TREATMENT OR PREVENTION OF METABOLIC BONE DISORDER

The invention provides a method of preventing or treating metabolic bone disorder, comprising the step of administering a composition comprising phosphopeptide salt and phytoestrogen. Also provided is a composition for the treatment or prevention of a metabolic bone disorder, the composition comprising phosphopeptide salt and phytoestrogen.
TREATMENT

The invention relates to the treatment or prevention of metabolic bone disorder by administration of a composition comprising phosphopeptides, particularly as a salt, and phytoestrogen. The invention also relates to compositions for use in such a method, to the extraction of phytoestrogen, and in particular, the invention relates particularly to a nutriceutical composition for enhancing calcium absorption.

Background

There are a number of medical indications resulting from inadequate calcium, whether due to excessive secretion, inadequate absorption in the GIT or other causes. One of the most well known of these is metabolic bone diseases, such as osteoporosis. Osteoporosis is a common metabolic bone disease (MBD) affecting many individuals, particularly a significant number of post-menopausal women. As much as 15-50% of bone mass may be lost in these subjects during the first 10 years of menopause.

A number of factors are known to affect bone health including genetics, exercise and diet. Some dietary factors can compromise bone health by increasing the rate at which calcium is released from the skeleton and excreted from the body. Excess calcium resorption into the blood over deposition into the bone can result in thinning of the bone matrix.

Calcium supplements in the form of tablets, or increased dietary intake of calcium - rich foods are encouraged as a preventative measure of MBD including osteoporosis. However, absorption of the calcium is dependent on its form (some salts are more soluble and some are more easily absorbed), and the presence of substances eg oxalates in spinach, rhubarb, beet greens or carrots (which bind to calcium, making it unusable). The presence of acids or high amounts of fats can also impede calcium absorption. Due to inefficient absorption of the calcium, the effectiveness of treatment or prevention of osteoporosis simply by increased calcium intake is therefore limited.
Some studies indicate that urinary calcium loss, rather than calcium intake, is the predominant factor contributing to variations in calcium balance among women. Thus, the amount of calcium retained by the bones appears to be at least as important as the actual amount consumed, even though adequate calcium intake throughout life is essential for good bone health. It is thought that some dietary factors can compromise bone strength by increasing the rate of release of calcium from the bone, which is then excreted from the body. One dietary factor shown to increase urinary calcium loss is animal protein. Some studies show that, relative to animal meat protein, soy (plant) protein causes less urinary calcium to be excreted. Thus, plant proteins rather than animal proteins are thought to be more beneficial for health.

Phytoestrogens are plant derived chemicals which possess oestrogenic activity. Isoflavones is a general name given to derivatives of 3-phenyl-4H-1-benzopyran-4-one. Of particular interest are isoflavone phytoestrogens which occur in the *Leguminosae* plant family. They bind to animal (including human) estrogen receptors, although generally less strongly (and thus have a weaker effect) than estrogen. Particular phytoestrogens with known effect are genestean, diadzein and biochanin A, which have similar known structures. Formononetin and glycitein are isoflavones less effective as estrogen mimics.

Phytoestrogen isoflavones can be isolated from members of the *Leguminosae* plant family. These include but are not limited to lentils, chick peas, alfalfa, soya beans, soy plant parts such as soya hypocotyls and clover or clover plant products such as clover leaves and various varieties of beans. Red clover (*Trifolium pratense*) and subterranean clover (*Trifolium subterranean*) are particularly suitable sources.

Phytoestrogens frequently exist naturally with a bound sugar moiety in:

(i) the glucone (or glycosidic) form (eg. bound to glucose with a β-glucosidic linkage),

(ii) the glucone form and a bound malonyl moiety, or
(iii) the glucone form and a bound acetyl moiety.

As the aglucone or aglycone forms are biologically active, these are the desirable end-products. One way to cleave the sugar moiety is use of the $\beta$-glucosidase enzyme, although this would be expensive in large scale production. One advantage of using red clover as an isoflavone source is its natural content of $\beta$-glucosidase to avoid the need for it to be added. Alternatively, hydrochloric acid has been used to hydrolyse the glucone isoflavones. It is also known that the aglucone forms are not water-soluble and can be extracted with organic solvents (eg. ethyl acetate, hexane) but the glycosidic forms are water soluble.

Another source of phytoestrogens is to extract them from plant material, particularly the leaves and stems of Leguminosae plants. Extraction of isoflavones from plants has been attempted in many ways. One way is a process described in Japanese Patent 7-173148 which primarily relates to the separation of genistein from an isoflavone mixture using carbon di-, tri- or tetrachloride. The method of obtaining the isoflavone mixture is described as mixing soya bean with water and organic solvent. The isoflavones are soluble in water and can be recovered by an ion-exchange resin. The isoflavones are then freed from lipophilic impurities by extraction with an organic solvent, and then hydrolysed to form a mixture of the aglycone form of the isoflavones. The phytoestrogen products sought to be extracted were the estrogenic isoflavones, genistein and diadzein, being the two main isoflavones in soya.

International patent application no. WO 98/49153 (by Novogen Inc) describes a number of the difficulties of phytoestrogen extraction and describes an extraction method which allows an enzyme such as a $\beta$-glucosidase to react with the source material in order to convert the glycosides of the isoflavones into their aglycone forms, which are then extracted into a water-immiscible solvent eg, ethyl acetate, from which they are then recovered. Enzyme treatment is necessary when the source material is soya grits, but when it is red clover, direct extraction is possible as the aglycones are the main forms present. Example 3 of the patent uses 50% ethanol for extraction, followed by another extraction with
ethyl acetate. This procedure is similar to the old method of Wong (1962)\(^1\) who also used red clover. The earlier method of Walter (1941)\(^3\) is a two-step procedure involving methanol extraction of soya bean flakes followed by acidic hydrolysis.

Soya beans are also a known source of phytoestrogen. It has been speculated that soya foods or soya products may play a beneficial role in the treatment of diseases such as cancer, including breast cancer, menopause and/or post-menopausal symptoms, heart disease and related conditions, and osteoporosis.

Some calcium supplements are known. One of the disadvantages of known calcium treatments is poor patient adherence to the regimens prescribed. This may be for a number of reasons, including a philosophical reluctance to take tablets of any sort and/or a dislike for the particular calcium-rich foods which provide calcium in a form readily absorbable in the GIT. There is a need for a composition which can provide increased calcium intake while also achieving a high level of consumer acceptance. Soy milk has been identified as such a product with a sufficiently high calcium content and high consumer acceptance. However, the amount of calcium absorbed from such soy milk is often not sufficient to satisfy the desirable intake, particularly if it is needed to overcome calcium deficiency disorders. Further, often a significant volume of such product must be consumed and consumers may deliberately or inadvertently consume an inadequate amount.

Accordingly, investigations have been carried out to develop a product which is substantially comprised of a primary component well accepted by consumers as healthy (and desirably “natural”), with sufficiently concentrated calcium that a relatively small volume (less than 200 mls, preferably around 100 mls, to ensure a daily dose is fully consumed) will provide an adequate calcium supplement, where the product also facilitates absorption of calcium. Additionally, investigations have been carried out as to more cost-efficient means of using phytoestrogens from plants in a safe and biologically effective manner with a view to utilising presumed or known therapeutic benefits from these substances.
Known methods require the extraction of phytoestrogens from plant material, which result in significantly increased costs (e.g., by substantial use of enzymes, which are expensive), and also significant capital costs and running costs with large-scale extraction processes involving a number of steps, particularly where dangerous or hazardous chemicals are used in the process. Such chemicals require careful handling and/or management and may have harmful residues in the end product. Further, in the case of treatment of osteoporosis, combination with calcium in particular forms has been investigated.

**Summary of the Invention**

In one aspect, the invention relates to a method of preventing or treating metabolic bone disorder, comprising the step of administering a composition comprising phosphopeptide salt and phytoestrogen.

In a preferred embodiment, the phosphopeptide is from a milk protein. More preferably, the phosphopeptide is a casein phosphopeptide salt of a divalent cation. Most preferably, the salt is a casein calcium phosphopeptide.

The phytoestrogen is preferably selected from the group consisting of diadzein, genistein, biochanin A, formononetin and glycitein. More preferably, genistein and biochanin A are used. Desirably, the phytoestrogen is in the aglucone form, although normally the sugar moiety linkage can be cleaved in the digestive tract.

The composition for preventing or treating the metabolic bone disorder preferably includes phosphopeptide salt and phytoestrogen in the ratio from 5:1 to 15:1 (phosphopeptide salt : phytoestrogen). More preferably, the ratio is 10:1. The composition is most preferably 10:1 bound calcium to phytoestrogen. The bound calcium is most preferably casein calcium phosphopeptide.

Surprisingly, it has been found that a combination of calcium phosphopeptide and phytoestrogens as described above have a synergistic effect in increasing bone-mass and reversing the effects of MBD.
The composition may be used in the prevention or treatment of a MBD characterised by calcium loss, or a condition in which calcium retention would be beneficial, particularly disorders such as osteoporosis, ostemalacia and secondary nutritional hypoparathyroidism.

In another aspect, the invention relates to a composition when used in the treatment or prevention of a metabolic bone disorder, the composition comprising phosphopeptide salt and phytoestrogen.

In a further aspect, the invention relates to use of a composition in the preparation of a medicament for the treatment or prevention of a metabolic bone disorder, the composition comprising phosphopeptide salt and phytoestrogen.

The invention also relates to a method of preparing a composition comprising phosphopeptide salt and phytoestrogen, which composition is used in the treatment or prevention of metabolic bone disorder, comprising the step of incorporating active ingredients in suitable pharmaceutical formulations such as tablet or capsule.

Preferably, in at least these aspects, the disorder is osteoporosis. The phosphopeptide salt is preferably casein calcium phosphopeptide and the ratio of the salt to the phytoestrogen is 10:1.

The method and the composition in accordance with the invention may also be used to prevent or reduce loss of calcium such as urinary calcium loss, to prevent or reduce calcium resorption, or preferably to increase or stimulate calcium retention. Most preferably calcium retention in bones is enhanced so as to alleviate or prevent metabolic bone disorders such as osteoporosis. The invention therefore also provides a method of enhancing calcium retention in a subject, comprising the step of administering a composition comprising phosphopeptide salt and phytoestrogen. In a particularly preferred embodiment, the calcium retention is in the bone, and the composition is casein calcium phosphopeptide and phytoestrogen in the ratio 10:1.
The phytoestrogen may be isolated from a single plant such as one from the *Leguminosae* family, or a source comprising a combination of two or more plants or members of the *Leguminosae* family. The phytoestrogen may be extracted using aqueous and organic solvents conventionally used in such processes or according to the processes described below.

In another aspect of the invention, it has been surprisingly found that phytoestrogen can be recovered in significant quantities from seeds of *Leguminosae* is used. Preferably, clover seeds are used, particularly red clover. The seeds are milled and the hulls removed. The phytoestrogens are then able to be used, i.e. bioavailable. This has the advantage that it is not necessary to extract the phytoestrogen from the plant material and seeds are easier to handle.

The composition in accordance with the invention may be prepared by admixing the phosphopeptide salt and phytoestrogen, preferably at concentrations such that the final proportion of phosphopeptide salt to phytoestrogen is 10:1. Most preferably, the composition comprises bound calcium (i.e. casein calcium phosphopeptide) to phytoestrogen at a ratio of 10:1. The composition is preferably administered at a dose of about 200 to 400 mg of CPP and about 40 mg of phytoestrogen per day, in one or two doses for adult humans.

The composition may be formulated with one or more suitable carriers, diluents or excipients, or in accordance with Remington’s Pharmaceutical Sciences, Merck Publishing Co, Easton, PA, USA. Other compounds such as dietary supplements including but not limited to vitamins and minerals may be present in this composition. The composition may be formulated for administration orally, transdermally by injection or for delivery by implantation of suitable devices.

In another aspect, the invention provides a composition for providing supplemental dietary calcium comprising:

ionic calcium;
milk protein-derived phosphopeptides;

a pharmaceutically acceptable carrier.

Preferably, the carrier is water. Preferably the calcium is naturally derived. Soy milk provides a natural source of calcium in a pharmaceutically acceptable carrier. The composition may be soy milk (where the isoflavone concentration is adequate) and milk protein-derived phosphopeptides.

In a preferred aspect of the invention, the composition also includes isoflavones. Preferably, the isoflavones are also derived naturally. In a most preferred form of the invention, the isoflavones are predominantly in an aglucose form. Again, soy milk contains naturally some phytoestrogen isoflavones. More such isoflavones may be added where necessary.

The phosphopeptide is preferably a casein phosphopeptide salt of a divalent cation. Most preferably, the salt is a casein calcium phosphopeptide.

The invention also relates to use of a composition as described above for the treatment of calcium deficiency disorders, including metabolic bone diseases such as osteoporosis. Further, the invention also includes a method of treating calcium deficiency disorders, such as metabolic bone diseases like osteoporosis by administration of a composition as described above in a therapeutically effective amount.

In a preferred form, the ratio of calcium phosphopeptide salt to phytoestrogen (or isoflavone) is between 10:1 and 2:1. Preferably, 100 ml of the composition contains:

- caseopeptide - about 40 mg
- isoflavones - about 8 to 21 mg
- calcium - about 150 mg
- soy protein - about 3.5 g.

The phosphopeptide of the invention may be produced by methods such as those described in Australian patent numbers 600225, 653401 and an alternative method is disclosed in Australian provisional patent application PR0819. These disclosures are incorporated herein by this reference. Further, the disclosures in the specifications of AU653401, WO 93/03707 and WO 98/40406 disclose phosphopeptide complexes which are suitable for use for the invention. These disclosures are also incorporated by reference. The invention extends to compositions as described above which include phosphopeptides produced by these methods.

The invention also includes the further addition of supplements to the composition. Preferably, the supplements include Vitamin D. Further, flavourings may also be added.

It will be understood that the term “comprises” (or its grammatical variants) as used in this specification is equivalent to the term “includes” and should not be taken as excluding the presence of other elements or features. It will also be understood that the term “consisting essentially of” (or its grammatical variants) when used in this specification and the claims means that the composition comprises only those components which it is said to “consist essentially of”, together with trace impurities and additional common components such as excipients as would be appreciated by one skilled in the art.

**Detailed Description of the Invention**

A preferred embodiment of the invention will now be described with reference to the following non-binding examples.

The preparation of calcium phosphopeptides is described in the references referred to above and incorporated herein.

Phytoestrogens may also be extracted using one or more of the methods
referred to above. Alternatively, phytoestrogen from *Leguminosae* plant seeds can be used. Seeds from red clover have been found particularly suitable. For example, the following red clover seed types, sourced from the Hamilton Genetic Resource Centre of the Department of Agriculture in Victoria, Australia, have been found to contain significant amounts of phytoestrogen (w/w) as follows:

- Colenso 3.1%
- Redquin 5.2%
- PAC19 3.8%
- ASTRED 4.0%

Other types of clover, such as white and pink clover, also have phytoestrogen in their seeds.

**Example 1**

In a first example, 20 grams of each of the above types of red clover seeds were milled to powder form. The phytoestrogen content of the remaining portion of the seeds was then measured by TLC and HPLC as known in the art using an organic solvent (eg methanol and acetonitrile). It was found that the seeds had about 50 grams per kilogram of total seed (including hulls) of phytoestrogen, compared with about 3 grams per kilogram in mature plant material by known phytoestrogen extraction methods.

Of practical importance, however, it was not necessary to extract the phytoestrogen from the seed in order for it to be pharmaceutically effective.

**Example 2**

In order to take advantage of the combined effect discovered between calcium phosphopeptide and phytoestrogen, a tablet composition was determined, and tablets were manufactured, composed as follows:
### Table 1

<table>
<thead>
<tr>
<th>Tablet Composition</th>
<th>Percentage</th>
<th>Milligram</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calcium Phosphopeptide</td>
<td>67</td>
<td>402</td>
</tr>
<tr>
<td>Phytoestrogen</td>
<td>7</td>
<td>42</td>
</tr>
<tr>
<td>Povidone</td>
<td>10</td>
<td>60</td>
</tr>
<tr>
<td>Cross-povidone</td>
<td>8</td>
<td>48</td>
</tr>
<tr>
<td>Magnesium Stearate</td>
<td>1</td>
<td>6</td>
</tr>
<tr>
<td>Microcrystalline Cellulose</td>
<td>7</td>
<td>42</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>100</strong></td>
<td><strong>600</strong></td>
</tr>
</tbody>
</table>

The phytoestrogen may be extracted using known methods from plant material, or a sufficient amount of ground clover seeds may be added (preferably with the hulls extracted) to provide the required amount of phytoestrogen. The remainder of the seed is harmless and no deleterious effects are experienced by incorporating it.

It will be seen that the ratio of calcium phosphopeptide to phytoestrogen was about 10:1. As will be appreciated by one skilled in the art, povidone and cross-povidone are commonly used in tablets to provide structure to a tablet so that it can survive packaging, transportation, storage and consumption. Magnesium stearate is used to facilitate the manufacturing process by reducing friction between components of the tablets. The microcrystalline cellulose is included to promote disassociation of the tablet upon contact with water as the cellulose absorbs water and expands, therefore breaking the tablet apart and exposing the active ingredients where required.
Example 3

The effectiveness of the phosphopeptide-phytoestrogen composition was then tested in chickens, which provide a convenient platform for measuring increased calcium absorption. A composition manufactured in accordance with Example 2 was added to the normal feed. Of two groups of chickens, one being used as a control, the composition made up approximately 1% by weight of the total food intake of the chickens. Over a period of two weeks, it was found that the bone and mineral content of the chickens consuming the composition (as measured by bone ash content) increased by between 10 and 50% compared with chickens in the control group.

In conditions such as rickets and osteoporosis, administration of the compound (preferably with Vitamin D supplements) improves symptoms and reverse bone-mineral content loss.

Example 4

400 mg of phosphopeptide derived from milk protein in accordance with the method described in Australian patent application no. PR0819 was added to 1.0 litres of water. 120 mg of isoflavones naturally present in commercially available soy milk or derivable by the methods described above, 1.5 g of ionic calcium and 3.5 g of soy protein were added, and the mixture agitation.

Example 5

In this example, a commercially available flavouring known to be suitable for flavouring soy milk was added to the composition described in example 4. Otherwise, a composition was prepared in the same manner.

The composition of examples 4 and 5 may then be prepared in a concentrated form which has significant advantages for promoting the sale of the product to likely purchasers. In one form of the invention, a sufficiently concentrated form may be prepared such that only 100 mls of the composition is
required. Such a composition has the advantage that it can be quickly consumed. Its composition would be primarily water and contain:

Table 2

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Per 100 ml</th>
<th>Per 250 ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Caseopeptide (CPP)</td>
<td>40 mg</td>
<td>100 mg</td>
</tr>
<tr>
<td>Isoflavones</td>
<td>8.8 to 21 mg</td>
<td>22 to 53 mg</td>
</tr>
<tr>
<td>Calcium</td>
<td>150 mg</td>
<td>375 mg</td>
</tr>
<tr>
<td>Soy Protein *</td>
<td>3.5 g</td>
<td>8.75 g</td>
</tr>
</tbody>
</table>

* Soy protein has the advantage of USFDA approval of a health claim for soy protein and cardiovascular disease risk reduction.

From a purchaser's viewpoint, it is consumed quickly more like a medicine than a drink. This is attractive to some consumers of the product who are in need of increased calcium intake but desire a “natural” treatment. There are certain health products already known on the market (eg. bacterial supplements) which are effectively promoted in small containers such that each container represents an appropriate daily dose. An aspect of this invention is that the product is marketable and acceptable to consumers is an advantage over known treatments.

Thus, a composition and method of treatment is disclosed of a synergistic effect of phytoestrogen and phosphopeptides combined to treat a serious medical problem.

It will be understood that the invention disclosed and defined in this specification extends to all alternative combinations of two or more of the individual features mentioned or evident from the text. All of these different
combinations constitute various alternative aspects of the invention.

References


3. Walter ED "Genistin and its aglucone, genistein" Vol 63, p 3273, December 1941
CLAIMS

The invention is defined by the following claims:

1. A method of preventing or treating metabolic bone disorder, comprising the step of administering a composition comprising phosphopeptide salt and phytoestrogen.

2. A method according to claim 1 in which the phosphopeptide salt is derived from a milk protein.

3. A method according to claim 2 in which the phosphopeptide salt is a casein phosphopeptide salt of a divalent cation.

4. A method according to claim 3 in which the salt is a casein calcium phosphopeptide.

5. A method according to any one of claims 1 to 4 in which the phytoestrogen is selected from the group consisting of diadzein, genistein, biochanin A, formononetin and glycitein.

6. A method according to claim 5 in which the phytoestrogen is selected from genistein and biochanin A.

7. A method according to any one of claims 5 and 6 in which the phytoestrogen is in the aglucose form.

8. A method according to any one of claims 1 to 7 in which the composition has a ratio of phosphopeptide salt : phytoestrogen of between 5:1 to 15:1.

9. A method according to claim 8 in which the ratio is 10:1.

10. A method according to any one of claims 1 to 9 for the prevention or treatment of a MBD characterised by calcium loss, or a condition in which
calcium retention would be beneficial.

11 A method according to claim 10 when used to treat osteoporosis, ostemalacia or secondary nutritional hypoparathyroidism.

12 A composition for the treatment or prevention of a metabolic bone disorder, the composition comprising phosphopeptide salt and phytoestrogen.

13 A composition according to claim 12 in which the phosphopeptide salt is derived from a milk protein.

14 A composition according to claim 13 in which the phosphopeptide salt is a casein phosphopeptide salt of a divalent cation.

10 15 A composition according to claim 14 in which the salt is a casein calcium phosphopeptide.

16 A composition according to any one of claims 12 to 15 in which the phytoestrogen is selected from the group consisting of diadzein, genistein, biochanin A, formononetin and glycetin.

15 17 A composition according to claim 16 in which genistein and biochanin A are used.

18 A composition according to any one of claims 16 and 17 in which the phytoestrogen is in the aglucone form.

19 A composition according to any one of claims 12 to 18 in which the composition has a ratio of phosphopeptide salt : phytoestrogen of between 5:1 to 15:1.

20 A composition according to claim 19 in which the ratio is 10:1.

21 Use of a composition according to any one of claims 12 to 20 in the preparation of a medicament for the treatment or prevention of a metabolic
bone disorder.

22 A method of preparing a composition comprising phosphopeptide salt and phytoestrogen, for the treatment or prevention of a metabolic bone disorder, comprising the step of incorporating active ingredients in suitable pharmaceutical formulations such as tablet or capsule.

23 A method of enhancing calcium retention in the bone, comprising administration of a composition according to any one of claims 12 to 20.

24 A method according to any one of claims 1 to 11 or a composition according to any one of claims 12 to 20 in which the phytoestrogen is isolated from a single type of plant such as one from the *Leguminosae* family, or a source comprising a combination of two or more types of plants or members of the *Leguminosae* family.

25 A method according to any one of claims 1 to 11 or a composition according to any one of claims 12 to 20 in which the phytoestrogen is sourced from seeds of *Leguminosae*.

26 A method according to claim 25 in which clover seeds are used.

27 A method according to claim 25 in which red clover seeds are used.

28 A method according to any one of claims 1 to 11 and 23 to 27 in which the composition is administered at a dose of about 200 to 400 mg of calcium phosphopeptide and about 40 mg of phytoestrogen per day, in one or two doses for adult humans.

29 The composition of claim 28 formulated with one or more suitable carriers, diluents or excipients for administration orally, transdermally by injection or for delivery by implantation of suitable devices.

30 A composition for providing supplemental dietary calcium consisting
essentially of:

ionic calcium;

milk protein-derived phosphopeptides;

a pharmaceutically acceptable carrier.

5 31 A composition according to claim 30 in which the carrier is water.

32 A composition according to any of claims 30 to 31 in which the calcium is naturally derived.

33 A composition according to any one of claims 30 to 32 in which the phosphopeptide is a casein phosphopeptide salt of a divalent cation.

10 34 A composition according to claim 33 in which the salt is a casein calcium phosphopeptide.

35 A composition according to any one of claims 30 to 34 in which substantially all components are derived from living sources without the use of organic solvents.

15 36 A composition according to any one of claims 30 to 35 including soy milk.

37 A composition according to any one of claims 30 to 36 further including isoflavones.

38 A composition according to claim 37 in which the isoflavones are derived naturally.

20 39 A composition according to claim 37 or 38 in which the isoflavones are predominantly in an aglucone form.

40 A composition according to any one of claims 30 to 39 in which the ratio of
calcium phosphopeptide salt to phytoestrogen (or isoflavone) is between 10:1 and 2:1.

41 A composition according to any one of claims 30 to 39 wherein 100 ml of the composition contains:

5

- caseopeptide - about 40 mg
- isoflavones - about 8 to 21 mg
- calcium - about 150 mg
- soy protein - about 3.5 g.

42 A composition according to any one of claims 30 to 41 which further includes Vitamin D.

43 A composition according to any one of claims 30 to 42 further including flavourings.

44 A nutriceutical or pharmaceutical including a composition according to any one of claims 30 to 43.

45 The use of a composition according to any one of claims 30 to 44 for the treatment of calcium deficiency disorders, including metabolic bone diseases such as osteoporosis in a therapeutically effective amount.

46 A method of treating calcium deficiency disorders, including metabolic bone diseases such as osteoporosis, comprising administration of a therapeutically effective amount of a composition according to any one of claims 30 to 44.
# INTERNATIONAL SEARCH REPORT

**A. CLASSIFICATION OF SUBJECT MATTER**

Int Cl:
- A61K 35/20, 35/78; A23J 7/00; A61P 19/08, 19/10

According to International Patent Classification (IPC) or to both national classification and IPC

**B. FIELDS SEARCHED**

Minimum documentation searched (classification system followed by classification symbols)
- IPC A61K 35/20, 35/78, A23J 7/00

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
- AU: IPC as above

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
- WPAT: phosphopeptide, phytoestrogen, isoflavone, bone, osteoporosis
- MEDLINE: phosphopeptide, phytoestrogen, isoflavone, bone, osteoporosis

**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

<table>
<thead>
<tr>
<th>Category*</th>
<th>Citation of document, with indication, where appropriate, of the relevant passages</th>
<th>Relevant to claim No.</th>
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<tr>
<td>P, Y</td>
<td>WO 00/45650 A (NUTRAHEALTH LTD) 10 August 2000, page 1, page 14 lines 6-20, claims 1-4, 29-30</td>
<td>1-46</td>
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</tbody>
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* Further documents are listed in the continuation of Box C

**See patent family annex**

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**Date of the actual completion of the international search**

09 July 2001

**Date of mailing of the international search report**

16 July 2001

**Authorized officer**

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Form PCT/ISA/210 (second sheet) (July 1998) COPGLA
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<tr>
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<th>Citation of document, with indication, where appropriate, of the relevant passages</th>
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INTERNATIONAL SEARCH REPORT
Information on patent family members

This Annex lists the known "A" publication level patent family members relating to the patent documents cited in the above-mentioned international search report. The Australian Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

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END OF ANNEX