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(54) Titre : PRODUITS A MACHER COMESTIBLES POUR ANIMAUX  
(54) Title: EDIBLE ANIMAL CHEWS

(57) **Abrégé/Abstract:**

An edible animal chew comprising: a partially hydrolysed collagen; and/or an alkyl succinate modified starch, wherein the edible animal chew has a tensile toughness greater than 200 PMA.

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## (54) Title: EDIBLE ANIMAL CHEWS

(57) Abstract: An edible animal chew comprising: a partially hydrolysed collagen; and/or an alkyl succinate modified starch, wherein the edible animal chew has a tensile toughness greater than 200 PMA.



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## **Edible Animal Chews**

### **Background**

Edible animal chews provide oral care and mental stimulation through occupation for  
5 domestic animals, particularly dogs. The material properties of current edible animal  
chews are provided by starch, which is sometimes included in the form of flours. The  
longest chewing time per gram of synthetic animal chews has been provided by  
combining the material properties of starch with the use of an aerated structure.

10 Alternative, longer lasting, animal chews can be formed from animal hide (raw-hide),  
which requires no, or only minimal, processing. Despite providing longer chewing time,  
commercial exploitation of these animal-hide-based chews have several major  
drawbacks. These products have been linked to several cases of damage to teeth, as  
well as intestinal damage and microbacterial poisoning. Furthermore, the inconsistent  
15 size, shape and properties of the rawhide, which are affected by the part of the animal  
hide being used, the quality of the butchering and subsequent processing, results in an  
inconstant product not suited to large scale commercialisation.

### **Brief Description of the Figures**

20

Figure 1A provides the tensile toughness of various edible animal chews (Powders  
Samples 1-5), wherein each edible animal chew comprises pork skin protein (PSP).

25 Figure 1B provides ratio of SME (mechanical energy transferred by the extruder) to  
rotations per minute (RPM) seen for various edible animal chews (Powders Samples 1-  
5).

30 Figure 2 shows triangular contour plots of the expanded hot strength of the edible  
animal chew of the present disclosure, when the edible animal chew comprises A) 0%  
and B) 10 wt.% hydrolysed sodium octenyl succinate starch.

35 Figure 3 shows triangular contour plots of the expanded cross section of the edible  
animal chew of the present disclosure, when the edible animal chew comprises A) 0%  
and B) 10 wt.% hydrolysed sodium octenyl succinate starch.

Figure 4 shows triangular contour plots of the tensile strength with A) 0% and B) 10 wt.% hydrolysed sodium octenyl succinate starch.

5 Figure 5 shows triangular contour plots of the shear resistance of the edible animal chew of the present disclosure, when the edible animal chew comprises A) 0% and B) 10 wt.% hydrolysed sodium octenyl succinate starch.

10 Figure 6 shows triangular contour plots of the hardness (conical probe) of the edible animal chew of the present disclosure, when the edible animal chew comprises A) 0% and B) 10 wt.% hydrolysed sodium succinate starch.

15 Figure 7 shows A) daily and B) weekly lasting time data for edible animal chews containing pork skin protein in comparison to currently available daily and weekly products.

20 Figure 8 shows a comparison of the tensile toughness of edible animal chews formed by using partially hydrolysed collagen with those formed by combining natural collagen extract with gelatine (hydrolysed collagen).

25 Figure 9 shows the tensile test equipment (which can be used to determine tensile toughness) and samples. A) cutting press; B) ribbon (flat sheet) extrudate; C) a tensile bar cutting stamp [ISO 527-2; 75 mm long, 10 mm wide, 5 mm centre]; D) a tensile bar during texture analysis; and E) a tensile bar after analysis – the tensile bar is positioned within the grips of the tensile testing machine so that its full shoulder is exposed. F) a general plot of the force response curve of a texture analysis test plotted on axes of stress (MPa) versus strain (%). The dimensions of the central break point (width and depth) are accounted for in the plot.

### Detailed Description

30

The presently-disclosed subject matter is illustrated by specific but non-limiting examples throughout this description. The examples may include compilations of data that are representative of data gathered at various times during the course of

development and experimentation related to the present invention(s). Each example is provided by way of explanation of the present disclosure and is not a limitation thereon.

5 All references to singular characteristics or limitations of the present disclosure shall include the corresponding plural characteristic(s) or limitation(s) and vice versa, unless otherwise specified or clearly implied to the contrary by the context in which the reference is made.

10 All combinations of method or process steps as used herein can be performed in any order, unless otherwise specified or clearly implied to the contrary by the context in which the referenced combination is made.

15 While the following terms used herein are believed to be well understood by one of ordinary skill in the art, definitions are set forth to facilitate explanation of the presently-disclosed subject matter.

20 Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which the presently-disclosed subject matter belongs. Although any methods, devices, and materials similar or equivalent to those described herein can be used in the practice or testing of the presently-disclosed subject matter, representative methods, devices, and materials are now described.

25 Following long-standing patent law convention, the terms "a", "an", and "the" refer to "one or more" when used in this application, including the claims. Thus, for example, reference to "an animal chew" may include a plurality of such animal chews, and so forth.

30 Unless otherwise indicated, all numbers expressing quantities, properties, and so forth used in the specification and claims are to be understood as being modified in all instances by the term "about". Accordingly, unless indicated to the contrary, the numerical parameters set forth in this specification and claims are approximations that can vary depending upon the desired properties sought to be obtained by the presently-disclosed subject matter.

35 As used herein, the term "about," when referring to a value or to an amount of mass, weight, time, volume, concentration or percentage is meant to encompass variations of

in some embodiments  $\pm 20\%$ , in some embodiments  $\pm 15\%$ , in some embodiments  $\pm 10\%$ , in some embodiments  $\pm 5\%$ , in some embodiments  $\pm 1\%$ , in some embodiments  $\pm 0.5\%$ , and in some embodiments  $\pm 0.1\%$  from the specified amount, as such variations are appropriate to perform the disclosed invention(s).

5

As used herein, ranges can be expressed as from "about" one particular value, and/or to "about" another particular value. It is also understood that there are a number of values disclosed herein, and that each value is also herein disclosed as "about" that particular value in addition to the value itself. For example, if the value "10" is disclosed, then "about 10" is also disclosed. It is also understood that each unit between two particular units is also disclosed. For example, if 10 and 15 are disclosed, then 11, 12, 13, and 14 are also disclosed.

10

The terms "component" and "portion" are used interchangeably throughout the present disclosure to mean a part or portion.

15

As used herein, "pet food" means a composition intended for oral consumption to meet one or more nutritional needs of a pet. Pet food expressly excludes items that are capable of being orally ingested but are not intended to be ingested, such as rocks. The terms "pet food" and "pet food product" are used interchangeably throughout this disclosure. A pet food may be, for example, in certain embodiments, a treat, a chew, a biscuit, a gravy, a supplement, a topper and any combination thereof.

20

As used herein, "dietary composition" refers to any composition utilized as part of the diet for a dog. This includes, but is not limited to, a pet food, a treat, a chew, a biscuit, a gravy, a supplement, a topper, and any combination thereof.

25

As used herein, "nutritionally balanced" and/or "nutritionally complete" refers to a composition capable of sustaining life as the sole dietary ration for an animal, without the need for any other substance, except possibly water.

30

As used herein, the terms "animal" or "pet" mean a domestic animal including, but not limited to, a domestic dog, cat, horses cow, ferret, rabbit, pig, or the like. Domestic dogs and cats are particular examples of pets. "Dog" includes adult dogs, between 1 year of age and 7 years of age; seniors, older than 7 years of age; and super-seniors,

35

older than 11 years of age. For the purpose of this disclosure, "dog" does not include puppies under the age of 1 year.

5 All lists of items, such as, for example, lists of ingredients, are intended to and should be interpreted as Markush groups. Thus, all lists can be read and interpreted as items "selected from the group consisting of ... list of items ... "and combinations and mixtures thereof."

10 All percentages in the present disclosure are listed as percent by weight on the total weight of the material or mixture, unless explicitly noted otherwise.

15 In a first aspect, there is provided an edible animal chew. The edible animal chew may comprise a partially hydrolysed collagen and/or alkyl succinate modified starch. The edible animal chew may have a tensile toughness greater than 200 MPa.

In a second aspect, there is provided a method of making an edible animal chew. The method may comprise:

20 providing an edible chew composition comprising a partially hydrolysed collagen and/or alkyl succinate modified starch;

heating the edible chew composition to form a flowable edible chew composition;

extruding the flowable edible chew composition to form an extrudate;

allowing the extrudate to cool and/or harden to form the edible animal chew.

25 In a third aspect, there is provided an edible animal chew according to the first aspect, wherein the edible animal chew is produced by a method according to the second aspect.

30 Edible chews that are resilient against chewing and last longer deliver increased oral care efficacy and an enhanced chewing experience to the animal. The present inventors have found that the partially hydrolysed collagen and succinate modified starch (i.e. either used alone or together) provide edible animal chews that have improved physical properties in comparison to previous edible animal chews; for example, one of more of the following properties is improved: hardness, shear force,  
35 ductility, tensile strength and toughness. By improving the physical properties of the

edible animal chews, the present inventors have devised edible animal chews with an improved, that is, a longer, lasting time.

As used herein, "lasting time" refers to the time taken for an animal to completely  
5 consume the product, that is, the time from which the animal first begins to chew the  
product to the time when the animal swallows the last pieces of the product. The lasting  
time excludes any time that the animal may be playing with the product but not actually  
chewing it. By increasing the lasting time of an animal chew, this increases the  
propensity of the chew to clean the teeth and gums to a greater extent. Additionally,  
10 the net calorie intake per chew can be reduced, in view of the increased amount of  
chewing an animal has to perform to finish the chew.

It should be noted that an animal "chew" is quite distinct from an animal food or pet  
food. An animal chew differs from an animal food in at least the size of the pieces, the  
15 time taken for the animal to consume each piece, in the number of pieces per serving  
and/or in nutritional content. Thus, the largest pieces of food are smaller than those in a  
chew, and pieces of food are generally consumed faster than chews. For example, an  
animal chew may have a longest dimension of at least about 5 cm, optionally at least 7  
cm, optionally at least about 10 cm, optionally at least about 15 cm.

20

### **Edible animal chew**

Described herein is an edible animal chew. The edible animal chew comprises a  
partially hydrolysed collagen and/or an alkyl succinate modified starch. In other words,  
25 the edible animal chew may contain the partially hydrolysed collagen and alkyl  
succinate modified starch or it may contain partially hydrolysed collagen and lack alkyl  
succinate modified starch or it may contain alkyl succinate modified starch and lack  
partially hydrolysed collagen.

30 Collagen, in its natural state, comprises aggregates (fibrils) of collagen molecules  
(tropocollagen). Each tropocollagen molecule comprises a triple helix of protein chains  
(polypeptide strands). Partially hydrolysed collagen, as described herein, may indicate  
a collagen-derived product, in which collagen, in its natural state, that has been  
subjected to a process that involves contacting the collagen with water so that at least  
35 some of collagen fibrils have untangled from one another and/or tropocollagen triple

helices have untangled (i.e. the triple helix has unwound, partially or fully, to form three protein chains) to form the partially hydrolysed collagen. There may or may not have been some hydrolysis and shortening of the peptide strands during the partial hydrolysis. In an embodiment, there may have been little or no shortening of the peptide chains during the partial hydrolysis. The partially hydrolysed collagen has preferably been formed such that at least some of the partially hydrolysed collagen is water insoluble, and preferably at least some of the partially hydrolysed collagen is water soluble. The partially hydrolysed collagen, before inclusion in the animal chew, is typically not gelatin, which is the result of a hydrolysis process that involves shortening the protein chains of collagen to an extent that the gelatin is entirely water soluble (at least at concentrations of about 1-2wt% in water at 25°C). Gelatin, for example, will typically give a clear solution in water, whereas the partially hydrolysed collagen as used herein, will typically form a cloudy mixture with water, with some of the partially hydrolysed collagen being dissolved and some of the partially hydrolysed collagen being suspended in the water (e.g. when mixed with water in a 1:15 wt/wt ratio, i.e. with one part by weight partially hydrolysed collagen to 15 parts by weight water at a temperature of 25°C).

The partially hydrolysed collagen may have a Bloom gel strength of at least 300 g, optionally at least 350 g, optionally at least 400 g, optionally at least 450 g, optionally from 300 g to 600 g, optionally from 300 g to 550 g, as measured in accordance with ISO 9665. The "Bloom gel strength" is a measure of the ability of a material to form a gel. The Bloom gel strength is the weight in grams required to depress the gel a distance of 4 mm with a piston having a cross-sectional area of 1 cm<sup>2</sup>, the gel first having been cooled for a defined time under defined conditions. Thus, the higher the Bloom gel strength of a material is, the greater the ability of that material to form a gel. The gel for this measurement has a standard concentration of 6.67% and has been kept 17 hours at 10 °C prior to measurement.

The partially hydrolysed collagen may have a molecular weight of at least 60kDa, optionally at least 70 kDa, as measured using gel permeation chromatography (GPC), optionally from 60kDa to 100 kDa, optionally from 70 kDa to 90 kDa, optionally from 75 kDa to 85 kDa, optionally about 80 kDa.

The partially hydrolysed collagen, before incorporation into the edible animal chew, may be formable by a process comprising forming a wet collagen product in the form of a fibrous mass from a hide or skin at a temperature of 50 °C or less, followed by drying the wet collagen product using a contact dryer, optionally having a surface temperature  
5 of 150°C or higher, to obtain the partially hydrolysed collagen in the form of a powder.

The partially hydrolysed collagen, before incorporation into the edible animal chew, may have been formed by a method comprising the steps of: producing at a temperature of 50 °C or less a wet collagen product by subjecting a hide or skin to a  
10 size reduction step; an alkaline and/or an oxidizing treatment step; and a neutralizing step; followed by drying of said wet collagen product using a contact dryer, to obtain the partially hydrolysed collagen in the form of a powder. Optionally, the drying is carried out using a contact dryer having a surface temperature of more than 150 °C, optionally 155 °C or higher, optionally 160 °C or higher, optionally a surface  
15 temperature of  $165 \pm 4$  °C.

Optionally, the temperature of the steps for producing the wet collagen product is kept at 45 °C or less and preferably 40 °C or less. The wet collagen product may be produced by subjecting the hide or skin to a size reduction step, e.g. by cutting or  
20 grinding the skin or hide into pieces, and then dispersing the pieces in water. The hide or skin dispersed in the water may be subjected to an alkaline and/or an oxidising treatment. The alkaline treatment may involve the water in which the hide or skin is dispersed containing an alkaline agent, such as a Group I or Group II hydroxide, e.g. sodium hydroxide or calcium hydroxide, in an amount of at least 1wt%, optionally at  
25 least 2 wt%, such that the pH of the water is at least 10, optionally at least 11, optionally at least 12, but preferably not greater than 12.5. The alkaline treatment may be for at least 10 seconds to 10 minutes, optionally 10 seconds to five minutes, optionally 10 seconds to a minute,

30 The oxidising treatment may involve an oxidising agent, such as hydrogen peroxide, being present in the water in which the hide or skin is dispersed, and the oxidising agent may be present with the alkaline agent.

The animal hide or skin is selected from the group consisting of bovine, porcine and  
35 poultry. Preferably, the hide or skin is bovine hide or skin.

The neutralizing treatment may result in a pH value of neutral or 6 or less, preferably from 5 to 6. The neutralising treatment may involve contacting the skin or hide with water containing an acid. The acid may be selected from lactic acid, hydrochloric acid, carbon dioxide, acetic acid, ethylene diamine tetraacetic acid, ammonium chloride, propionic acid, and fumaric acid. Carbon dioxide or acetic acid are the preferred acid. The method of partially hydrolysing the collagen may avoid allowing the collagen to contact water having a pH of less than 4.2, optionally less than 5.

Preferably, drying the wet collagen product is carried out within 24 hours after the size reduction of the skin or hide, preferably within 12 hours after the size reduction of the skin or hide, more preferably within 6 hours after the size reduction of the skin or hide, and most preferably within one hour of the neutralisation step.

The partially hydrolysed collagen may be produced in the method described in WO2011/149356, which is incorporated herein by reference, to produce a collagen powder.

Partially hydrolysed collagen may be obtained commercially, e.g. products sold under the trade name Kapro B95, available from D.C.P Ingredients B.V.

In some examples, the edible animal chew comprises 10 wt.% or more partially hydrolysed collagen. In some examples, the edible animal chew comprises 11 wt.% or more, in some examples, 12 wt.% or more, in some examples, 13 wt.% or more, in some examples, 14 wt.% or more, in some examples, 15 wt.% or more, in some examples, 16 wt.% or more, in some examples, 17 wt.% or more, in some examples, 18 wt.% or more, in some examples, 19 wt.% or more, in some examples, 20 wt.% or more, in some examples, 21 wt.% or more, in some examples, 22 wt.% or more, in some examples 23 wt.% or more, in some examples 24 wt.% or more, in some examples, 25 wt.% or more, in some examples, 26 wt.% or more, in some examples, 27 wt.% or more, in some examples, 28 wt.% or more, in some examples, 29 wt.% or more, in some examples, 30 wt.% or more, in some examples, 31 wt.% or more, in some examples, 32 wt.% or more, in some examples, 33 wt.% or more, in some examples, 34 wt.% or more, in some examples, 35 wt.% or more, in some examples, 36 wt.% or more, in some examples, 37 wt.% or more, in some examples, 38 wt. % or

more, in some examples, 39 wt.% or more, in some examples, 40 wt.% or more, in some examples, 41 wt.% or more, in some examples, 42 wt.% or more, in some examples, 43 wt.% or more, in some examples, 44 wt.% or more, in some examples, 45 wt.% or more, in some examples, 50 wt.% or more, in some examples, 55 wt.% or more, in some examples, about 60 wt.% partially hydrolysed collagen.

In some examples, the edible animal chew comprises 60 wt.% or less partially hydrolysed collagen. In some examples, the edible animal chew comprises 55 wt.% or less, in some examples, 50 wt.% or less, in some examples, 45 wt.% or less, in some examples, 44 wt.% or less, in some examples, 43 wt.% or less, in some examples, 42 wt.% or less, in some examples, 41 wt.% or less, in some examples, 40 wt.% or less, in some examples, 39 wt.% or less, in some examples, 38 wt.% or less, in some examples, 37 wt.% or less, in some examples, 36 wt.% or less, in some examples, 35 wt.% or less, in some examples, 34 wt.% or less, in some examples, 33 wt.% or less, in some examples, 32 wt.% or less, in some examples, 31 wt.% or less, in some examples, 30 wt.% or less, in some examples, 29 wt.% or less, in some examples, 28 wt.% or less, in some examples, 27 wt.% or less, in some examples, 26 wt.% or less, in some examples, 25 wt.% or less, in some examples, 20 wt.% or less, in some examples, 19 wt.% or less, in some examples, 18 wt.% or less, in some examples, 17 wt.% or less, in some examples, 16 wt.% or less, in some examples, 15 wt.% or less, in some examples, 14 wt.% or less, in some examples, 13 wt.% or less, in some examples, 12 wt.% or less, in some examples, 11 wt.% or less, in some examples, about 10 wt.% partially hydrolysed collagen.

In some examples, the edible animal chew comprises 10 wt.% to 60 wt.% partially hydrolysed collagen. In some examples, the edible animal chew comprises 11 wt.% to 55 wt.%, in some examples, 12 wt.% to 50 wt.%, in some examples, 13 wt.% to 45 wt.%, in some examples, 14 wt.% to 44 wt.%, in some examples, 15 wt.% to 43 wt.%, in some examples, 16 wt.% to 42 wt.%, in some examples, 17 wt.% to 41 wt.%, in some examples, 18 wt.% to 40 wt.%, in some examples, 19 wt.% to 39 wt.%, in some examples, 20 wt.% to 38 wt.%, in some examples, 25 wt.% to 37 wt.%, in some examples, 26 wt.% to 36 wt.%, in some examples, 27 wt.% to 35 wt.%, in some examples, 28 wt.% to 34 wt.%, in some examples, 29 wt.% to 33 wt.%, in some examples, 30 wt.% to 32 wt.%, in some examples, 31 wt.% to 32 wt.% partially hydrolysed collagen. In some examples, the edible animal chew comprises 10 wt.% to

25 wt.%, in some examples, 10 wt.% to 15 wt.%, in some examples, 15 wt.% to 20 wt.%, in some examples, 25 wt.% to 45 wt.%, in some examples, 25 wt.% to 35 wt.%, in some examples, 35 wt.% to 45 wt.% partially hydrolysed collagen.

5 The edible animal chew may comprise an alkyl succinate modified starch. An alkyl succinate modified starch may be defined as a starch having covalently bound thereto an alkyl succinate. An alkyl succinate starch may be defined as a starch in which at least some of the hydroxyl groups of the starch have been reacted with an alkyl succinate or an alkyl succinate precursor, such as an alkyl succinate anhydride, such  
10 that the succinate group of the alkyl succinate is attached to the oxygen of the hydroxyl group. The present inventors have found that, while natural collagen (i.e. extracted collagen, but not hydrolysed or detangled) can assist in creating animal chews with high toughness, there is a disadvantage in using it, particularly in extruded products. In an extruded product, the starting materials are typically passed through a heated  
15 extruder. However, natural collagen melts at a relatively low temperature, and the resultant viscosity of the composition being extruded is low; this in turn makes the composition difficult to extrude. Similarly, some natural collagen may contain a relatively high amount of fat, which, again, can lead to low viscosity of the composition in a heated extruder. The present inventors have found, however, that the addition of  
20 an alkyl succinate modified starch to a composition comprising collagen, whether natural or partially hydrolysed, raises the viscosity of the composition, allowing for more effective extrusion. Additionally, as is shown in the Examples below, embodiments of compositions comprising partially hydrolysed collagen and alkyl succinate modified starch have been found to have improved properties in at least one of the following:  
25 hardness, shear force, ductility, tensile strength and toughness.

In some examples, alkyl succinate is a C4 to C12 alkyl succinate. In some examples, alkyl succinate is a C5 to C11 alkyl succinate, in some examples, a C6 to C10 alkyl succinate, in some examples, a C7 to C9 alkyl succinate, in some examples, a C8 alkyl  
30 succinate.

In some examples, the succinate is a metal succinate. In some examples, the metal is selected from Group 1 metals, Group 2 metals and Group 3 metals or a combination thereof. In some examples, the metal succinate is selected from sodium succinate,

potassium succinate, magnesium succinate, calcium succinate and aluminium succinate or a combination thereof.

5 Optionally, the alkyl succinate modified starch is selected from sodium octenyl succinate starch, calcium octenyl succinate starch, potassium octenyl succinate starch, aluminium octenyl succinate starch and any combination thereof. Alkyl succinate modified starch is available commercially, e.g. products sold under the trade names Clearam® and Cleargum®, available from Roquette®.

10 In some examples, the alkyl succinate modified starch acts as an emulsifier. In some examples, the alkyl succinate modified starch binds fats in the edible animal chew composition, reducing the disruptive effects of fat on processing. In some examples, the alkyl succinate modified starch allows water and steam to disperse evenly within the edible animal chew composition during processing.

15 In some examples, the modified starch is formed by modification of a starch selected from maize starch, potato starch (optionally, high viscosity potato starch), tapioca starch or combinations thereof.

20 In some examples, the alkyl succinate modified starch is formed by chemical addition of alkyl succinate groups to a hydrolysed dextrin. In some examples, the alkyl succinate modified starch is a thin boiling alkyl succinate modified starch. A thin boiling alkyl succinate modified starch contributes little viscosity during processing under heat and hydration.

25 The edible animal chew may comprise 1 wt.% or more alkyl succinate modified starch. The edible animal chew may comprise 2 wt.% or more, in some examples, 3 wt.% or more, in some examples, 4 wt.% or more, in some examples, 5 wt.% or more, in some examples, 6 wt.% or more, in some examples, 7 wt.% or more, in some examples, 8  
30 wt.% or more, in some examples, 9 wt.% or more, in some examples, 10 wt.% or more, in some examples, 11 wt.% or more, in some examples, 12 wt.% or more, in some examples, 13 wt.% or more, in some examples, 14 wt.% or more, in some examples, 15 wt.% or more, in some examples, 16 wt.% or more, in some examples, 17 wt.% or more, in some examples, 18 wt.% or more, in some examples, 19 wt.% or more, in  
35 some examples, 20 wt.% or more, in some examples, 21 wt.% or more, in some

examples, 22 wt.% or more, in some examples, 23 wt.% or more, in some examples, 24 wt.% or more, in some examples, about 25 wt.% alkyl succinate modified starch.

5 The edible animal chew may comprise 25 wt.% or less, in some examples, 24 wt.% or less, in some examples, 23 wt.% or less, in some examples, 22 wt.% or less, in some examples, 21 wt.% or less, in some examples, 20 wt.% or less, in some examples, 19 wt.% or less, in some examples, 18 wt.% or less, in some examples, 17 wt.% or less, in some examples, 16 wt.% or less, in some examples, 15 wt.% or less, in some examples, 14 wt.% or less, in some examples, 13 wt.% or less, in some examples, 12 wt.% or less, in some examples, 10 wt.% or less, in some examples, 9 wt.% or less, in some examples, 8 wt.% or less, in some examples, 7 wt.% or less, in some examples, 6 wt.% or less, in some examples, 5 wt.% or less, in some examples, 4 wt.% or less, in some examples, 3 wt.% or less, in some examples, 2 wt.% or less, in some examples, about 1 wt.% alkyl succinate modified starch.

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The edible animal chew may comprise 1 wt.% to 25 wt.%, in some examples, 2 wt.% to 24 wt.%, in some examples, 3 wt.% to 23 wt.%, in some examples, 4 wt.% to 22 wt.%, in some examples, 5 wt.% to 21 wt.%, in some examples, 6 wt.% to 20 wt.%, in some examples, 7 wt.% to 19 wt.%, in some examples, 8 wt.% to 18 wt.%, in some examples, 9 wt.% to 17 wt.%, in some examples, 10 wt.% to 16 wt.%, in some examples, 11 wt.% to 15 wt.%, in some examples, 12 wt.% to 14 wt.%, in some examples, 12 wt.% to 13 wt.%, in some examples, 13 wt.% to 15 wt.% alkyl succinate modified starch. The edible animal chew may comprise 1 wt.% to 10 wt.%, in some examples, 10 wt.% to 20 wt.% alkyl succinate modified starch.

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The edible animal chew may comprise partially hydrolysed collagen, of which some is water-soluble and some is water insoluble. Water solubility of the partially hydrolysed collagen may be determined by dispersing an amount of partially hydrolysed collagen in water at a concentration of 1 – 2 wt%, letting the partially hydrolysed collagen swell for 15 minutes at 25 °C, then raising the temperature of the water to 60 °C, and then allowing to stand for 30 minutes at 60 °C. The mixture of water and partially hydrolysed collagen is then centrifuged at 8000 rpm at 40 °C for 7 minutes. The mixture is then filtered using a one-way 0.2 µm filter at 40 °C. The centrifuging and the filtering serve to remove the non-water-soluble collagen from the mixture. The mixture is then analysed to determine the amount of protein (i.e. the partially hydrolysed

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collagen) remaining in the mixture, which will be the amount of water-soluble protein (i.e. the partially hydrolysed collagen) in the mixture. The amount of protein remaining in the mixture (in g/cm<sup>3</sup>) divided by the amount of protein initially added to the water (in g/cm<sup>3</sup>) gives the amount wt% of water-soluble partially hydrolysed collagen. The amount of protein remaining in the mixture may be determined using any suitable method, e.g. the biuret method that uses tartrate as a reagent in an alkaline copper sulphate solution, and in which the concentration of the protein in the mixture is determined by a colorimetric test using UV/VIS spectroscopy at wavelength 546 nm at 25°C, using a plastic or glass cell with a path length of 1 cm, and the absorbance being measured against a calibration curve that can be produced using standardized protein (e.g. BSA) stock solutions (available commercially, e.g. from Sigma Aldrich or Merck, e.g. available from Merck using the order number 1.10307.0500).

The edible animal chew may comprise 5 wt.% or more of a water-soluble component of partially hydrolysed collagen. In some examples, the edible animal chew comprises 6 wt.% or more, in some examples, 7 wt.% or more, in some examples, 8 wt.% or more, in some examples, 9 wt.% or more, in some examples, 10 wt.% or more, in some examples, 11 wt.% or more, in some examples, 12 wt.% or more, in some examples, 13 wt.% or more, in some examples, 14 wt.% or more, in some examples, 15 wt.% or more, in some examples, 16 wt.% or more, in some examples, 17 wt.% or more, in some examples, 18 wt.% or more, in some examples, 19 wt.% or more, in some examples, 20 wt.% or more, in some examples, 21 wt.% or more, in some examples, 22 wt.% or more, in some examples, 23 wt.% or more, in some examples, 24 wt.% or more, in some examples, 25 wt.% or more, in some examples, 26 wt.% or more, in some examples, 27 wt.% or more, in some examples, 28 wt.% or more, in some examples, 29 wt.% or more, in some examples, about 30 wt.% of a water-soluble component of partially hydrolysed collagen.

The edible animal chew may comprise 30 wt.% or less of a water-soluble component of partially hydrolysed collagen. In some examples, the edible animal chew comprises 29 wt.% or less, in some examples, 28 wt.% or less, in some examples, 27 wt.% or less, in some examples, 26 wt.% or less, in some examples, 25 wt.% or less, in some examples, 24 wt.% or less, in some examples, 23 wt.% or less, in some examples, 22 wt.% or less, in some examples, 21 wt.% or less, in some examples, 20 wt.% or less, in some examples, 19 wt.% or less, in some examples, 18 wt.% or less, in some

examples, 17 wt.% or less, in some examples, 16 wt.% or less, in some examples, 15 wt.% or less, in some examples, 14 wt.% or less, in some examples, 13 wt.% or less, in some examples, 12 wt.% or less, in some examples, 11 wt.% or less, in some examples, 10 wt.% or less, in some examples, 9 wt.% or less, in some examples, 8 wt.% or less, in some examples, 7 wt.% or less, in some examples, 6 wt.% or less, in some examples, about 5 wt.% or less.

The edible animal chew may comprise 5 wt.% to 30 wt.%, in some examples, 6 wt.% to 29 wt.%, in some examples, 7 wt.% to 28 wt.%, in some examples, 8 wt.% to 27 wt.%, in some examples, 9 wt.% to 26 wt.%, in some examples, 10 wt.% to 25 wt.%, in some examples, 11 wt.% to 24 wt.%, in some examples, 12 wt.% to 23 wt.%, in some examples, 13 wt.% to 22 wt.%, in some examples, 14 wt.% to 21 wt.%, in some examples, 15 wt.% to 20 wt.%, in some examples, 16 wt.% to 19 wt.%, in some examples, 17 wt.% to 18 wt.% of a water-soluble component of partially hydrolysed collagen e. In some examples, the edible animal chew may comprise 5 wt.% to 10 wt.%, in some examples, 10 wt.% to 15 wt.%, in some examples, 15 wt.% to 20 wt.% of a water-soluble component of partially hydrolysed collagen.

It should be noted that the properties of the partially hydrolysed collagen differ from those of a mixture formed by mixing natural collagen (which is substantially water-insoluble) with gelatine (which is water soluble). In particular, the tensile toughness of an edible animal chew obtained by using partially hydrolysed collagen is greater than that obtained by using a mixture formed by mixing the equivalent amount of natural collagen and gelatine.

The partially hydrolysed collagen may comprise a 30:70 to 70:30 (wt:wt) mixture of water-soluble component and non-water soluble component. In some examples, the partially hydrolysed collagen may comprise a 35:65 to 65:35 (wt:wt) mixture, in some examples, a 40:60 to 60:40 (wt:wt) mixture, in some examples, a 45:55 to 55:45 (wt:wt) mixture, in some examples, about a 50:50 (wt:wt) mixture of a water-soluble component and a non-water-soluble component.

The edible animal chew may comprise (i) partially hydrolysed collagen and (ii) additional natural collagen. In some examples, all of the collagen in the edible animal chew may be partially hydrolysed collagen. In some examples, the additional natural

collagen may constitute 1 wt.% or more of the edible animal chew. In some examples, the additional natural collagen may constitute 2 wt.% or more, in some examples, 3 wt.% or more, in some examples, 4 wt.% or more, in some examples, 5 wt.% or more, in some examples, 10 wt.% or more, in some examples, 15 wt.% or more, in some examples, 16 wt.% or more, in some examples, 17 wt.% or more, in some examples, 18 wt.% or more, in some examples, 19 wt.% or more, in some examples, 20 wt.% or more, in some examples, 21 wt.% or more, in some examples, 22 wt.% or more, in some examples, 23 wt.% or more, in some examples, 24 wt.% or more, in some examples, 25 wt.% or more, in some examples, 26 wt.% or more, in some examples, 27 wt.% or more, in some examples, 28 wt.% or more, in some examples, 29 wt.% or more, in some examples, about 30 wt.% of the edible animal chew. In some examples, the additional natural collagen may constitute 30 wt.% or less, in some examples, 29 wt.% or less, in some examples, 28 wt.% or less, in some examples, 27 wt.% or less, in some examples, 26 wt.% or less, in some examples, 25 wt.% or less, in some examples, 24 wt.% or less, in some examples, 23 wt.% or less, in some examples, 22 wt.% or less, in some examples, 21 wt.% or less, in some examples, 20 wt.% or less, in some examples, 19 wt.% or less, in some examples, 18 wt.% or less, in some examples, 17 wt.% or less, in some examples, 16 wt.% or less, in some examples, 15 wt.% or less, in some examples, 10 wt.% or less, in some examples, 5 wt.% or less, in some examples, 4 wt.% or less, in some examples, 3 wt.% or less, in some examples, 2 wt.% or less, in some examples, about 1 wt.% of the edible animal chew. In some examples, the additional natural collagen may constitute 1 wt.% to 30 wt.% , in some examples, 2 wt.% to 29 wt.% , in some examples, 3 wt.% to 28 wt.% , in some examples, 4 wt.% to 27 wt.%, in some examples, 5 wt.% to 26 wt.%, in some examples, 10 wt.% to 25 wt.%, in some examples, 15 wt.% to 24 wt.%, in some examples, 16 wt.% to 23 wt.%, in some examples, 17 wt.% to 22 wt.%, in some examples, 18 wt.% to 21 wt.%, in some examples, 19 wt.% to 20 wt.% of the edible animal chew. In some examples, the additional natural collagen comprises 1 wt.% to 5 wt.%, in some examples, 15 wt.% to 25 wt.%, in some examples, 20 wt.% to 30 wt.% of the edible animal chew. The “additional natural collagen” may be collagen extract having a low fat content, e.g. a fat content of 8wt% or less, preferably 7wt% or less, preferably 6wt% or less, preferably 5wt% or less and/or a high protein content, e.g. a protein content of at least 800 g/kg of the extract (g/kg), optionally at least 850 g/Kg, optionally at least 875 g/kg, optionally at least 900 g/kg (e.g. as measured according to ISO 5983:1998). Natural collagen, as used herein, indicates that the collagen may

have been extracted, but has not been hydrolysed or detangled, such that most (e.g. at least 90wt%, e.g. at least 95wt%) or all of the collagen is present in the form of fibrils comprising tropocollagen triple helices and there has been little or no detangling of the fibrils or of the triple helices and little or no chain shortening of the proteins (e.g. due to hydrolysis). Natural collagen extract, e.g. with low fat content, may be obtained commercially, e.g. products sold under the trade name Valocoll, which is a natural porcine collagen extract, available from Sonac / Darling Ingredients.

In some examples, the edible animal chew comprises a mixture of (i) partially hydrolysed collagen and (ii) additional natural collagen in which 10 wt.% to 100 wt.% of the total amount of collagen is partially hydrolysed collagen. In some examples, the edible animal chew comprises (i) partially hydrolysed collagen and (ii) additional natural collagen in which 10 wt.% to 30 wt.%, in some examples, 15 wt.% to 27 wt.%, in some examples, 20 wt.% to 26 wt.%, in some examples, 21 wt.% to 25 wt.%, in some examples, 22 wt.% to 24 wt.%, in some examples, 23 wt.% to 24 wt.% of the total amount of collagen is partially hydrolysed collagen. In some examples, the edible animal chew comprises (i) partially hydrolysed collagen and (ii) additional natural collagen in which 55 wt.% to 75 wt.%, in some examples, 60 wt.% to 70 wt.%, in some examples, 61 wt.% to 69 wt.%, in some examples, 62 wt.% to 69 wt.%, in some examples, 63 wt.% to 68 wt.%, in some examples, 64 wt.% to 68 wt.%, in some examples, 65 wt.% to 67 wt.%, in some examples, 66 wt.% to 67 wt.% of the total amount of collagen is partially hydrolysed collagen. In some examples, the edible animal chew comprises (i) partially hydrolysed collagen and (ii) additional natural collagen, wherein the relative wt% of (i):(ii) is from 80:20 to 20:80, optionally 70:30 to 30:70, optionally 60:40 to 40:60, optionally 45:55 to 55:45, optionally about 50:about 50. The "additional natural collagen" may be collagen extract having a low fat content, e.g. a fat content of 8wt% or less, preferably 7wt% or less, preferably 6wt% or less, preferably 5wt% or less and/or a high protein content, e.g. a protein content of at least 800 g per kg of the extract (g/kg), optionally at least 850 g/Kg, optionally at least 875 g/kg, optionally at least 900 g/kg (e.g. as measured according to ISO 5983:1998). Optionally, the additional natural collagen may have a fat content of from 8 to 30 wt%, optionally from 8 to 20 wt%.

Collagen is typically obtained in an extract form, and the extract is normally a mixture of the collagen compound and impurities, such as fat. Preferably, the collagen used in the composition or method has a low fat content. Preferably, the fat content in the collagen used in the composition or method is 8wt% or less, preferably 7 wt% or less, preferably 6 wt% or less, preferably 5wt% or less (e.g. as measured by ISO 5983).  
5 "The collagen used in the composition or method" may be the partially hydrolysed collagen and/or any other collagen used in the composition/method, such as the "additional natural collagen", which may be collagen extract having a low fat content, e.g. a fat content of 8wt% or less, preferably 7 wt% or less, preferably 6 wt% or less, preferably 5wt% or less and/or a high protein content, e.g. a protein content of at least  
10 800 g per kg of the extract (g/kg), optionally at least 850 g/Kg, optionally at least 875 g/kg, optionally at least 900 g/kg (e.g. as measured according to ISO 5983:1998).

In some examples, the edible animal chew comprises unmodified starch. In some  
15 examples, the unmodified starch may be derived from corn, wheat, modified wheat, tapioca, sorghum, potato, sweet potato, rice, oat, beets, barley, soy, other cereals or grains, and mixtures thereof. The unmodified starch may be maize starch or potato starch. The potato starch may be high viscosity potato starch. Tapioca starch, pea starch, mixtures thereof or mixtures of tapioca starch and/or pea starch and any of the  
20 aforementioned types may also be used. The starch used may be one type of starch or may alternatively comprise a mixture of starches. Pure or substantially pure starches may be used if desired. The type(s) of starch(es) used may be characterised by starch profiles having all possible proportions of amylopectin, intermediates and amylose. The exact source(s) of starch used is not critical. In general, the starch source(s) is(are)  
25 selected on the basis of cost and palatability considerations. The unmodified starch may comprises maize starch, which, when included in an edible animal chew, produces an animal chew with higher strength characteristics than other starches, e.g. potato starch.

30 The edible animal chew may comprise 5 wt.% or more unmodified starch. The edible animal chew may comprise 7 wt.% or more, in some examples, 10 wt.% or more, in some examples, 11 wt.% or more, in some examples, 12 wt.% or more, in some examples, 13 wt.% or more, in some examples, 14 wt.% or more, in some examples, 15 wt.% or more, in some examples, 17 wt.% or more, in some examples, 20 wt.% or  
35 more, in some examples, 21 wt.% or more, in some examples, 22 wt.% or more, in

some examples, 23 wt.% or more, in some examples, 24 wt.% or more, in some examples, 25 wt.% or more, in some examples, 27 wt.% or more, in some examples, 30 wt.% or more, in some examples, 33 wt.% or more, in some examples, 35 wt.% or more, in some examples, 36 wt.% or more, in some examples, 37 wt.% or more, in some examples, 38 wt.% or more, in some examples, 39 wt.% or more, in some examples, 40 wt.% or more, in some examples, 45 wt.% or more, in some examples, about 50 wt.% unmodified starch.

The edible animal chew may comprise 50 wt.% or less unmodified starch. The edible animal chew may comprise 45 wt.% or less, in some examples, 40 wt.% or less, in some examples, 39 wt.% or less, in some examples, 38 wt.% or less, in some examples, 37 wt.% or less, in some examples, 36 wt.% or less, in some examples, 35 wt.% or less, in some examples, 33 wt.% or less, in some examples, 30 wt.% or less, in some examples, 27 wt.% or less, in some examples, 25 wt.% or less, in some examples, 24 wt.% or less, in some examples, 23 wt.% or less, in some examples, 22 wt.% or less, in some examples, 21 wt.% or less, in some examples, 20 wt.% or less, in some examples, 17 wt.% or less, in some examples, 15 wt.% or less, in some examples, 14 wt.% or less, in some examples, 13 wt.% or less, in some examples, 12 wt.% or less, in some examples, 11 wt.% or less, in some examples, 10 wt.% or less, in some examples, 7 wt.% or less, in some examples, about 5 wt.% unmodified starch.

The edible animal chew may comprise 5 wt.% to 50 wt.% unmodified starch. The edible animal chew may comprise 7 wt.% to 45 wt.%, in some examples, 10 wt.% to 40 wt.%, in some examples, 11 wt.% to 39 wt.%, in some examples, 12 wt.% to 38 wt.%, in some examples, 13 wt.% to 37 wt.%, in some examples, 14 wt.% to 36 wt.%, in some examples, 15 wt.% to 35 wt.%, in some examples, 17 wt.% to 33 wt.%, in some examples, 20 wt.% to 30 wt.%, in some examples, 21 wt.% to 27 wt.%, in some examples, 22 wt.% to 25 wt.%, in some examples, 23 wt.% to 24 wt.% unmodified starch. The edible animal chew may comprise 5 wt.% to 20 wt.%, in some examples, 17 wt.% to 27 wt.%, in some examples, 30 wt.% to 40 wt.% unmodified starch.

“Unmodified starch” indicates that the starch has not had any chemical species covalently bound to it, e.g. it has not had an alkyl succinate bound to it. However “unmodified starch” may be gelatinized or ungelatinized starch. Gelatinized starch is produced when starch is heated in water, e.g. if the edible animal chew is produced

using extrusion in which a composition comprising starch and water is heated. At least a portion of the unmodified starch may be gelatinized unmodified starch. The term “gelatinized unmodified starch” as used herein means unmodified starch that has been processed in the presence of water such that its native granular structure has been destroyed, that the crystalline regions of the unmodified starch have been melted, and the starch converted to a water-soluble form of amylose molecules. Importantly, the effect of such processing is to convert the native unmodified starch, which is essentially indigestible, into a form which is digestible.

The edible animal chew may comprise a plasticiser. The plasticizer may be or comprise water. The plasticiser may comprise water and another plasticiser. The plasticizer or additional plasticiser may comprise a polyol, esters of citric acid or urea. Suitable polyols include glycol, diethylene glycol, alkylene glycols, polyalkylene glycol, sorbitol, glycerol, glycerol monoesters and the like. The plasticiser may comprise glycerol, water or a mixture thereof. The glycerol and/or glycol may function as both a plasticiser and a humectant.

The edible animal chew may comprise a humectant. The humectant may comprise sucrose, sodium chloride, sorbitol, glycerine, starch hydrolysate, glucose, maltose, lactose, gums, galactose, citric acid, alanine, glycine, high fructose corn syrup, tartaric acid, malic acid, xylose, PEG 400, PEG 600, propylene glycol, aminobutyric acid, mannitol, mannose, or lactulose. In some examples, the humectant may be selected from propylene glycol, glycerine, and starch hydrolysate, and combinations thereof.

In embodiment, at least 60wt% of the animal chew comprises partially hydrolysed collagen and/or alkyl succinate modified starch, and optionally a component selected from natural collagen a plasticizer (excluding water), and unmodified starch. In embodiment, at least 70 wt%, optionally at least 75wt%, optionally at least 80wt%, optionally at least 85wt%, optionally at least 90wt% of the animal chew comprises partially hydrolysed collagen and/or alkyl succinate modified starch, and optionally a component selected from natural collagen, a plasticizer (excluding water), and unmodified starch. The remaining weight % may be components other than those listed in this paragraph and those components may be selected from, for example, water, salt, flavourings, palatants, meat powder, e.g. liver powder, vitamins, minerals, flour and preservatives.

In embodiment, at least 70wt% of the animal chew comprises partially hydrolysed collagen and/or alkyl succinate modified starch, a plasticizer (excluding water), and optionally, unmodified starch and/or natural collagen. In embodiment, at least 70wt%  
5 of the animal chew comprises partially hydrolysed collagen, alkyl succinate modified starch, a plasticizer (excluding water), unmodified starch and natural collagen. In embodiment, at least 75wt%, optionally at least 80wt%, optionally at least 85wt%, optionally at least 90wt% of the animal chew comprises partially hydrolysed collagen and/or alkyl succinate modified starch, a plasticizer (excluding water), and optionally,  
10 unmodified starch and/or natural collagen. The remaining weight % may be components other than those listed in this paragraph and those components may be selected from, for example, water, salt, flavourings, palatants, meat powder, e.g. liver powder, vitamins, minerals, flour and preservatives,

15 Preferably, the edible animal chew comprises less than 10 wt% fat, optionally less than 8wt% fat, optionally less than 7wt% fat, optionally less than 6wt% fat, optionally less than 5wt% fat, optionally less than 4wt% fat, optionally less than 3wt% fat. This has been found to produce a chew that can be formed easily, and expanded if desired, e.g. formed in an extrusion process, and have desired chew characteristics.

20 The edible animal chew may be an elongate edible animal chew. The elongate animal chew may have a length, which is along the longest dimension of the animal chew, and a cross section, perpendicular to the length, and the shape of the cross section may be substantially constant along the length of the animal chew. The cross-sectional shape  
25 of the animal chew may be any regular or irregular shape. The cross-sectional shape may be selected from a circle, an oval, or an n-sided regular shape, and optionally n is selected from 3 to 8, optionally 4, 5, 6 or 7. The regular shape may, for example, be selected from a triangle, a square, a rectangle, a hexagon, a heptagon and an octagon.

30 The elongate animal chew may have an internal cellular structure (e.g. it is an expanded, e.g. thermally expanded, animal chew in the form of a solid foam) and/or have a cross-sectional area (i.e. the area of the cross section perpendicular to its length) of at least 900 mm<sup>2</sup>, optionally at least 1000 mm<sup>2</sup>, optionally at least 2000 mm<sup>2</sup>, optionally at least 2000 mm<sup>2</sup>, optionally at least 3000 mm<sup>2</sup>, optionally at least 3500  
35 mm<sup>2</sup>, optionally at least 3700 mm<sup>2</sup>, optionally at least 4000 mm<sup>2</sup>. The elongate animal

chew may have an internal cellular structure (e.g. it is an expanded animal chew in the form of a solid foam) and/or have a cross-sectional area (i.e. the area of the cross section perpendicular to its length) of from 900 mm<sup>2</sup> to 5500 mm<sup>2</sup>, optionally 1000 mm<sup>2</sup> to 5500 mm<sup>2</sup>, optionally 2000 mm<sup>2</sup> to 5500 mm<sup>2</sup>, optionally 3000 mm<sup>2</sup> to 5500 mm<sup>2</sup>,  
5 optionally 4000 mm<sup>2</sup> to 5500 mm<sup>2</sup>.

The elongate edible animal chew may have at least one channel extending at least part way through, optionally all the way through, the animal chew, along the length of the animal chew (i.e. the longest dimension of the animal chew).

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The edible animal chew may have an internal cellular structure. The edible animal chew may be in the form of a solid foam. The internal cellular structure may be in the form of pores, which may be open or closed pores, which may have been formed, for example, by the expansion of gases within and/or the passage of gases through the  
15 edible animal chew during formation and before solidification, e.g. while being formed by extrusion. The gases may be selected from steam, air, nitrogen and a supercritical fluid, which may be selected from nitrogen and carbon dioxide. The edible animal chew may be a thermally expanded edible animal chew or an aerated animal chew.

20 The edible animal chew (or the material from which it is made) may have a tensile toughness of at least 200 MPa, optionally at least 300 MPa, optionally at least 400 MPa. The edible animal chew (or the material from which it is made) may have a tensile toughness of from 200 MPa to 800 MPa, optionally from 200 MPa to 700 MPa, optionally from 200 MPa to 600 MPa, optionally from 250 MPa to 600 MPa. The tensile  
25 toughness may be determined by plotting stress (in MPa) vs strain (in %), up until the breaking point, and determining the area beneath the curve (from 0% strain to the breaking point), which gives the tensile toughness in MPa.

The general method for determining the toughness may be as follows:

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- (i) cut the material of the edible animal chew into to a tensile bar by using an ISO 527-2 stamp (Zwick Roell; e.g. see Figure 9C);
- (ii) provide a tensile rig having upper and lower grips, and place the tensile bar into the lower grips of the tensile rig (e.g. as shown in Figure 9D; so that the full  
35 shoulder of the test piece is exposed) and tighten to hold the product (e.g.

enough that the product does not slip during testing, but not so tight that the product breaks);

(iii) position the upper grips of the tensile rig to align with the test piece (again, so that the full shoulder of the test piece is exposed). Tighten to hold the product, as for the lower grips.

(iv) minimise any tension or slack in the test piece;

(v) move the upper and lower grips apart by the texture analyser (TA) at 1 mm/s and the resistive force is recorded by the sensor through the breaking point of the test piece (the tensile bar).

The following equipment and apparatus may used (see Figure 9):

(i) Stable Micro Systems TA-HD.plus;

(ii) Tensile rig (A/HDT) – Max. load 500 kg;

(iii) Secateurs (only required for testing a commercially-available edible animal chew);

(iv) Tensile cutter (ISO 527-2; 75mm long, 10mm wide, 5mm in centre) – Zwick Roell;

(v) 100 or 500kg load cell.

The water content of the edible animal chew may be from about 5 to 20 wt%, optionally from about 8 to about 16 wt%, optionally about 10 to about 15 wt%, optionally about 11 to 14wt%, relative to the total weight of the chew.

In an embodiment, the edible animal chew of the present invention exhibits a cohesiveness measured by Texture Profile Analysis (as described herein) of 0.55 or greater. Preferably, the cohesiveness is 0.57 or greater, optionally 0.60 or greater, optionally 0.62 or greater, optionally 0.65 or greater, optionally 0.68 or greater. The higher the cohesiveness value, the greater the propensity of the chew to retain its structure while being chewed; this may assist in improving the cleaning properties of the chew.

The density of the edible animal chew may be 1.5 g cm<sup>-3</sup> or less, optionally 1.2 g cm<sup>-3</sup> or less, optionally 1 g cm<sup>-3</sup> or less. In other embodiments, the density of the edible animal chew is 0.95 g cm<sup>-3</sup> or less, or 0.90 g cm<sup>-3</sup> or less, or 0.80 g cm<sup>-3</sup> or less.

Preferably the density is less than 0.80 g cm<sup>-3</sup>, optionally 0.75 g cm<sup>-3</sup> or less. The

density of the edible animal chew may be from 0.5 g cm<sup>-3</sup> to 1.5 g cm<sup>-3</sup>, optionally from 0.5 g cm<sup>-3</sup> to 1.2 g cm<sup>-3</sup>, optionally from 0.5 g cm<sup>-3</sup> to 1 g cm<sup>-3</sup>, optionally from 0.6 g cm<sup>-3</sup> to 0.9 g cm<sup>-3</sup>, optionally from 0.6 g cm<sup>-3</sup> to 0.8 g cm<sup>-3</sup>, optionally from 0.7 g cm<sup>-3</sup> to 0.9 g cm<sup>-3</sup>, optionally from 0.7 g cm<sup>-3</sup> to 0.8 g cm<sup>-3</sup>.

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"Water activity" may be a measurement of the energy status of the water in a system; represented by a quotient between water's partial pressure in the food and pure water's partial pressure. It indicates how tightly water is bound, structurally or chemically, within a substance. This is measured by equilibrating the liquid phase (in the sample) with the vapour phase (in the headspace) and measuring the relative humidity of that space. The water activity (Aw) of the animal chew may be from about 0.40 to about 0.85, optionally from about 0.50 to about 0.85, more preferably from about 0.50 to about 0.80, and more preferably from about 0.60 to about 0.76, even more preferably from about 0.60 to about 0.70.

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In some examples, the edible animal chew is a thermally expanded edible animal chew. In some examples, thermal expansion provides an aerated structure to the edible animal chew.

20 In some examples, the animal is a canine. In some examples, the canine is a dog.

In an aspect, there is also provided: a method of making an edible animal chew, the method comprising:

- a. providing an edible chew composition comprising:
  - 25 partially hydrolysed collagen; and/or
  - alkyl succinate modified starch
- b. heating the edible chew composition to form a flowable edible chew composition;
- c. extruding the flowable edible chew composition to form an extrudate;
- 30 d. allowing the extrudate to cool and harden to form the edible animal chew.

The extrusion is carried out in an extruder. The flowable edible chew mixture may leave the extruder at a temperature above the melting point of the collagen. The flowable edible chew mixture may leave the extruder at a temperature such that steam is produced from water within the composition. The flowable edible chew mixture may

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leave the extruder at a temperature of 100 °C or more, and/or preferably a temperature of 150 °C or less. By extruding the composition, such that it exits the extruder at a temperature of 100 °C or more, this will, under normal ambient pressure (e.g. about 100 kPa), generate steam. The steam then causes expansion of the product, e.g. forming a cellular structure within the animal chew or, in other words, a foamed chew, which becomes a solid foam when the composition solidifies on cooling. This often leads to the animal chew having a density of 1 .0 g cm<sup>-3</sup> or less.

Optionally, the composition leaves the extruder at a temperature of from about 105°C to about 130°C, preferably at least 110°C to 125°C , and preferably from 115°C to 125°C, optionally from 118°C to 123°C, optionally about 120°C. These temperature ranges have been found to result in an expanded animal chew with desired chew characteristics, include strength. This may be termed thermal expansion of the chew.

Optionally, if the chew is not to be expanded (at least by thermal means), the flowable edible chew mixture may leave the extruder at a temperature of less than 100 °C, e.g. a temperature of between 40 °C and 90 °C, optionally between 50 °C and 70 °C. Preferably, even if the flowable edible chew mixture does not leave the extruder at a temperature of 100 °C or more, preferably the flowable edible chew mixture has been heated to a temperature within the extruder of at least 90 °C, optionally from 90 °C to 120 °C, optionally at least 100 °C, optionally from 100 °C to 120 °C, optionally from 100 °C to 110 °C.

In an embodiment, the edible chew mix is formed in a barrel extruder.

25

In an embodiment, the temperature of the barrel in the barrel extruder increases in the extrusion direction; this is particularly preferred when producing expanded chews, i.e. chews having an internal cellular structure.

30 The extruder may be a single screw extruder or a twin screw extruder.

The screw may rotate at a speed of from 80 rpm to 300 rpm, optionally a speed of from 80 rpm to 200 rpm, optionally a speed of from 80 rpm to 180 rpm, optionally a speed of from 80 rpm to 160 rpm.

35

The specific mechanical energy (SME) applied by the extruder may be from 60 kWhkg<sup>-1</sup> to 120 kWhkg<sup>-1</sup>, optionally from 80 kWhkg<sup>-1</sup> to 120 kWhkg<sup>-1</sup>, optionally from 90 kWhkg<sup>-1</sup> to 115 kWhkg<sup>-1</sup>, optionally from 95 kWhkg<sup>-1</sup> to 115 kWhkg<sup>-1</sup>.

5 In an embodiment, the extrusion is supercritical fluid extrusion, such that expansion of the flowable edible chew mixture is effected, such that, on cooling, the edible chew has an internal cellular structure. The supercritical fluid may be selected from nitrogen and carbon dioxide.

10 In aspect, there is also provided an edible animal chew producible by a method described herein.

Preferably the components of edible chew mixture are introduced into a cooker extruder, preferably a twin-screw cooker extruder, and the cooker extruder, during the  
15 act of extruding the edible chew mixture, subjects it to heating, to produce the flowable edible chew mixture, and shear, to homogenise and mix the components thoroughly, and the flowable edible chew mixture leaves the extruder, optionally with expansion of the mixture such that an internal cellular structure is formed in the mixture, and the edible chew mixture allowed to cool and harden to form the edible animal chew.

20

In an embodiment, there is also provided an edible animal chew comprising a blend of

- (i) a natural collagen extract;
- (ii) a partially hydrolysed collagen.

25 The natural collagen extract and the partially hydrolysed collagen may be as described herein.

The natural collagen extract may have fat content of 40wt% or less, optionally 30wt% or less, optionally from 8wt% to 20wt%, optionally 8wt% or less and/or a protein  
30 content of at least 800 g per kg of the extract. Optionally, (i) and (ii) are present in wt:wt ratio of 20:80 to 80:20. Optionally, the animal chew substantially lacks a modified starch. Optionally, components (i) and (ii) constitute at least 30wt% of the composition, optionally at least 50wt% of the composition, optionally at least 80wt% of the composition, and optionally at least some of the remaining wt% of the edible animal  
35 chew comprising a component selected from an unmodified starch, plasticiser and an

alkyl succinate modified starch. Optionally, the unmodified starch comprises a maize starch.

In an embodiment, there is also provided an edible animal chew comprising a partially  
5 hydrolysed collagen and an alkyl succinate modified starch. The edible animal chew  
may further comprise an unmodified starch. The partially hydrolysed collagen, the alkyl  
succinate modified starch and the unmodified starch may be as defined herein. In an  
embodiment, there is also provided an edible animal chew comprising a partially  
10 hydrolysed collagen and an alkyl succinate modified starch, an unmodified starch and  
protein extract, e.g. natural collagen extract, which may be protein extracted from  
animal skin, e.g. pork skin protein. In an embodiment, there is provided an edible  
animal chew that comprises an unmodified starch, which may be maize starch, partially  
hydrolysed collagen, an alkyl (e.g. octenyl) succinate modified starch, an protein  
15 extract, e.g. natural collagen extract, which may be protein extracted from animal skin,  
e.g. pork skin protein, and optionally other ingredients, which may be selected from  
salt, flavourings, vitamins, minerals, emulsifiers and flour. In an embodiment, there is  
provided an edible animal chew that comprises an unmodified starch in 45 to 55 parts  
by weight, which may be maize starch, partially hydrolysed collagen in 25 to 35 parts  
20 by weight, an alkyl (e.g. octenyl) succinate modified starch in 5 to 15 parts by weight,  
an protein extract, e.g. natural collagen extract, which may be protein extracted from  
animal skin, e.g. pork skin protein, in 5 to 15 parts by weight, and other ingredients in 0  
to 10% in parts by weight, the other ingredients may be selected from salt, flavourings,  
vitamins, minerals, emulsifiers and flour.

25 One of ordinary skill in the art will recognize that additional embodiments or  
implementations are possible without departing from the teachings of the present  
disclosure or the scope of the claims that follow. This detailed description, and  
particularly the specific details of the exemplary embodiments and implementations  
disclosed herein, is given primarily for clarity of understanding, and no unnecessary  
30 limitations are to be understood therefrom, for modifications will become obvious to  
those skilled in the art upon reading this disclosure and may be made without departing  
from the spirit or scope of the claimed invention(s).

**EXAMPLES**

The following illustrate examples of the compositions, methods and other aspects described herein. Thus, these Examples should not be considered as limitations of the present disclosure, but are merely in place to teach how to make embodiments of the edible animal chew, and to illustrate embodiments of the method.

**Example 1 – animal skin protein as a source of collagen**

Edible animal chews formed by combining pork skin protein (PSP) with sodium octenyl succinate starch (NaOSS) in an 80:20 ratio were produced by cooker extrusion. A variety of control samples were formed from 1) 100% pork skin protein; 2) 98.5 wt.% pork skin protein and 1.5 wt.% of a standard emulsifier [diacetyl tartaric acid ester of mono- and diglycerides (DATEM)]; 3) 80 wt.% pork skin protein and 20 wt.% wheat flour (WF); 4) 78.5 wt.% pork skin protein, 20 wt.% wheat flour and 1.5 wt.% DATEM. The control samples were labelled as follows:

<b>Control Sample Name</b>	<b>Control Sample Composition</b>
Powders Sample 1	20 wt.% NaOSS, 80 wt.% PSP
Powders Sample 2	20 wt.% WF, 1.5 wt.% DATEM, 78.5 wt.% PSP
Powders Sample 3	20 wt.% WF, 80 wt.% PSP
Powders Sample 4	98.5 wt.% PSP, 1.5 wt.% DATEM
Powders Sample 5	100 wt.% PSP

These control sample compositions were extruded under the following conditions:

Table 1 – Extrusion conditions

	<b>Powders</b>	<b>Water</b>	<b>Glycerol</b>	<b>Steam</b>	<b>Specific Mechanical Energy (SME)</b>	<b>Screw Speed</b>
	<b>kg/h</b>	<b>kg/h</b>	<b>kg/h</b>	<b>kg/h</b>	<b>Wh/kg</b>	<b>RPM</b>
Powders Sample 1	100	13	15	13	60	225
Powders Sample 2	100	13	15	13	55	400
Powders Sample	100	13	15	13	57	400

	3					
Powders Sample	4	100	14	15	13	39
Powders Sample	5	100	13	15	13	41
						300
						300

In all cases the vacuum was set at -0.6 bar gauge, with barrel temperature control over the seven barrel (B1-B7) extruder set as follows:

5

Nozzle	Barrel Temperature
B1	Uncontrolled
B2	150°C
B3	150°C
B4	Uncontrolled
B5	130°C
B6	110°C
B7	90°C

The tensile toughness for the pork skin protein and sodium octenyl succinate starch chew was compared with those of the control samples (see Figure 1A). As shown in Figure 1A, the tensile toughness of the edible animal chew containing pork skin protein and sodium octenyl succinate starch is significantly higher than those of the control samples. Depending on the control sample used as a reference, the performance improvement ranges from 80% to 200%. It is believed that a reason for this marked improvement in the tensile toughness is due to the clear and large increase in the ratio of SME (mechanical energy transferred by the extruder) to rotations per minute (RPM) seen for samples containing sodium octenyl succinate starch (see Figure 1B).

15

### **Example 2 – partially hydrolysed collagen**

Edible animal chews were formed by combining partially hydrolysed collagen, maize starch, high viscosity potato starch (HV starch) and sodium octenyl succinate starch in varying proportions (including combinations in which at least one ingredient was omitted) and processing the mixture in a cooker extruder. The proportions of each ingredient used are given in Table 2. Although it is possible to extrude formulations

20





<sup>1)</sup>									
<b>B2 (°C)</b>	112	102	105	109	104	113	114	120	101
<b>B3 (°C)</b>	116	106	104	107	106	102	110	107	106
<b>B5 (°C)</b>	83	101	88	85	95	94	88	89	96
<b>B6 (°C)</b>	66	67	66	67	68	66	67	66	65
<b>B7 (°C)</b>	56	62	57	57	58	57	56	58	58
<b>Vacuum (bar)</b>	0.31	0.33	0.31	0.31	0.3	0.31	0.31	0.31	0.3
<b>RPM</b>	155	90	138	140	125	95	140	155	90
<b>SME (kWhkg<sup>-1</sup>)</b>	95	110	92	100	95	95	99	95	101

	<b>N10</b>	<b>N11</b>	<b>N12</b>	<b>N13</b>	<b>N14</b>	<b>N15</b>	<b>N16</b>	<b>N17</b>
<b>Powders (kgh<sup>-1</sup>)</b> <sup>1)</sup>	95	95	95	95	95	95	95	95
<b>Water (kgh<sup>-1</sup>)</b>	17	17	17	17	17	17	17	17
<b>Glycerol (kgh<sup>-1</sup>)</b>	21	21	21	21	21	21	21	21
<b>B2 (°C)</b>	108	114	108	112	97	110	110	109
<b>B3 (°C)</b>	101	119	116	116	112	113	113	111
<b>B5 (°C)</b>	96	96	93	91	99	90	90	89
<b>B6 (°C)</b>	65	67	66	65	78	67	67	66
<b>B7 (°C)</b>	58	59	58	58	71	57	57	57
<b>Vacuum (bar)</b>	0.31	0.31	0.31	0.3	0.32	0.31	0.31	0.3
<b>RPM</b>	95	115	95	140	172	100	100	140
<b>SME (kWhkg<sup>-1</sup>)</b>	100	98	100	93	95	100	100	95

The tensile strength, ductility, toughness, shear force, hardness, expanded cross  
5 section, expanded hot strength and water activity of each edible animal chew formed  
above was determined. The results are provided in Tables 5. Test procedures are  
described later.

Table 5

Blend Code	Tensile Strength/MPa	Ductility/%	Toughness/MPa	Shear Force/kg	Hardness
N1	2.27	67.21	112.83	12.72810	2.454
N2	2.51	310.13	484.77	55.9138	1.808
N3	1.4	278.52	288.05	29.127	1.862
N4	2.2	58.96	94.39	13.4621	2.368
N5	3.06	399.01	681.43	65.7718	1.749
N6	1.68	224.73	251.51	45.5163	1.663
N7	2.15	63.59	99.88	13.7588	2.2
N8	1.5	247.79	282.38	29.1715	1.619
N9	3.17	386.4	702.24	78.4549	1.998
N10	2.93	378.57	664.71	68.2958	1.808
N11	1.95	186.71	256.59	29.6097	1.93
N12	1.94	214.89	303.24	36.2693	2.254
N13	1.6	273.93	318.07	32.0543	1.797
N14	1.26	119.11	123.48	18.1328	2.255
N15	1.72	187.89	239.6	29.0367	1.954
N16	1.73	202.55	250.99	35.293	1.928
N17	1.85	254.17	337.84	33.287	2.003

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Blend Code	Expanded Cross Section/mm <sup>2</sup>	Expanded Hot Strength/%	Water Activity/1
N1	992	16	0.755
N2	3385	87	0.682
N3	4077	69	0.702
N4	1032	14	0.71
N5	3046	81	0.692
N6	4170	75	0.694

N7	1092	19	0.665
N8	6293	73	0.691
N9	3332	81	0.66
N10	3785	77	0.679
N11	5111	73	0.69
N12	3956	80	0.707
N13	4801	73	0.672
N14	4520	63	0.698
N15	4222	83	0.664
N16	4990	79	0.685
N17	4798	72	0.684

### Expansion Characteristics

The behaviour of partially hydrolysed collagen formulations under thermal expansion was investigated. In order to achieve this the extruder was reconfigured with a hollow cylinder nozzle. All other aspects of the extruder configuration were equivalent. Thermally expanding the material into a hollow cylindrical shape has been found to provide a useful gauge for comparing expansion properties as the shape is prone to sagging presenting a useful “worst case” scenario. This is due to the void in the middle of the shape. The outer wall of the hollow cylinder nozzle is perfectly circular.

### *Definition of “Expanded Hot Strength”*

The product is placed on a bench to cool following extrusion and thermal expansion. On the bench the product will sag and distort if the material does not have enough strength when hot. It is critical that this effect be minimised in order for a viable product to be made in a desired shape. After cooling we can measure two attributes of the expansion using simple geometry. These are termed the Expanded Hot Strength (%) which is essentially a ratio of the height (H) and width (W) on the cut face of an extruded cylindrical product. For a product that does not sag these two parameters would be the same and the Expanded Hot Strength would be 100%. For a product that totally collapses the width (W) would be much larger than the height (H) giving rise to

an Expanded Hot Strength that tends towards 0%. This is defined mathematically in Equation 1.

$$\text{Expanded Hot Strength (\%)} = \frac{H}{W} \times 100 \quad (1)$$

5

*Definition of "Expanded Cross Section"*

A key benefit of thermal expansion is that the volume of the product can be much larger for a given weight (low density). Larger chews are more challenging to eat and last longer, increasing stimulation for the animal. For this reason, the size to which a material can be expanded and maintain its shape is an important parameter. This has been compared by measuring the area of the cross-section of the chew, which can be approximated to an elliptical shape after sagging. This is defined mathematically in Equation 2.

15

$$\text{Expanded Cross Section (mm}^2\text{)} = \pi HW \quad (\text{Area of an ellipse}) \quad (2)$$

Figure 2 shows how the expanded hot strength (%) changes when the proportions of maize starch, HV potato starch and partially hydrolysed collagen are altered with no inclusion (Figure 2A) and with 10 wt.% of hydrolysed sodium octenyl succinate starch (Figure 2B). (The triangular contour plots were of Figures 2A and 2B, and those in the other Figures, were produced using "MODDE" software, available from "Umetrics".) In both cases the overriding trend is an increase in hot strength with increasing inclusion of partially hydrolysed collagen. Two subtler trends can be observed. The first is that in both plots, maize starch is more conducive to driving hot strength than HV potato starch, especially at higher inclusions of partially hydrolysed collagen. Secondly, a slightly higher hot strength is possible when hydrolysed sodium octenyl succinate starch is not included in the recipe (although this effect is minor).

Figure 3 shows how the expanded cross section (mm<sup>2</sup>) changes when the proportions of maize starch, HV potato starch and partially hydrolysed collagen are altered with no inclusion (Figure 3A) and with 10 wt.% inclusion of hydrolysed sodium octenyl succinate starch (Figure 3B). The overriding trend is that the expanded cross section (mm<sup>2</sup>) increases with increasing inclusion of partially hydrolysed collagen up to a

maximum point after which the inclusion of further partially hydrolysed collagen gives rise to a reduction in the expanded cross section. The highest expanded cross section in the study is achieved in a formulation with 10 wt.% hydrolysed sodium octenyl succinate starch, 30 wt.% to 50 wt.% HV potato starch and 35 wt.% to 60 wt.% partially hydrolysed collagen with the absence of maize starch.

#### Expanded Product Appearance

The appearance of the products extruded under thermal expansion was compared. In general, very low levels of partially hydrolysed collagen (N1, N4 and N7) resulted in total collapse of the products whilst very high levels of partially hydrolysed collagen (N2, N5, N9 and N10) resulted in a dull brown product with small bubbles and a “breadly” appearance. Larger bubble structures were visible for products with higher values for the expanded cross section parameter which tend to be in the mid-range of partially hydrolysed collagen inclusion (N3, N6, N8, N11, N12, N13, N14, N15, N16, and N17).

#### Tensile Properties

The overriding trend in the tensile properties of the extruded products are that the tensile strength (Figure 4), ductility and toughness all increase with increasing inclusion of partially hydrolysed collagen. Maize starch provides greater tensile strength than HV potato starch. Slightly tougher textures are generated when 10 wt.% hydrolysed sodium octenyl succinate starch is included. A description of the texture methods used to generate this data can be found below.

#### Hardness and Shear Resistance

The shear resistance (Figure 5) increases substantially with increase inclusion of partially hydrolysed collagen. The hardness (Figure 6) as measured with the “conical probe” method broadly decreases with increased inclusion of partially hydrolysed collagen. This shows a benefit of using partially hydrolysed collagen in dog chew formulations as the material is more difficult to break down by chewing (shear resistant) whilst being less likely to induce tooth fracture (softer texture). The function is enhanced whilst safety is improved. The trends are broadly the same with 10 wt.%

hydrolysed sodium octenyl succinate starch as without its inclusion. A description of the texture methods used to generate this data can be found below.

### **Example 3**

5

Several other edible animal chews containing partially hydrolysed collagen and sodium octenyl succinate starch have been formulated by the inventors, with various additional components, which may include concentrated collagen extract, unmodified starch (maize starch and/or HV potato starch), plasticisers (water and/or glycerol) and various  
10 flavourings and other additives. The formulations are given in Table 6. Various tests were performed in these formulations, with the results given in Table 7.

Table 6

	Thermal Expansion			
	Chewhide Mark I [wt.%]	Chewhide Mark II Alternate A [wt.%]	Chewhide Mark II Alternate B [wt.%]	Chewhide Mark II Alternate C [wt.%]
Partially hydrolysed bovine collagen	30-40	10-30	10-30	10-30
Natural porcine collagen extract	—	1-5	—	—
Pork skin protein	—	—	1-5	1-5
Glycerol	10-20	10-20	10-20	10-20
Water	10-15	10-15	10-15	10-15
Maize Starch	10-15	35-40	35-40	35-40
HV Starch	5-15	—	—	—
OSA starch	5-10	1-5	1-5	—
Modified OSA starch	—	—	—	1-5
Other additives (including salt, flavourings, vitamins, minerals, emulsifier, flour)	1-10	1-10	1-10	1-10
	Gas Expansion			
	Chewhide Mark II Alternate A [wt.%]	Chewhide Mark II Alternate B [wt.%]	Chewhide Mark II Alternate A [wt.%]	Chewhide Mark II Alternate B [wt.%]
Partially hydrolysed bovine collagen	20-30	20-30	20-30	20-30
Natural porcine	20-30	20-30	—	—

collagen extract				
Drinde 1015 / SF	—	—	20-30	20-30
Glycerol	10-20	10-20	10-20	10-20
Water	10-20	5 -12	5 -12	5 -12
OSA starch	10-15	—	—	—
Modified OSA starch	—	10-15	10-15	10 -20
Other additives (including salt, flavourings, vitamins, minerals, emulsifier, flour)	1-10	1-10	1-10	1-10
Collagen Peptides or Wheat Gluten Hydrosylate	—	1-10	1-10	1-10

The Chewhide Mark II trial was formulated using the following throughput [kg/h] settings:

SCF Flow Rate [kg/h]	Gas Type	Glycerol	Water	Powder Blend
0.0151	N <sub>2</sub>	22.10	18.00	98.10

- 5 'Partially hydrolysed collagen' contains approximately 50 wt.% water-insoluble component and 50 wt.% water soluble component) and is derived from bovine raw materials by mechanical and heat treatment, as described herein. It is supplied in powder form.
- 10 OSA indicates sodium octenyl succinate starch, supplied in particulate form.

HV Starch is potato starch.

- 15 'Natural porcine collagen extract' is a concentrated collagen extracted from natural food-grade porcine bones. It comprises at least 920 g/kg of protein (according to ISO 5983:1998) and at most 40 g/kg fat. It is supplied in particulate form.

- 20 'Pork skin protein' is pork skin protein that contains 10-15% fat and supplied in particulate form. It may be obtained commercially, e.g. under the trade name Drinde 1015/SF, available from Essentia Protein Solutions.

Table 7

	<i>Mark I</i>	<i>Mark II</i>	<i>Commercially- Available Product 3 (‘CAP3’) PCT/GB14/050964</i>			<i>Rawhide Roll</i>	<i>Commercially- Available Product 4</i>
<b>Hardness [kg]</b>	127.264	218.16	174.25	138.8	114.05	618.188	96.761
<b>Stress [kg/mm<sup>2</sup>]</b>	0.113	0.314	0.177	0.164	0.121	0.580	0.065
<b>IRS</b>	0.607	0.306	0.52	0.516	0.587	0.418	0.465
<b>RRS</b>	0.895	0.702	0.771	0.758	0.809	0.568	0.811
<b>Cohesiveness</b>	0.719	0.565	0.657	0.643	0.632	0.354	0.422
<b>Resilience</b>	0.265	0.143	0.232	0.209	0.205	0.175	0.129
<b>Density [g/cm<sup>3</sup>]</b>	0.718	0.728	0.644	0.628	0.601	0.527	0.660

IRS indicates instantaneous recoverable springiness; RRS indicates retarded recoverable springiness, cohesiveness therefore represents how well the product withstands a second deformation relative to how it behaved under the first deformation, 5 resilience is indicative of how much work the sample does in trying to regain its original shape; IRS, RRS, cohesiveness and resilience may be measured in accordance with the methods described in WO2014155113 or US2016100553.

#### **Example 4**

10

A study was conducted to compare three collagen supply options namely pork skin protein, partially hydrolysed collagen and concentrated collagen extract. A control sample was made using the formulation of a commercially-available edible animal chew (denoted CAP2 in the Figures) for comparison against an existing technology.

15

As hydrolysed sodium octenyl succinate starch (OSA) is required to aid the processing of pork skin protein, all of the powder formulations were normalised to comprise 80 wt.% of the respective collagen source and 20 wt.% hydrolysed sodium octenyl succinate starch. Each formulation was treated under the cooker extrusion process

conditions shown in Table 8. These conditions gave extruded materials that served as test pieces for comparison of the textural properties delivered by each of the collagen sources.

5 Table 8

	Control (Commercially-Available Edible Animal Chew, CAP2)	80 wt.% Pork Skin Protein & 20 wt.% OSA starch	80 wt.% partially hydrolysed collagen & 20 wt.% OSA starch	80 wt.% concentrated collagen & 20 wt.% OSA starch	80 wt.% partially hydrolysed collagen & 20 wt.% Hydrolysed maize starch
<b>Powders</b> [kgh <sup>-1</sup> ]	100	100	95	95	95
<b>Water</b> [kgh <sup>-1</sup> ]	13	10	18	18	18
<b>Glycerol</b> [kgh <sup>-1</sup> ]	15	17	22	22	22
<b>Steam</b> [kgh <sup>-1</sup> ]	13	13	0	0	0
<b>EPP</b> [bar]	30	20	93	5	83
<b>B2</b> [°C]	110	110	125	108	110
<b>B3</b> [°C]	110	110	101	108	108
<b>B5</b> [°C]	90	90	94	87	87
<b>B6</b> [°C]	80	80	67	65	65
<b>B7</b> [°C]	70	70	60	58	58
<b>SME</b> [kWhkg <sup>-1</sup> ]	65	65	86	35	85
<b>RPM</b>	175	190	175	300	170
<b>Vac</b> [bar]	0.4	0.4	0.26	0.28	0.26

Hydrolysed sodium octenyl succinate starch is often more expensive than many native starches. A separate formulation was extruded consisting of 80 wt.% partially hydrolysed collagen and 20 wt.% of a cheaper unmodified hydrolysed maize starch to emphasise the cost benefit that the relative versatility of partially hydrolysed collagen brings. The tensile toughness was determined for these formulations. The texture results are shown in Table 9.

Table 9

<b>Formulation</b>	<b>Tensile Ductility [%]</b>	<b>Tensile Toughness [MPa.]</b>	<b>Tensile Strength [Mpa]</b>
Control (Commercially-Available Edible Animal Chew, CAP2)	37.95	102.06	3.41
80 wt.% Pork Skin Protein & 20 wt.% OSA starch	517.00	503.00	2.40
80 wt.% Partially hydrolysed collagen & 20 wt.% OSA starch	512.76	627.60	2.64
80 wt.% Concentrated porcine collagen extract & 20 wt.% OSA starch	424.76	592.91	2.29
<i>80% Partially hydrolysed collagen &amp; 20% Hydrolysed maize starch</i>	-	692.30	-
<b>Formulation</b>	<b>Shear Resistance [kg]</b>	<b>Conical Probe Hardness [kg]</b>	
Control (Commercially-Available Edible Animal Chew, CAP2)	10.67	3.73	
80 wt.% Pork Skin Protein & 20 wt.% OSA starch	20.16	1.87	
80 wt.% Partially hydrolysed collagen & 20 wt.% OSA starch	30.89	1.06	
80 wt.% Concentrated porcine collagen extract & 20 wt.% OSA starch	20.84	1.09	
<i>80 wt.% Partially hydrolysed collagen &amp; 20 wt.% Hydrolysed maize starch</i>	-	-	

The results shown in Table 9 show the following:

- 5 All of the collagen based formulations significantly outperform the Control (a commercially-available edible animal chew) formulation for toughness. This shows the benefits of using collagen in comparison to the materials that were used to make the commercially-available edible animal chew. Of the normalised powder formulations consisting of 80 wt.% of the respective collagen source and 20 wt.% hydrolysed
- 10 sodium octenyl succinate starch, the formulation containing partially hydrolysed

collagen delivers the toughest texture, followed by the formulations containing concentrated collagen extract and pork skin protein. The formulation containing 80 wt.% partially hydrolysed collagen and 20 wt.% unmodified hydrolysed maize starch (e.g. Glucidex 2) is the toughest of all the samples measured. When this information is considered alongside the compatibility with thermal expansion that partially hydrolysed collagen brings, the commercial viability of this formulation is clear.

Table 9 also shows that the most shear resistant (driving chew time) and softest (driving safety) sample in the study also came from use of partially hydrolysed collagen.

#### **Example 5 – Lasting time and Palatability Tests**

Tests to determine the length of time an edible animal chew lasts were performed using a panel of dogs. The objective of lasting time trials is to measure the amount of time required for a dog to consume a sample. These tests were performed using samples designed for daily and weekly feeding regimens.

For the “daily” feeding regimen, samples were extruded in a shape that mimics the shape of a commercially-available edible animal chew (denoted ‘CAP2’ in the figures; samples were extruded through an approximately x shaped die). These samples were then cut to appropriate weights for daily feeding (approximately 16–40 g, depending on the sizes of the dogs in the feeding panel). This approach allowed for comparison of the results with existing data on commercially-available edible animal chew products (e.g. CAP2).

For the “weekly” feeding regimen, samples were extruded in a shape that mimicked the shape of a commercially-available edible animal chew (denoted ‘CAP3’ in the figures; samples were extruded through an approximately x shaped die). The low hot strength of the material means that this shape collapses, resulting in a much flatter final product. These samples were then cut to appropriate weights for weekly feeding (approximately 70–160 g, depending on the sizes of the dogs in the feeding panel). This approach allowed for comparison of the results with existing data on larger format products that are currently commercially available.

Due to the disparity in product weights between the samples fed and the comparison products, it is preferable to normalise the median measured time by dividing it by the sample/product weight in order to give a lasting time per gram (measured in seconds per gram). This measure was found to be largely consistent between the daily and  
5 weekly regimen feeding trials, and allows comparison with existing products

The lasting time per gram in the daily regimen of pork skin protein based samples is considerably higher than that of existing products; approximately twice to three times that of the commercially-available products that were tested (see Figure 7A; different  
10 commercially available products, i.e. animal chews, are labelled 'CAP1', 'CAP2' and 'CAP3' in Figures 7A). Figure 7B shows the median lasting times (not per gram) recorded for weekly format pork-skin-protein-based samples according to the present disclosure (referred to as "Chewhide" in the figures) versus those recorded for different sizes of other commercially-available edible animal chews. The sizes of the dogs used  
15 in each trial are included in parentheses. The solid bars on the right represent the actual recorded data, and the dotted bars represent the expected lasting time of samples cut to the weights of the control, commercially-available products (interpolated from actual lasting time data). It is clear that a weekly regimen chew made from pork skin protein has the potential to provide a step change in lasting time versus existing  
20 products.

A palatability trial was also undertaken with the objective of assessing whether dogs have a preference for eating a pork skin protein based chew versus a standard product. A commercially-available chew (denoted CAP2 in the Figures) was used as an  
25 example of a standard product, as it is starch-based chew, containing palatants designed to make it appealing to a dog. The pork-skin-protein-based samples tested did not include any additional palatants. The results showed that dogs have a clear preference for the pork-skin-protein-based chews, with these chews being chosen over a the commercially-available product on approximately 90% of occasions.

30

**Example 6 – partially hydrolysed collagen vs. a mixture of natural collagen and gelatine (hydrolysed collagen)**

Edible animal chews were formed from a mixture of concentrated natural collagen  
35 extract and gelatine, and their tensile toughness was determined. The measured

tensile toughness was compared with that of edible animal chews formed from partially hydrolysed collagen. The tensile toughness was significantly higher for edible animal chews formed from partially hydrolysed collagen than for those formed by simply blending gelatine with concentrated natural collagen extract (see Figure 8).

- 5 Additionally, the alternate blends utilising a mixture of concentrated natural collagen extract and gelatine collapsed when thermally expanded.

#### Texture Test Procedures

10 The partially hydrolysed collagen may comprise a 30:70 to 70:30 (wt:wt) mixture of water-soluble component and non-water soluble component. In some examples, the partially hydrolysed collagen may comprise a 35:65 to 65:35 (wt:wt) mixture, in some examples, a 40:60 to 60:40 (wt:wt) mixture, in some examples, a 45:55 to 55:45 (wt:wt) mixture, in some examples, about a 50:50 (wt:wt) mixture of a water-soluble component and a non-water-soluble component.

15

This texture analysis methodology pulls products apart giving information on their ductility, tensile strength and toughness. This method is applied to flat sheet (ribbon) extrudates which can be formed into tensile bars.

20 The following equipment and apparatus are used (see Figure 9):

- (vi) Stable Micro Systems TA-HD.plus;
- (vii) Tensile rig (A/HDT) – Max. load 500 kg;
- (viii) Secateurs (only required for testing a commercially-available edible animal chew);
- 25 (ix) Tensile cutter (ISO 527-2; 75mm long, 10mm wide, 5mm in centre) – Zwick Roell;
- (x) 100 or 500kg load cell.

#### Sample preparation and testing:

- 30 (vi) Cut an extrudate into to a tensile bar by using an ISO 527-2 stamp (Zwick Roell; see Figure 9C).
- (vii) Place the tensile bar into the lower grips of a tensile rig (Figure 9D; so that the full shoulder of the test piece is exposed) and tighten enough that the product does not slip during testing, but not so tight that the product breaks.
- (viii) Position the upper grips of the tensile rig to align with the test piece (again, so  
35 that the full shoulder of the test piece is exposed). Tighten as described above.

- (ix) Minimise any tension or slack in the test piece.
- (x) The grips are moved apart by the texture analyser (TA) at 1 mm/s and the resistive force is recorded by the sensor through the breaking point of the test piece.

5

Data analysis:

Figure 9F shows a general plot of the force response curve from a texture analysis test plotted on axes of stress (MPa) versus strain (%). The dimensions of the central break point (width and depth) are accounted for in the plot. From a plot of this type, the following parameters can be determined:

10

Tensile Strength (MPa) is the peak resistive force from test piece;

Ductility (%) is the elongation to break, that is, the ability to deform without failure;

Toughness (MPa) is the area under the curve and provides a compound measure

15

taking into account both the tensile strength and the ductility.

It is believed that the toughness of an edible animal chew is strongly correlated with the lasting time per gram in feeding tests.

## 20 Hardness Tests by the conical probe method

This texture analysis method uses a 30° cone geometry and applies the "Stress Relaxation" procedure whereby the probe indents the material/product by 3 mm and the force/stress acting on the probe during a 30 second hold period is measured. This characterises the hardness of the material (but can be used to characterise other attributes not considered in this study).

25

The following equipment and apparatus are used

- (i) Stable Micro Systems TA-HD.plus
- (ii) Lower tensile rig
- (iii) 30° Stainless steel cone (P/30CSS) – Max Load 50 kg
- (iv) Probe adapter (AD/100) – Max load 50 kg
- (v) 100 kg load cell

30

35 Sample preparation, testing and data analysis:

- (i) Ribbon shaped extrudates were cut to a length of 40 mm then damped with 10 mm of the extrudate protruding upwards (this length is long enough that the texture data is not influenced by the clamp, but short enough that bending is not significant).
- 5 (ii) The probe enters the product at a speed of 1 mm per second to a distance of 3 mm then holds its position for 30 seconds before being withdrawn at 1 mm per second. The force response from the material is measured giving rise to a plot of the form depicted below. The material hardness (kg) can be determined by the peak force as shown.

10

A 6 mm probe was found to be an adequate measure to approximate hardness in the analysis of samples made from pork (or beef) skin protein. The 6 mm probe measure was found to be a poor approximation of the material hardness in partially hydrolysed collagen samples as it is a compound measure of shear resistance and hardness. The high shear resistance of the partially hydrolysed collagen samples skewed the results so the 6 mm probe method was discarded and the conical probe method used exclusively for these samples.

15

#### Shear Resistance – Warner Brazler Rig

20

This texture analysis method uses a “Λ” shaped blade (Warner Brazler Blade) and a slit die base plate to determine the shear resistance.

The following equipment and apparatus are used for analysis on 10 mm wide by 5 mm tall strip or ribbon shaped sample of unlimited length:

25

- (i) Stable Micro Systems TA-HD.plus ;
- (ii) Heavy Duty Platform (HDP/90) – Max Load 300 kg;
- (iii) Warner Brazler Blade – Max Load 25 kg;
- (iv) Probe adapter (AD/100) – Max load 50 kg (bespoke probes can be made that negate the use of the Probe Adaptor and allow the test to be rated to 250 kg);
- 30 (v) Base plate with 3 mm slit;
- (vi) 100kg load cell.

Sample preparation, testing and data analysis:

The ribbon shaped sample was placed in the apparatus and the Warner Brazler Blade is used to cut through the sample. The peak force [kg] required to shear the sample provides a measure of the shear resistance.

## 5 Feeding trial descriptions

### *Lasting time*

The dog is given a chew, and a stopwatch is started. The timing continues until the dog has finished eating the chew, with any pauses of more than 5 seconds recorded. The  
 10 pauses are then summed and subtracted from the time on the stopwatch to give an “eating time”. Each dog on the panel (approximately 10 dogs) is fed three times, with the spacing determined by the feeding guide of the product (daily or weekly).

### *Palatability*

15 Each dog on the panel (approximately 30 dogs) is fed a sample of each product to be compared. The products are then cut into small pieces and placed within mesh topped boxes, allowing the product to be smelt but not seen. The dog is then allowed to smell  
 20 each box before choosing which it prefers; the dog is then allowed to eat the chosen sample. This is repeated eight times over two days, with the order of the boxes rotated to ensure a fair test.

### Example 7 – further formulation

As a further formulation for the edible animal chew, the following ingredients were  
 25 combined as shown in Table 10 below.

Table 10

Ingredient	Parts (by weight)
Maize starch	45-55
Partially hydrolysed collagen	25-35
OSA modified starch	5 - 15
Pork skin protein	5 - 15
Other additives (including salt, flavourings, vitamins, minerals, emulsifier, flour)	1-10

This formulation was found to be particularly effective as an edible animal chew.

**CLAIMS**

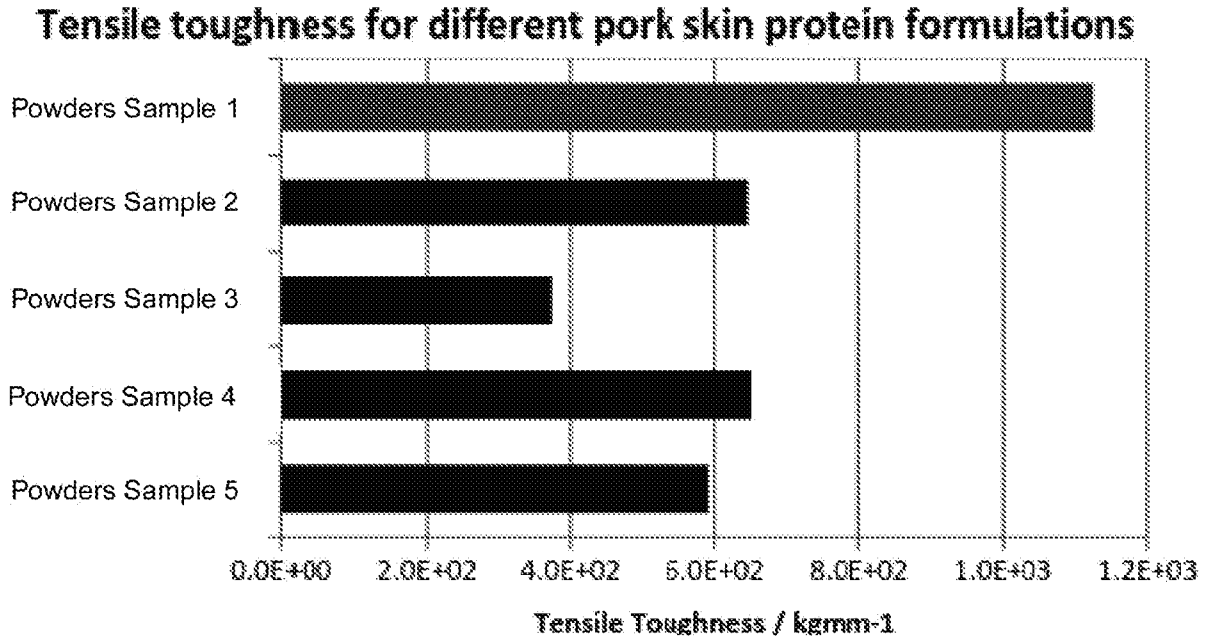
1. An edible animal chew comprising:  
a partially hydrolysed collagen; and/or  
5 an alkyl succinate modified starch, wherein the edible animal chew has a tensile toughness greater than 200 MPa.
2. The edible animal chew according to claim 1, wherein the alkyl succinate is a C4 to C12 alkyl succinate.
3. The edible animal chew according to claim 1 or claim 2, wherein the succinate is  
10 a metal succinate.
4. The edible animal chew according to any one of the preceding claims, wherein the alkyl succinate modified starch is selected from sodium octenyl succinate starch, calcium octenyl succinate starch, potassium octenyl succinate starch, aluminium octenyl succinate starch and any combination thereof.
- 15 5. The edible animal chew according to any one of the preceding claims, wherein the edible animal chew comprises a partially hydrolysed collagen and wherein the partially hydrolysed collagen has a Bloom gel strength of at least 300, as measured in accordance with ISO 9665.
6. The edible animal chew according to any one of the preceding claims, wherein  
20 the partially hydrolysed collagen has a molecular weight of at least 60kDa, optionally at least 70 kDa, as measured using gel permeation chromatography (GPC).
7. The edible animal chew according to any one of the preceding claims, wherein the partially hydrolysed collagen, before incorporation into the edible animal  
25 chew, is formable by a process comprising forming a wet collagen product in the form of a fibrous mass from a hide or skin at a temperature of 50 °C or less, followed by drying the wet collagen product using a contact dryer, having a surface temperature of 150°C or higher, to obtain the partially hydrolysed collagen in the form of a powder.
- 30 8. The edible animal chew according to any one of the preceding claims, wherein the partially hydrolysed collagen comprises a 30:70 to 70:30 mixture of a water soluble component and a non-water soluble component.
9. The edible animal chew according to any one of the preceding claims, further comprising a high purity natural collagen, in that the high purity natural collagen  
35 comprises less than 8 wt% fat.

10. The edible animal chew according to any one of the preceding claims, wherein the edible animal chew comprises 8 wt.% to 60 wt.% partially hydrolysed collagen.
11. The edible animal chew according to any one of the preceding claims, wherein  
5 the edible animal chew comprises 15 wt.% to 45 wt.% partially hydrolysed collagen.
12. The edible animal chew according to any one of the preceding claims, wherein the edible animal chew comprises 20 wt.% to 40 wt.% partially hydrolysed collagen.
- 10 13. The edible animal chew according to any one of the preceding claims, wherein the edible animal chew comprises 1 to 25 wt.% alkyl succinate modified starch.
14. The edible animal chew according to any one of the preceding claims, wherein the edible animal chew comprises from 1 wt.% to 20 wt% alkyl succinate modified starch.
- 15 15. The edible animal chew according to any one of the preceding claims, wherein the edible animal chew further comprises unmodified starch.
16. The edible animal chew according to claim 15, wherein the edible animal chew comprises 50 wt.% or less unmodified starch.
17. The edible animal chew according to any one of claims 15 or 16, wherein the  
20 edible animal chew comprises 10 wt.% or more unmodified starch.
18. The edible animal chew according to any one of claims 15 to 17, wherein the edible animal chew comprises 15 wt.% or more unmodified starch.
19. The edible animal chew according to any one of claims 15 to 18, wherein the unmodified starch comprises maize starch.
- 25 20. The edible animal chew according to any preceding claim, wherein the edible animal chew comprises a plasticiser.
21. The edible animal chew according to claim 20, wherein the plasticiser is selected from glycerol and water.
22. The edible animal chew according to any one of claims 20 to 21, wherein the  
30 edible animal chew comprises 15 wt.% to 45 wt.% plasticiser.
23. The edible animal chew according to any one of the preceding claims, wherein the edible animal chew comprises less than 5 wt% fat.
24. The edible animal chew according to any preceding claim, wherein the edible animal chew is an elongate edible animal chew.

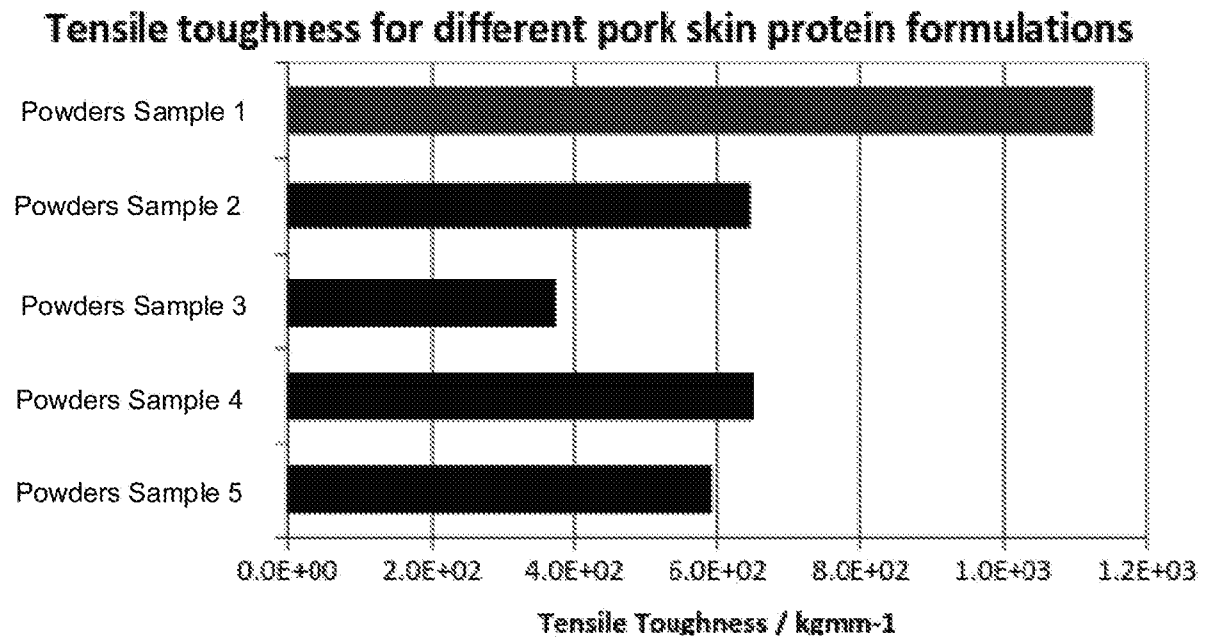
25. The edible animal chew according to claim 24, wherein the chew is elongate along an axis and at least one channel runs through the chew along the axis.
26. The edible animal chew according to any preceding claim, wherein the edible animal chew has been formed by extrusion.
- 5 27. The edible animal chew according to any preceding claim, wherein the edible animal chew has an internal cellular structure.
28. The edible animal chew according to claim 27, wherein the edible animal chew is a thermally expanded edible animal chew.
29. The edible animal chew according to any preceding claim, wherein the edible  
10 animal chew is aerated.
30. The edible animal chew according to any preceding claim wherein the alkyl succinate modified starch is sodium octenyl succinate starch.
31. The edible animal chew according to any preceding claim, wherein the tensile toughness of the edible animal chew is at least 300 MPa.
- 15 32. The edible animal chew according to any preceding claim, wherein the edible animal chew comprises components ('additives') other than those mentioned in the preceding claims, wherein the additives comprise 10 wt.% or less of the edible animal chew.
- 20 33. A method of making an edible animal chew, the method comprising:
- a. providing an edible chew composition comprising:  
partially hydrolysed collagen; and/or  
alkyl succinate modified starch;
- b. heating the edible chew composition to form a flowable edible chew composition;
- 25 c. extruding the flowable edible chew composition to form an extrudate;
- d. allowing the extrudate to cool and harden to form the edible animal chew.
34. The method according to claim 33, wherein the flowable edible chew composition leaves the extruder at a temperature above the melting point of the partially hydrolysed collagen.
- 30 35. The method according to claim 33, wherein the temperature is 100°C or more, such that steam causes expansion of the flowable edible animal chew mixture and the formation of an internal cellular structure in the resultant edible animal chew.
- 35 36. The method according to any one of claims 33 to 34, wherein the temperature is 150°C or less.

37. The method according to any one of claims 33 to 36, wherein the edible chew composition is formed in a barrel extruder.
38. The method according to claim 37, wherein the temperature of the barrel in the barrel extruder increases in the extrusion direction.
- 5 39. The method according to any one of claims 33 to 38, wherein the extruder is a single screw extruder or a twin screw extruder.
40. The method according to claim 38, wherein the screw rotates at 80 rpm to 300 rpm.
41. The method according to any one of claims 33 to 40, wherein the specific  
10 mechanical energy (SME) applied by the extruder is 60 kWhkg<sup>-1</sup> to 120 kWhkg<sup>-1</sup>.
42. The method according to any one of claims 33 to 41, wherein the extrusion is supercritical fluid extrusion.
43. The method according to claim 42, wherein the supercritical fluid is nitrogen or carbon dioxide.
- 15 44. An edible animal chew according to any one of claims 1 to 32, wherein the edible animal chew is produced by a method according to any one of claims 33 to 43.
45. An edible animal chew, comprising a blend of  
(i) a natural collagen extract, optionally having a fat content of 8wt% or less  
20 and/or a protein content of at least 800 g per kg of the extract; and  
(ii) a partially hydrolysed collagen.
46. An edible animal chew according to claim 45, wherein (i) and (ii) are present in wt:wt ratio of 20:80 to 80:20.
- 25 47. An edible animal chew according to claim 45 or claim 46, wherein the animal chew substantially lacks a modified starch.
48. An edible animal chew according to any of claims 45 to 47, wherein components  
30 (i) and (ii) constitute at least 30wt% of the composition, optionally at least 50wt% of the composition, optionally at least 80wt% of the composition, and optionally at least some of the remaining wt% of the edible animal chew comprising a component selected from an unmodified starch, plasticiser and an alkyl succinate modified starch.
- 35

49. An edible animal chew according to claim 48, wherein the unmodified starch comprises a maize starch.
50. The edible animal chew according to any of claims 1 to 32 or claims 45 to 49,  
5 wherein the edible animal chew further comprises diacetyl tartaric acid esters of mono- and diglycerides (DATEM).
51. The edible animal chew according to any of claims 1 to 32 or claims 45 to 49, further comprising a humectant.



*FIG. 1(A)*



*FIG. 1(B)*

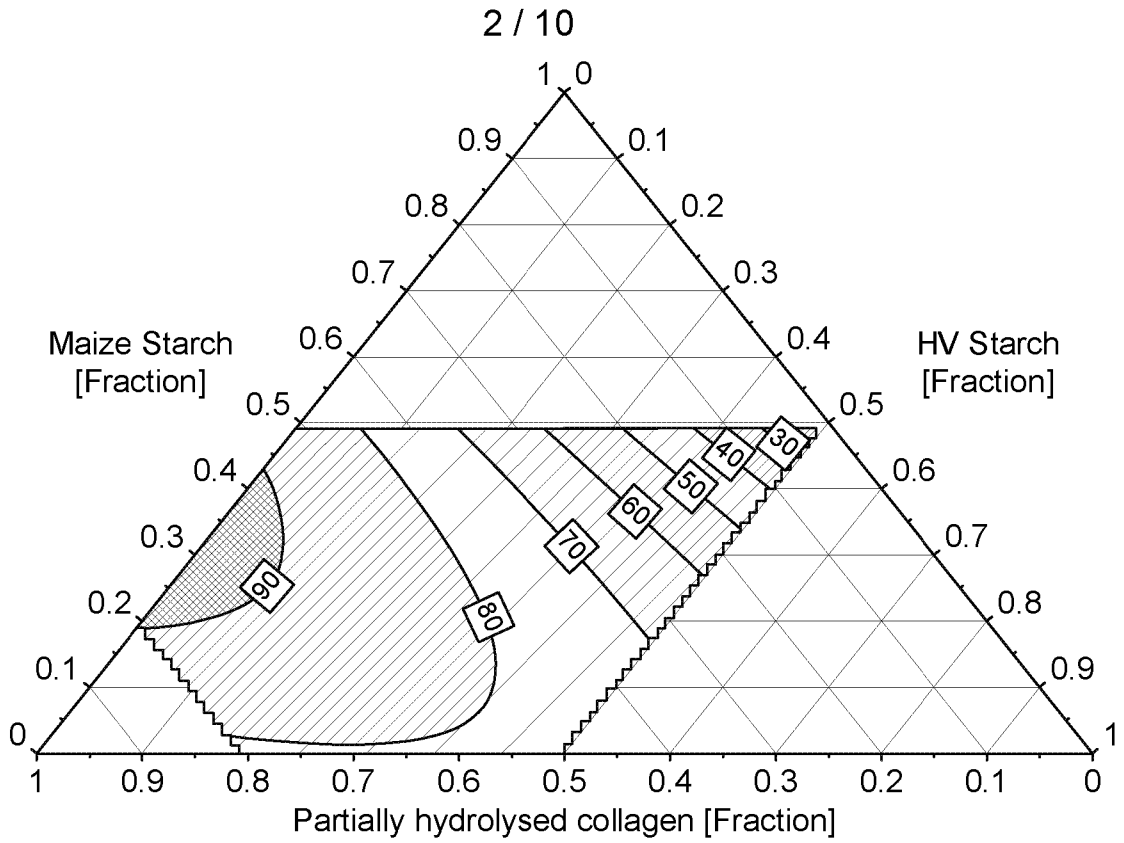


FIG. 2(A)

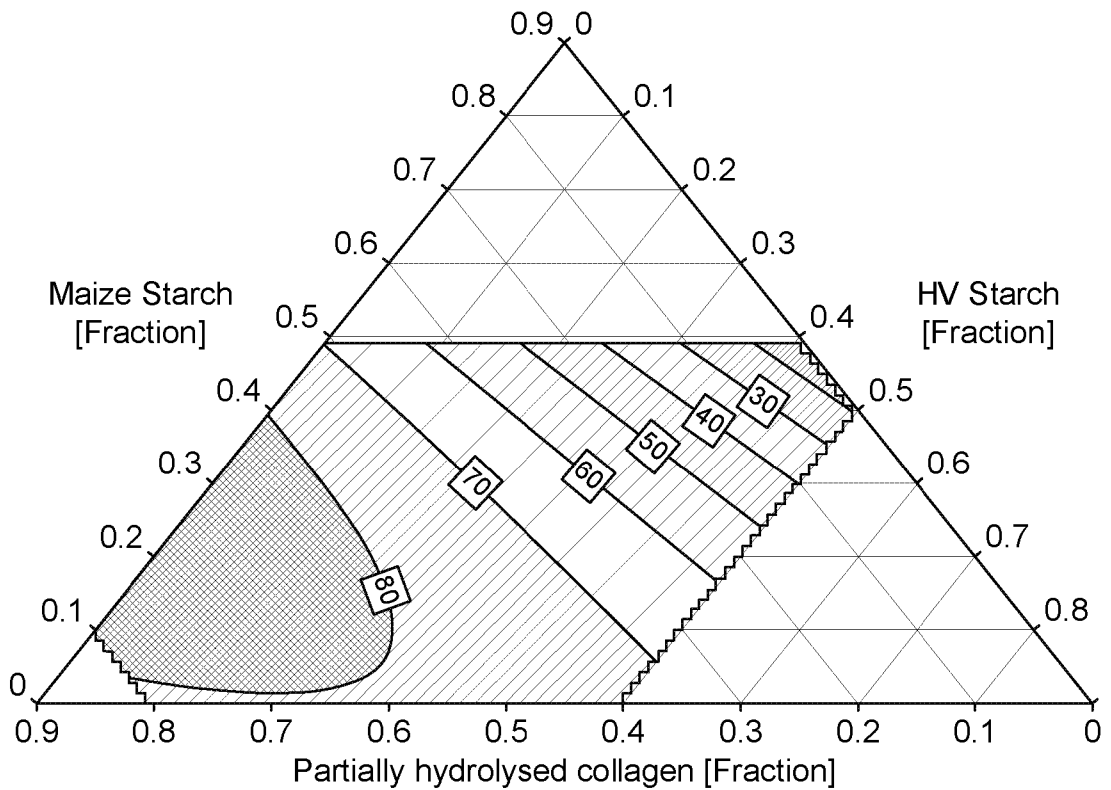


FIG. 2(B)

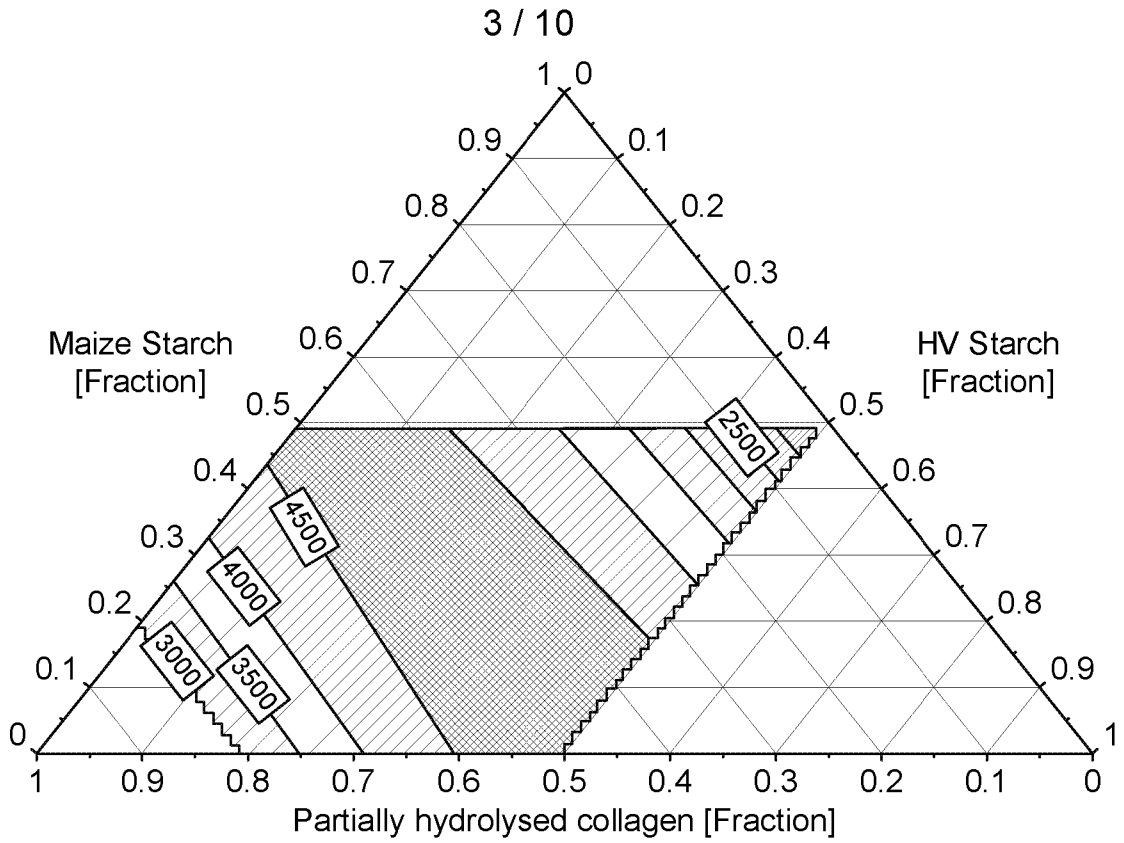


FIG. 3(A)

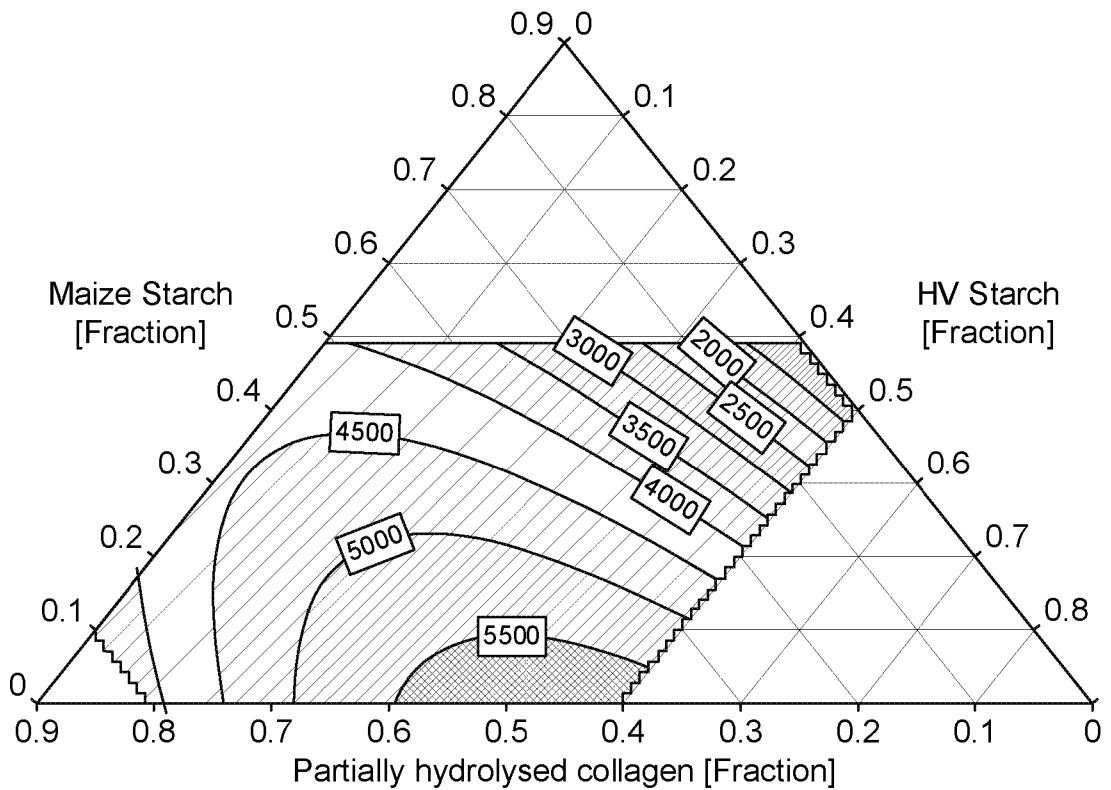


FIG. 3(B)

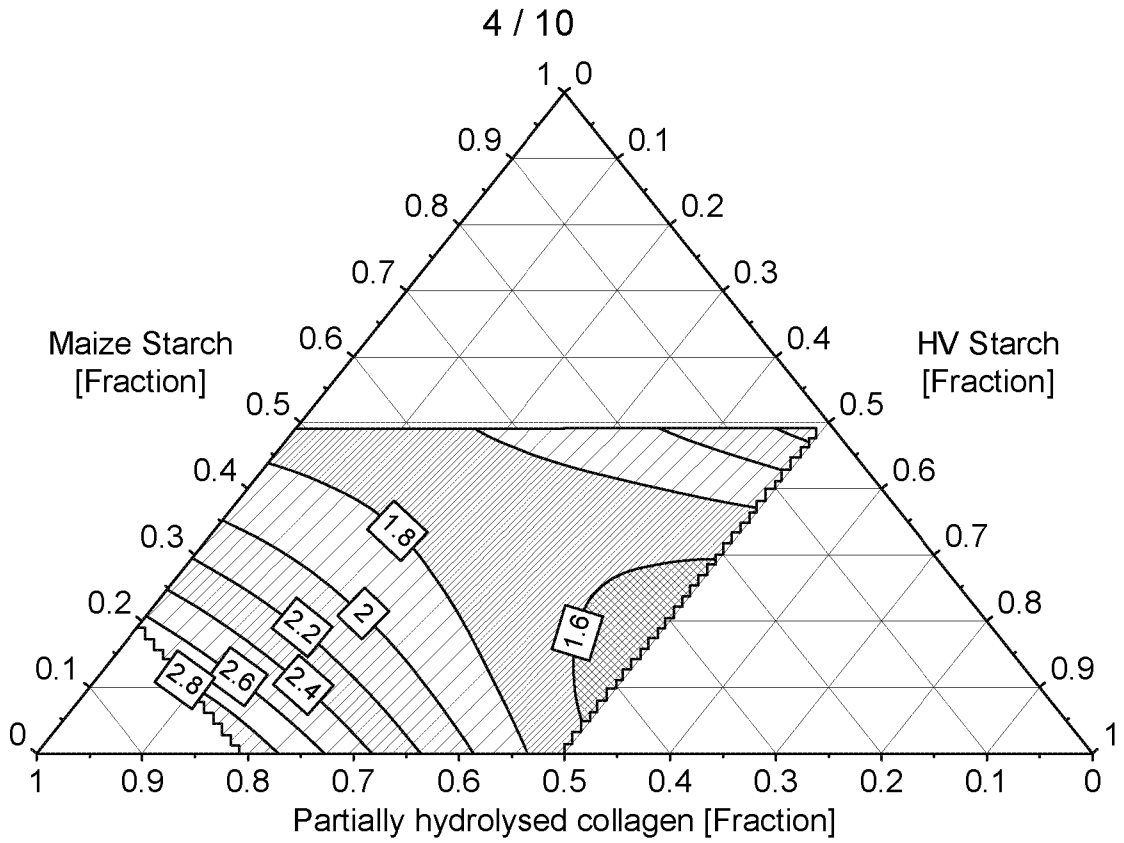


FIG. 4(A)

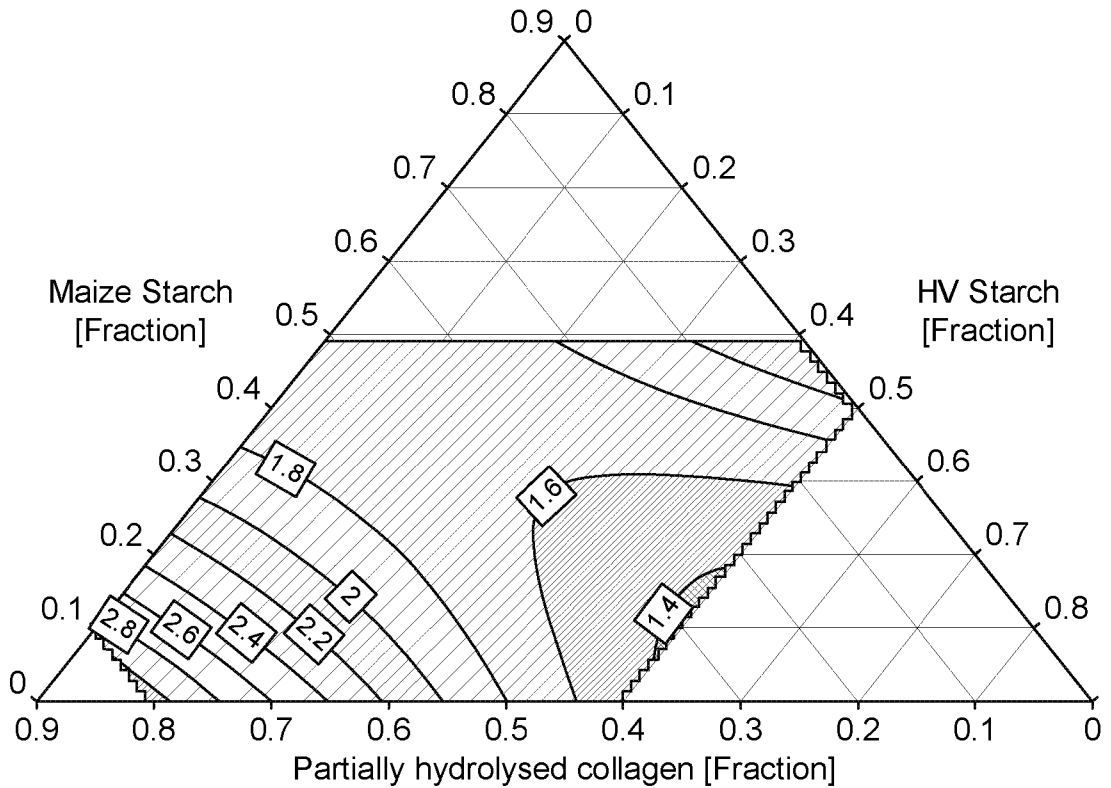


FIG. 4(B)

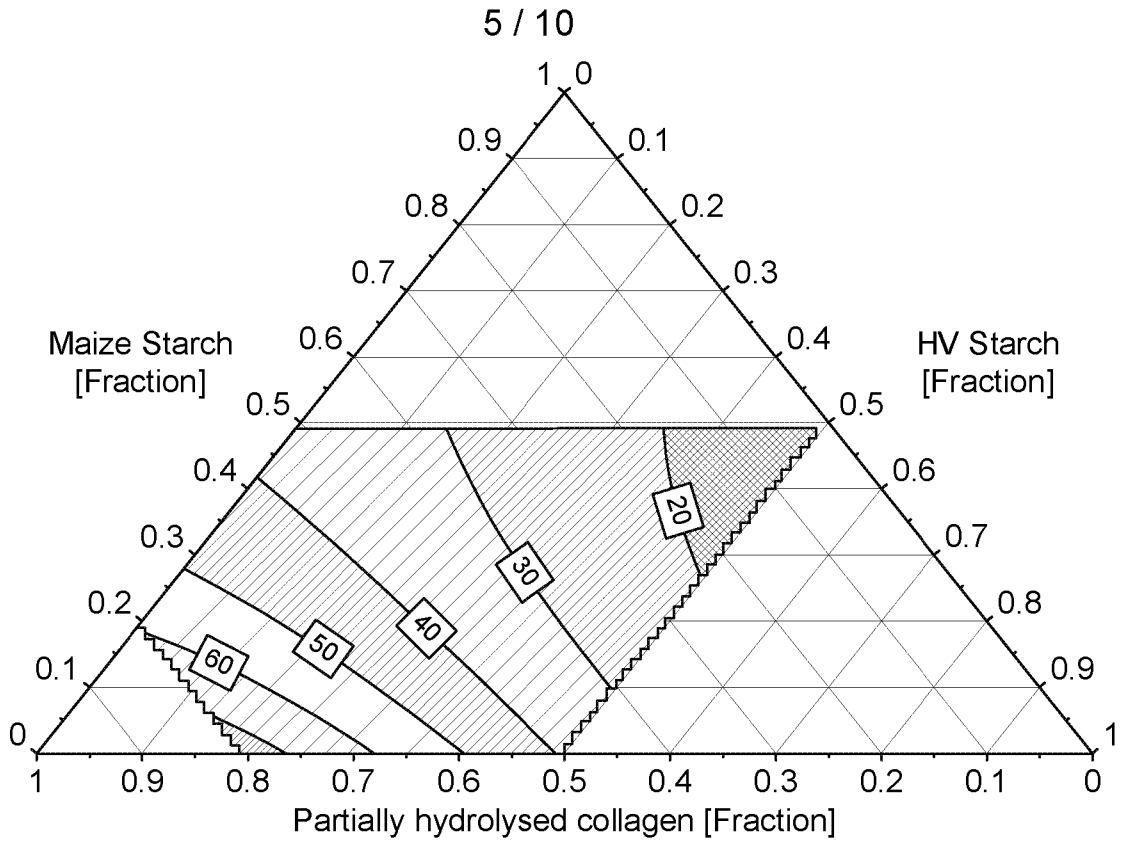


FIG. 5(A)

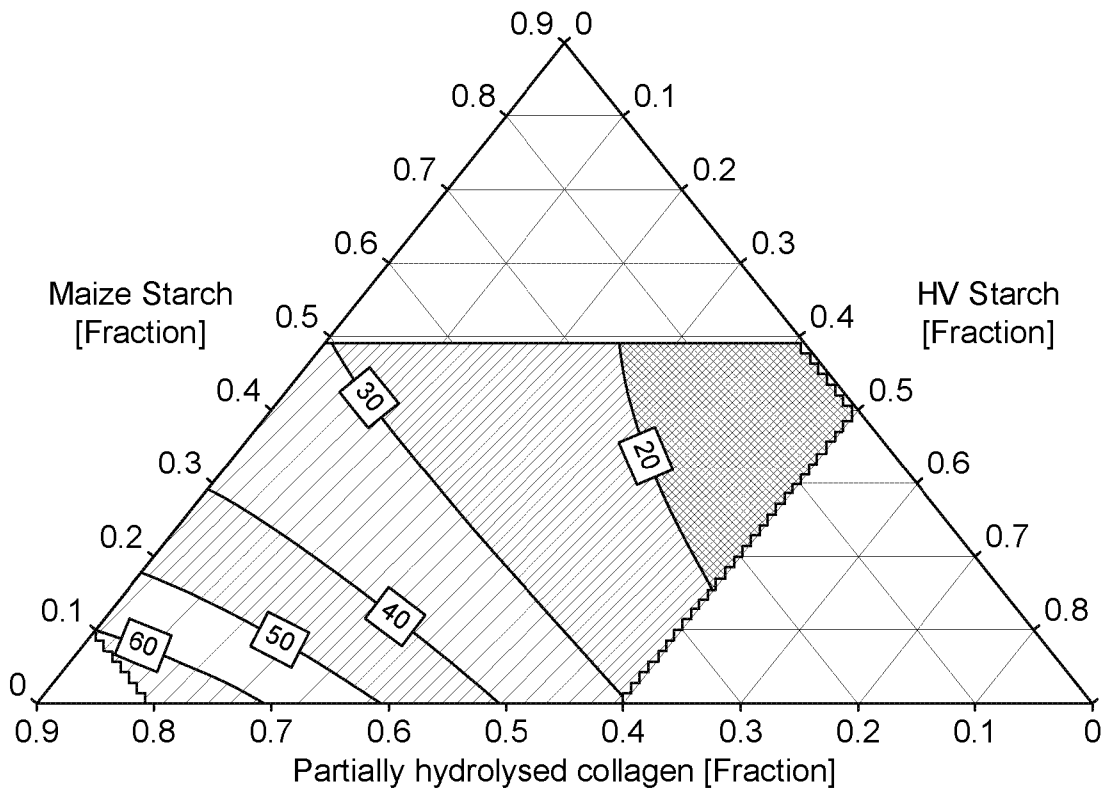


FIG. 5(B)

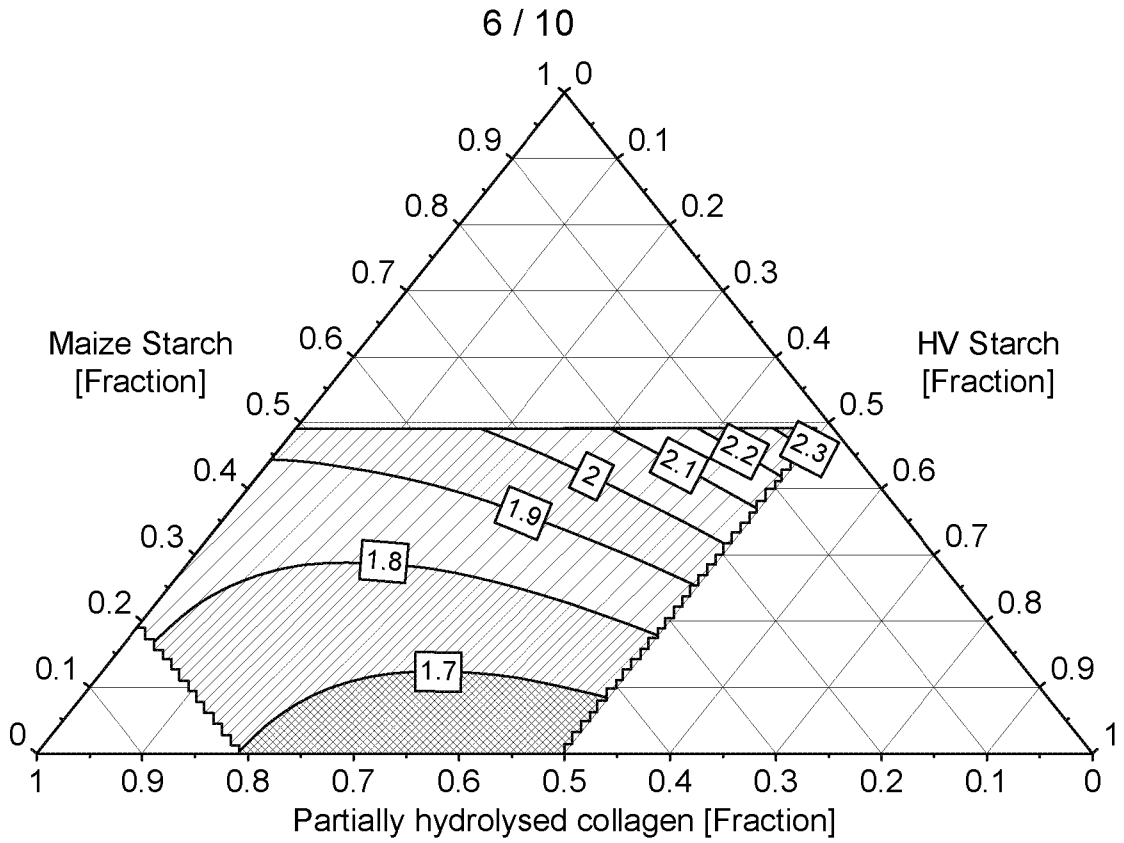


FIG. 6(A)

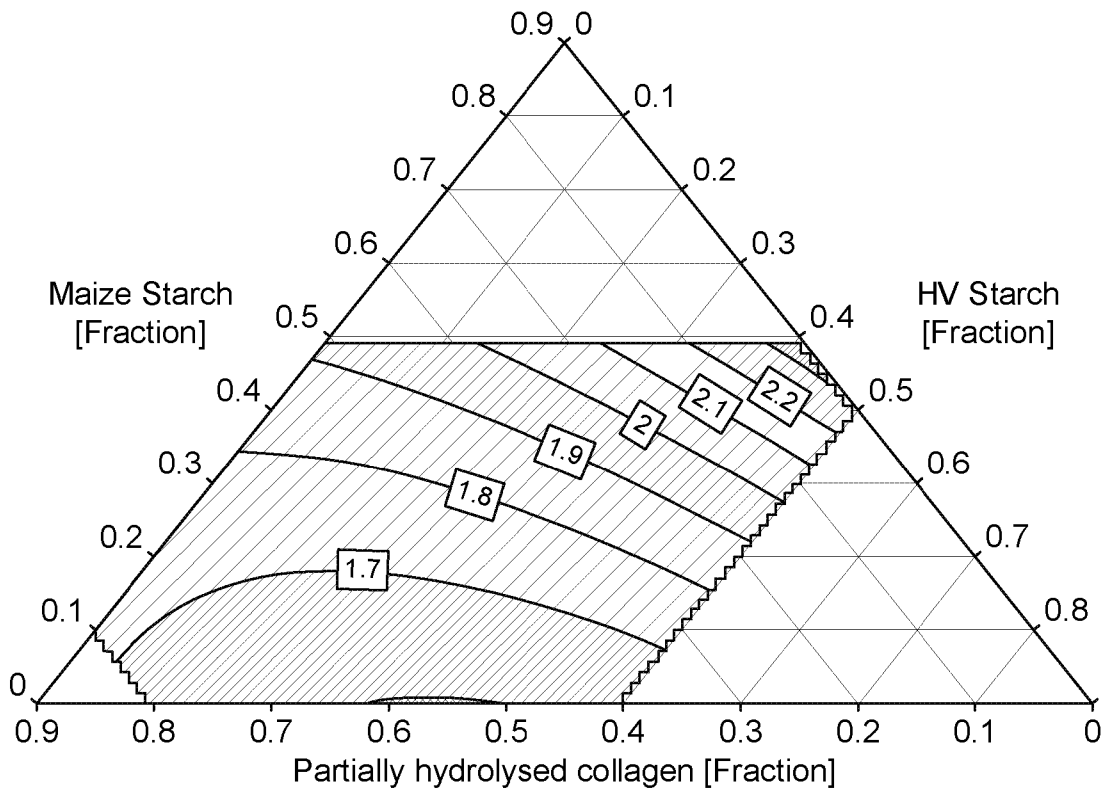


FIG. 6(B)

7 / 10

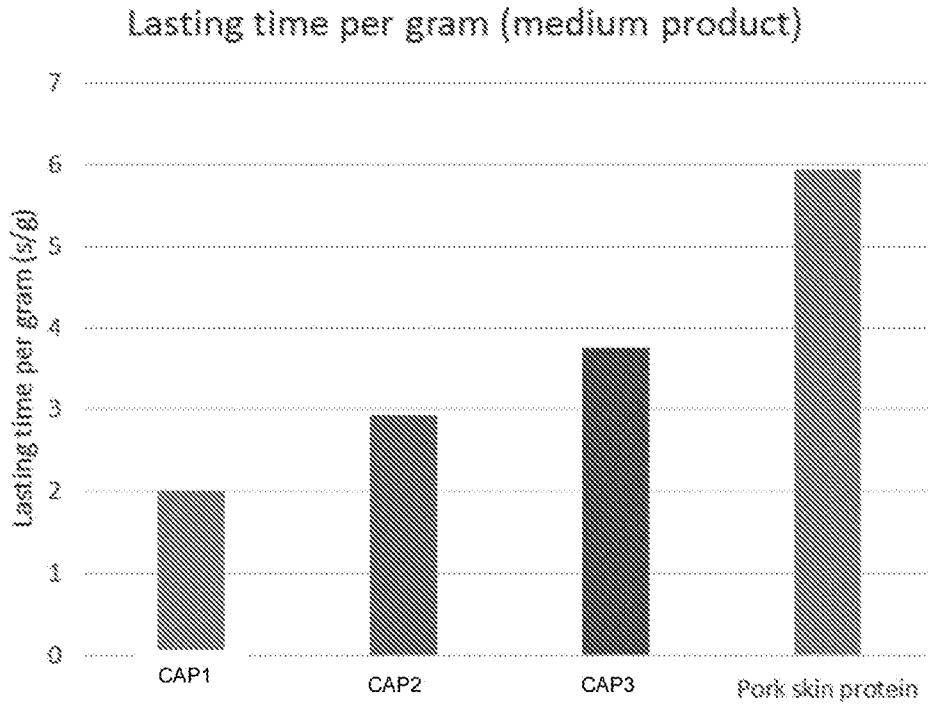


FIG. 7(A)

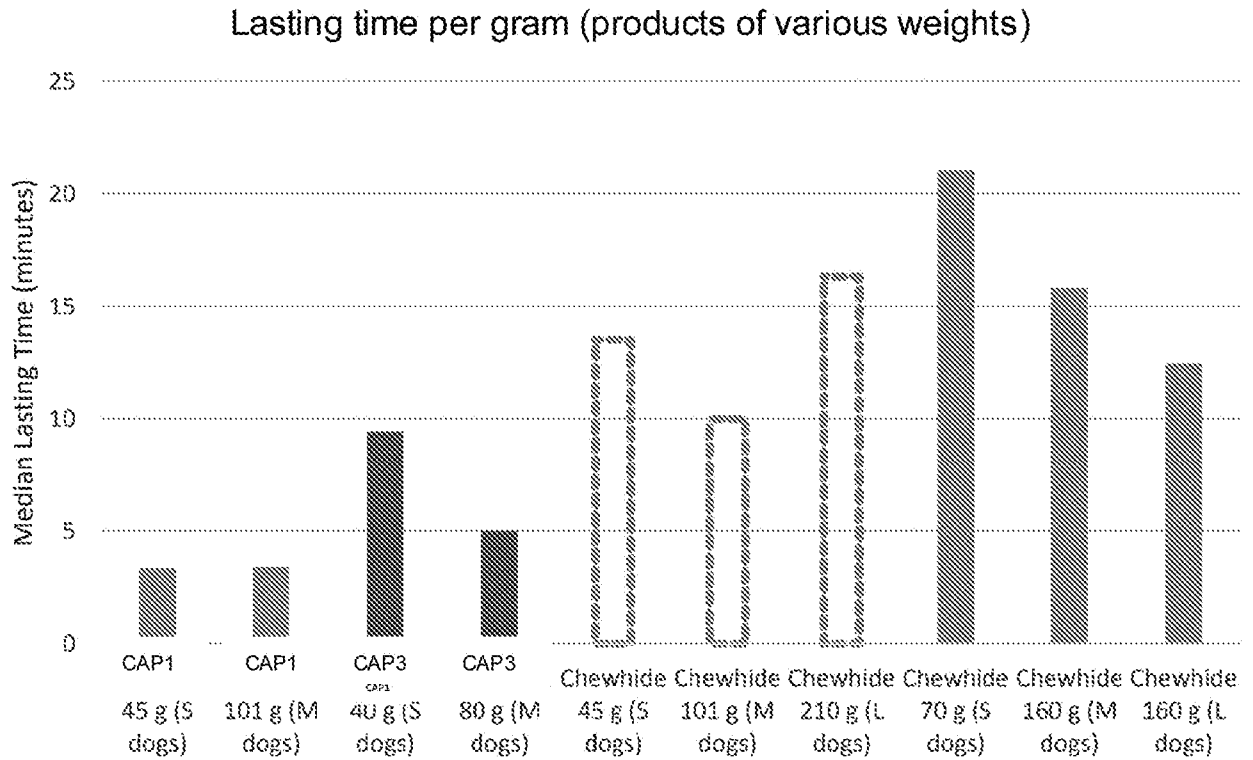
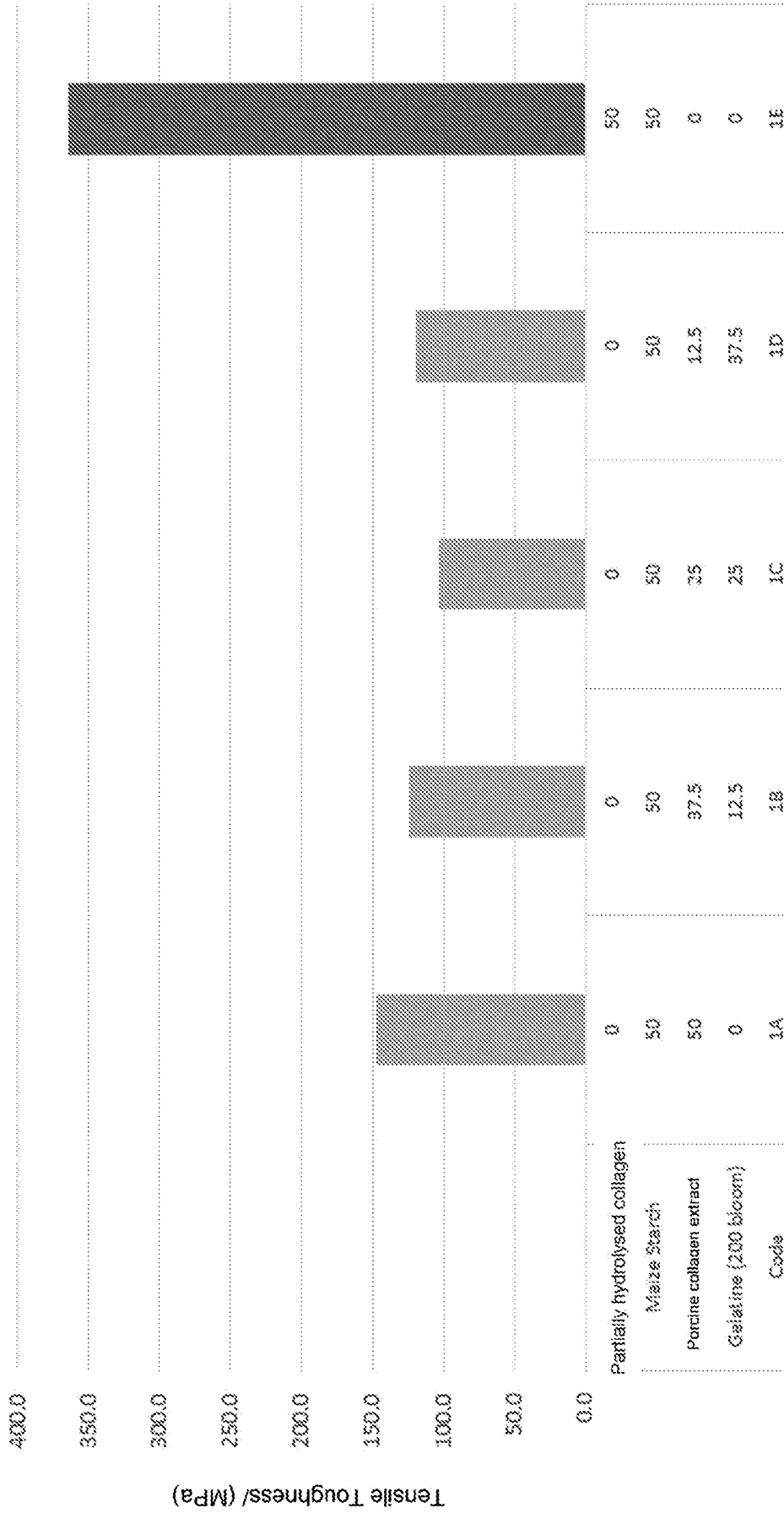
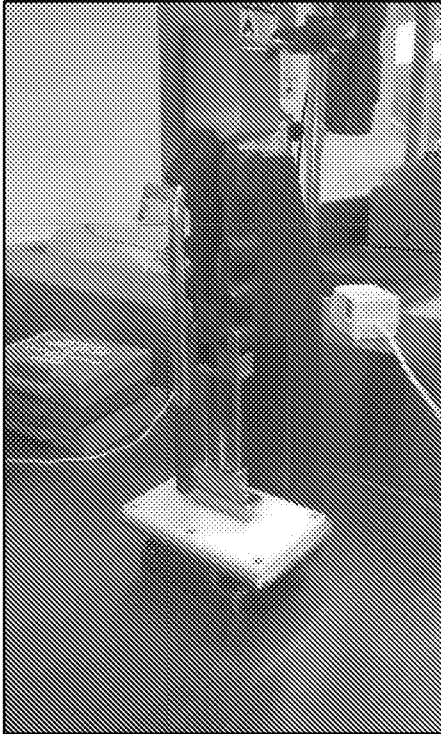


FIG. 7(B)

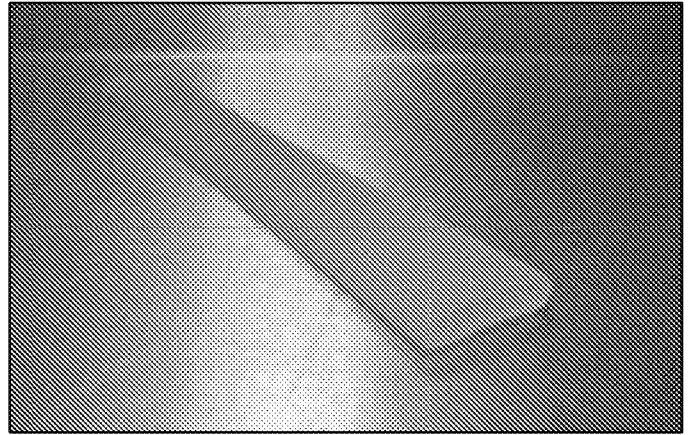


Blend Code and % Inclusion Powder Blend

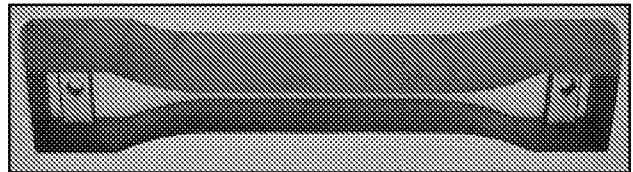
FIG. 8



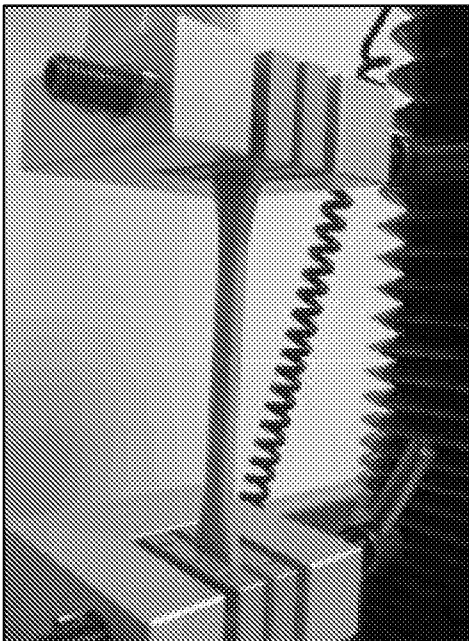
*FIG. 9(A)*



*FIG. 9(B)*



*FIG. 9(C)*



*FIG. 9(D)*



*FIG. 9(E)*

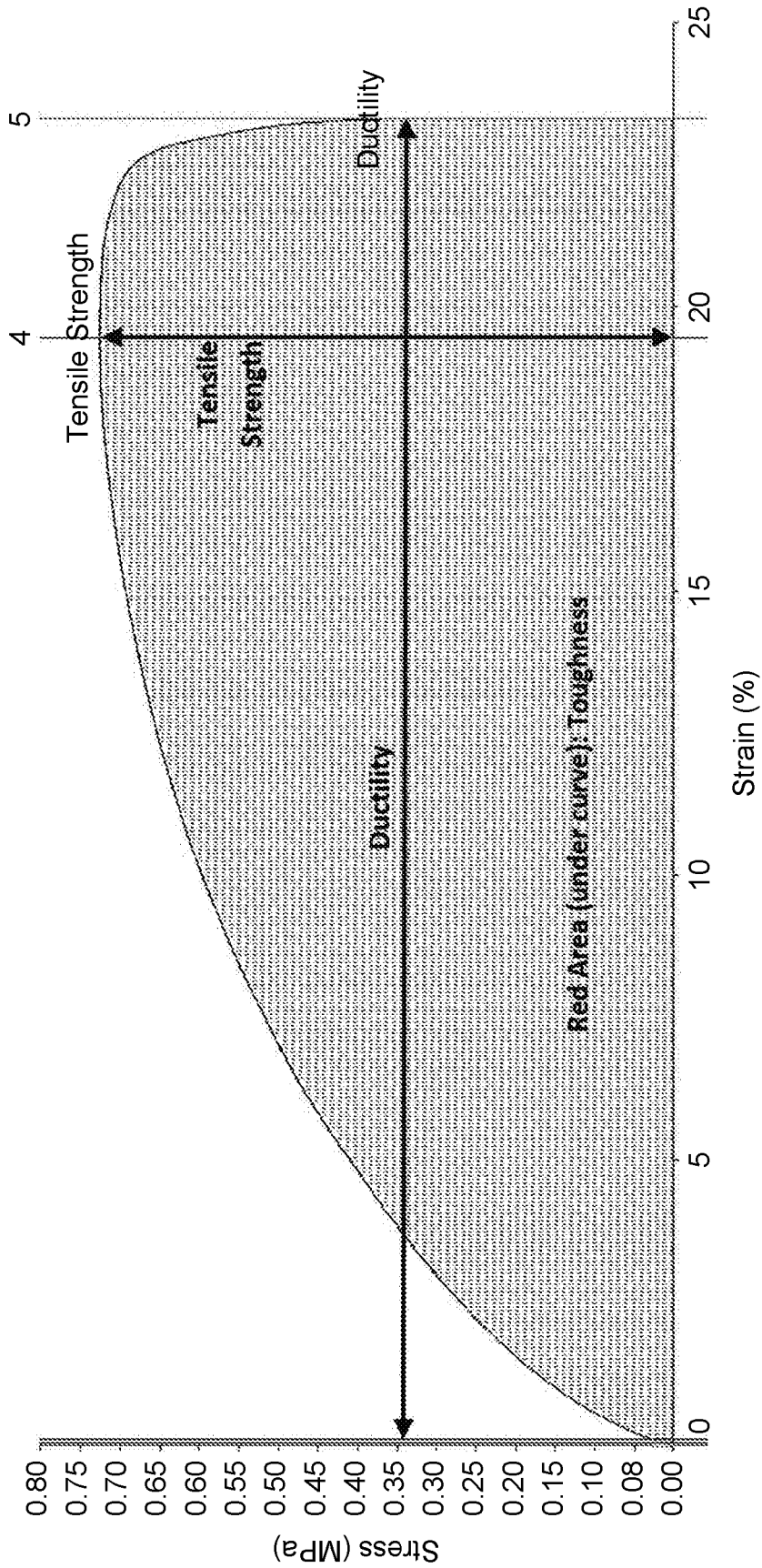


FIG. 9(F)