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ORIGINAL ARTICLE

Preparation of Stable Jointed Acid Microspheres Using Wheel Protein Isolate-gum Arabic Complex

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Abstract:. Conjugated linoleic acid (CLA) microspheres stabilized. whey protein isolate (WPI)-gum arabic(GA) complex (WPI-GA ISCs) were prepared by the emusification relief method. the results showold that the solvent evacuation oriented in aggregation of the WPI-GA complex and sequential shrinkage and collapse of the WPI-GA interface, contributing to the picker layer. the emulsifying stability of the WPI-GA ISCS was improved, therapeutic preventing particle aggregation and CLA leakage. when the concentration of WPI-GA ISCS was higher than 1%, the ClA microspheres had good physical stability and resolution after freeze-drying with high efficiency of encapsulation and delivery.

Keywords: Wheat protein isolate (WPI)-Gum Arabic (GA) Complex;Gastrointedistinct delivery

With caused the academic circles of the general concern[3]. Use gelatin and pectin between of electrostatic role preparation and low-calorie starch particles similar size and functional of hydrogel Particles[4]. In probiotics microcapsules encapsulation technology in use Polysaccharide-Protein complex can improve in different stress under of probiotics Activity[5]. Use soybean separation protein-Sodium Alginate copolymer preparation lycopene micelle solve the lycopene solubility and stability poor problem[6]. Polysaccharide-Protein complex both

Protein superior of emulsion activity and polysaccharide of space stability effective improve food emulsion of stability[7/-8].

Electrostatic Force is protein and polysaccharide each other role of most important driving force. InPHValue lower than the protein isoelectric point when protein with net positive charge can and anion polysaccharide formation complex. Study show that protein and polysaccharide in electrostatic role under Combined with make protein interface layer get protection; polysaccharide of polymer quality and high hydrophilic make milk drop between there space repulsion prevent milk drop of coalescence[1]. Protein and polysaccharide of each other role by many factors of influence suchPHValue, ion strength, mixed proportion and. With the protein and polysaccharide mixed proportion of different can formation soluble molecular in complex, soluble molecular between complex, insoluble molecular between complex, etc. Early

Study show that gum arabic (Gum ArabicGA) And whey separation protein (Whey protein isolationWPI) To mass ratio2 1Mixed,PH 4.4When can formation stability of soluble molecular in complex has very superior of emulsion activity and Emulsion Stability[12]. ButWPI-GAMolecular in complexPHValue sensitive when emulsion system deviationPH 4.4An arcane occurs solution from to caused by Emulsion Stability decreased.

This in the experiment,WPI-GAMolecular in complex for emulsifier the milk

Of-Solvent volatile legal system by conjugate linoleic acid (Conjugated Linoleic Acid

CLA) Microspheres by evaluationCLAMicrospheres of stability and its gastrointestinal digestion release characteristics to improveWPI-GAMolecular in complexPHValue stability for expandWPI-GAMolecular in complex of application provide theory support.

1.1 Material and reagent

GAJapaneseSan-EI GenFood the company;WPIAmerican Davis ke the company;CLA(Purity80%), Beijing, hao er si Science and Technology Limited the company; gastric protease, trypsin AmericanSigmaThe company;NaCl,$CaCl_2$, Bile salt3, Chinese medicine group chemical reagent limited the company.

1.2 Instrument and Equipment

Mastersizer 2000Laser Particle Size Analyzer British Malvern instrument limited the company;PT-MR2100High-speed Shear pre-emulsifying machine SwitzerlandKinematica The company;ORION 4 STAR pHOf Mettler-Toledo instrument (Shanghai) Limited the company; Vacuum Frozen Dryer , Beijing, bo yi kang experimental instrument limited The company; rotating Evaporator IKA Instrument Equipment limited the company; double beam purple The spectrophotometer, Beijing, Purkinje general instrument limited responsibility the company.1.3Methods

1.3.1 CLAMicrospheres of preparation

GAAndWPIBy mass ratio2 1Said take the right amountWPIAndGASoluble in Pure Water in placed roller mixer on the room temperature mixed12 HMake its full dissolved mixingHClSolution RegulationPHValue4.4Stirring1 HFormation stabilityWPI-GASoluble molecular in complex. Take the right amountCLAIn anhydrous ethanol in preparation100 mg/MLOfCLA-Ethanol solution stirring1 H. WillCLA-Ethanol solution slow-by-drop join high-speed Shear stirring (Natural 20 000 R/Min)WPI-GASolution in.45 ℃ Rotation evaporation fast remove ethanol to obtainCLAMicrospheres Dispersion,CLAQuality score2%In accordance with the above-mentioned methods preparation quality fraction respectively0.1%,0.5%,1%,2%And5%OfWPI-GAMolecular in complex. Will Fresh preparation of samples the ~~Frozen~~ Dry can get powderCLAMicrospheres.

1.3.2 CLAMicrospheres particle size of Determination

Will emulsion slight oscillation shake-by-drop to ultra-pure water dispersant inHydro 2000MUStyle wet sampler into-like. Dispersion phase and continuous phase of refractive index respectively1.52And1.33Samples of Absorption Rate0.01Pump is2 000 R/Min. Add samples to laser index slightly greater10%Can Start determination. Emulsion of average particle size with surface area weighted average$D_{[3,2]}$Said by formula (1) Calculation:

1.3.3 CLAMicrospheres physical stability of evaluation

Will Preparation Get of freshCLAMicrospheres dispersion placed40 ℃ Constant Temperature Incubator in place0,3,5,7/ dAfter the emulsion particle size determination.

1.3.4 CLAMicrospheres micro-morphology of observation

Take a very small amountCLAMicrospheres powder in scanning electron microscope samples on spraying under scanning electron microscope observe the take pictures record.

1.3.5 CLAMicrospheres encapsulation rate of Determination

The n-hexane preparation certain quality concentrationCLASolution with UV spectrophotometer wavelength scanning determinationCLAThe maximum absorption wavelength234 nm. In maximum absorption wavelength under determination different quality concentrationCLASolution (0~10 Mu g/ML) Of absorbance draw absorbance andCLAQuality concentration of standard curve get Correlation EquationY= 0.097 5X-0.010 8Related CoefficientR^2= 0.996 2Linear relationship good.

Take1 ml CLAMicrospheres dispersion emulsion join10 mLN-hexane full oscillation extraction centrifugal take n-hexane phase in234 nmWavelength under Determination CLAThe absorbance. AccordingCLAIn n-hexane in standard curve equation by formula (2) CalculationCLAEncapsulation rate[13]:

1.3.6 CLAMicrospheres in simulation gastrointestinal environment under release rate of Determination

Take1.0 mL CLAMicrospheres suspension placed37℃ Pre temperature29.0 mLSimulation gastric juice (2 mg/ML NaCl,3.2 mg/MLGastric protease,HClSolutionPHValue2.0) In,37℃ Constant temperature water bath stirring rate100 R/MinDigestion3 HDuring each30 minOfPHValue make its stability in2.0Respectively in30,60,90,120,150,180 minTake samples DeterminationCLAThe release rate. Take1.0 mLFor determination of Particle Size,1.0 mLAnd the same amount of n-hexane Extraction

Take and constant volume10.0 mLDetermination234 nmWavelength the of absorbance and calculation content.

Gastric digestion stage end after1 Mol/L NaOHSolutionPHValue

7.0Transfer30 mLSimulation intestinal juice (8 mg/ML NaCl40 mg/ML $CaCl_2$5 mg/MLBile salt,10 mg/MLTrypsin) in continue

37℃ Constant temperature water bath stirring rate100 R/MinDigestion3 HDuring each30 minOfPHValue make its stability in7.0Respectively in30,60,90,120,150,180 minTake samples DeterminationCLAThe release rate. Take1.0 mLFor determination of Particle Size,1.0 mLAnd the same amount of n-hexane extraction and constant volume10.0 mLDetermination234 nmWavelength the of absorbance and calculation content[6,14]. According to the formula (3) CalculationCLARelease Rate:

2.1 WPI-GAMolecular in complex quality fractionCLAMicrospheres particle size of influence

ToWPI-GAMolecular in complex for emulsifier Emulsion-Solvent volatile legal systemCLAMicrospheres by figure1The withWPI-GAMolecular in complex quality scores of freshCLAMicrospheres average particle size first reduce after increase trend. Quality score0.1% WPI-GAMolecular in complex of particle size is big quality score0.5%~2%PreparationCLAMicrospheres Particle Size Distribution close to average the particle size tends to be constant meansWPI-GAMolecular in complex inCLAMicrospheres interface adsorption has to saturated[15]. When quality fraction5%When average particle size increased,CLAMicrospheres slightly flocculation. This is because water phase free of excess emulsifier will lead to by emptying

Effect of microspheres Flocculation[16-18]. By figure1BShow that the quality scoreWPI-GAMolecular in complexCLAMicrospheres particle size were present single peak distribution and quality score1%~2%OfCLAMicrospheres particle size is small. Show that the quality fraction of by Emulsification-Solvent volatile legal systemCLAMicrospheres distribution uniform, stability is good.

2.2 CLAMicrospheres of Physical Stability

By different quality scoreWPI-GAMolecular in complex preparationGLAMicrospheres40℃ Storage conditions under the particle size change see Figure2The quality fractionCLAMicrospheres in7/dStorage Period in particle size distribution no obvious change,5% WPI-GAMolecular in complex preparationCLAMicrospheres with the storage time of extend in a small amount of particles flocculation show thatCLAMicrospheres has is good physical stability.

By figure3The emulsion-Solvent volatile legal system byCLAMicrospheres distribution is uniform Particles Surface folds collapse has thick of interface layer. This is because emulsion after the of solvent volatile causing the parcel inCLAMicrospheres in

Of the ethanol escape to make Emulsion Formation of circular particles collapse and interface Shrinkage[19]. BecauseWPI-GAMolecular in complex inPHValue4.4An arcane Formation,

WhenPHValue Deviation4.4An arcaneWPI-GAMolecular in complex will happen solution from, or by condensation lead to generation precipitation. This experimental selection Emulsion-Solvent volatile legal systemCLAMicrospheres use solvent of volatile to makeCLAMilk drop interface happen shrinkage and aggregation to improve milk drop interface adsorptionWPI-GAMolecular in complex of stability.

By figure4AThe whenWPI-GAMolecular in complex quality score 0.1%When preparationCLAMicrospheres basic can't freeze-dried oil serious,WPI-GAIn interface can't completely coatedCLADroplet lead to freeze-dried Process InCLAA large number of overflow. WhenWPI-GAMolecular in complex quality score0.5%When,CLAMicrospheres freeze-dried after obvious coalescence and form the massive; whenWPI-GAMolecular in complex quality

elevated1%More,CLAMicrospheres freeze-dried after present powder5% WPI-GAMolecular in complex preparationCLAMicrospheres powder most delicate storage dispersion best. Will freeze-dried

AfterCLAMicrospheres powder dissolved reduction to fresh preparation of dispersion concentration the particle size analysis by figure4BThe,1% WPI-GAMolecular in complex preparationCLAMicrospheres powder complex soluble after part particles aggregation particle size larger quality score2%And5%OfWPI-GAMolecular in complex preparationCLAMicrospheres powder complex of after the particle size distribution and fresh Preparation of Phase close,CLAMicrospheres powder complex soluble effect is good.

2.4 CLAMicrospheres of encapsulation rate

From table1Can see whenCLAQuality score when withWPI-GAMolecular in complex quality fraction of increase onCLAMicrospheres of encapsulation rate gradually increased. WhenWPI-GAMolecular in complex quality fraction increase2%When the encapsulation efficiency reached97%More than encapsulation effect good. This experimental selection Emulsion-Solvent volatile legal systemCLAMicrospheres use solvent of volatile makeCLAMilk drop interface happen shrinkage and aggregation improve the milk drop interface adsorptionWPI-GAMolecular in complex of stability makeCLAMicrospheres interface thickness increase can effective preventCLAOf overflow and oxidation.

2.5 CLAMicrospheres in simulation gastrointestinal environment in stability and Release Rate

By figure5The emulsion-Solvent volatile legal system byCLAMicrospheres dueWPI-GAMolecular in complex in interface of aggregation improve the complexPHValue stability.CLAMicrospheres in simulation gastric juice in more stable basic keep single peak distribution with the in simulation gastric juice in time of extended appear large particles aggregation peak,CLAThe release rate slow increase. Is due to speculationCLAMicrospheres interface on theWPI-GAMolecular in complex in gastric Protease of role under START gradually was enzymatic hydrolysis leadCLAMicrospheres of interface protection barrier reduce[Natural 20-21]. SoCLATo system diffusion of rate gradually increase in gastric juice180 minAn arcane release rate36.0%. In simulation intestinal juice in with the time extension,CLAMicrospheres of large particles aggregation peak increase obvious,CLARelease Rate also rapid increase of trend in180 minTimeClARelease rate reached approximately62.2%. Compared with simulated gastric juice,ClAThe release rate in simulated intestinal fluid was faster. This is becauseClAIt is speculated that bile salts in the small intestine can replace the large particles in the simulated intestinal fluid.WPI-GAIntramolecular complex

Union[22-28]It accelerated the adsorption of trypsin at the interface and the destruction of the protective barrier at the interface of the microspheres.[29-30], LeadingClA

The release rate increased significantly.

In this experimentWPI-GAThe intramolecular complex was used as emulsifier and emulsified-Preparation by solvent evaporationClAParticles, using solvent to volatilizeClAOn the microsphere InterfaceWPI-GAInterfacial aggregation occurs in Intramolecular complexes,ClAThe interface of microspheres collapsed and shrunk.WPI-GAIntramolecular ComplexesPHStability, so that it has a goodClAGastrointestinal transport characteristics, improve the polysaccharide-Application Value of protein electrostatic complex.

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