hydroxymorusin that is at least 75% pure.
12. The method of claim 11, wherein the plant is of the genus *Morus.*
13. The method of claim 11, wherein the plant is *Cortex Mori.*
14. The method of claim 11, wherein said purified 5-hydroxymorusin is at least 85% pure.
15. The method of claim 11, wherein said 5-hydroxymorusin is at least 95% pure.
16. The method of claim 11, wherein said 5-hydroxymorusin is at least 98% pure.
17. The method of claim 11, wherein said 5-hydroxymorusin is at least 99% pure.
18. The method of claim 11, further comprsing extracting root and/or bark from a Morus plant with ethanol, then with ethyl acetate, and then subjecting the extract

to silica gel chromatography and elution with petroleum ether: acetone.

19. A method of purifying 5-hydroxymorusin, comprising extracting root and/or bark from *Cortex Mori* with 90% ethanol, then with ethyl acetate, and then dissolving the extract in acetone and subjecting the acetone extract to silica gel chromatography and elution with petroleum ether: acetone in 8:2 (v/v).
20. The method of claim 19, wherein said said 5-hydroxymorusin is at least 95% pure.