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Plant and Plant-derived Compounds employed in Prevention of the Osteoporosis

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SUMMARY. Osteoporosis is now widely recognized as a public health problem since this disease, which increases bone fragility and thereby the risk of fractures, is associated with high mortality, morbidity and medical expenses throughout the world. The present work is a literature review of crude plant extracts and chemically defined molecules employed in the osteoporosis. The review refers to 69 plants with their families, parts used, type of extract, model tested and references. It also includes 46 compounds isolated from higher plants and which are classified in appropriate chemical groups. Some aspects of recent research with natural products from plants directed to the treatment or prevention of osteoporosis are discussed.

RESUMEN. "Plantas y compuestos derivados de plantas empleados en la prevención del osteoporosis". Actualmente la osteoporosis es reconocida como un problema de salud pública dado que esta enfermedad, que aumenta la fragilidad del hueso y por lo tanto el riesgo de fracturas, se asocia en todo el mundo a alta mortalidad y morbosidad y a altos costos médicos. El presente trabajo consiste en la revisión de la literatura con respecto a drogas vegetales, extractos semipuros y sustancias químicamente definidas, con actividad potencial en casos de osteoporosis. Esta revisión incluye 69 plantas, sus familias, partes utilizadas, tipos de extracto, modelos ensayados y la referencia correspondiente. Incluye también 46 sustancias aisladas de plantas superiores, clasificadas con respecto a su grupo químico. Se discuten algunos aspectos recientes de la investigación de productos naturales dirigida al tratamiento o prevención de la osteoporosis.

INTRODUCTION

Osteoporosis is a progressive systemic disease characterized by bone mass loss and the deterioration of bone microarchitecture, which leads to bone fragility and increased risk of fractures. It affects mainly postmenopausal women and is classified as primary (idiopathic), types I and II, and secondary. Type I, known as postmenopausal, is characterized by rapid bone loss and affects women shortly after the start of menopause, mainly in the trabecular bone and is associated with vertebrae and distal radio fractures. In contrast, type II, or senile, is associated with aging due to chronic deficiency of calcium, increase in parathormone activity and decrease in bone formation. The secondary type results from inflammatory processes, endocrine changes, multiple myeloma, sedentariness and the use of drugs such as heparin, corticoids and alcohol. The main risk factors for the onset of osteoporosis can be related to the person himself/herself (family history, white woman, presence of scoliosis, thin individuals, low stature, early gray hair) or to the environment in which he/she lives such as the use of alcohol and cigarettes (inhibitors of osteoblast multiplication); caffeine (increase in calcium excretion); sedentariness, malnutrition; diet rich in fibers; proteins and sodium (reduction in calcium absorption); nulliparity; amenorrhea due to exer-

KEY WORDS: Anti-osteoporotic, Bone deseases, Natural products, Osteoporosis, Plant extracts.
PALABRAS CLAVE: Antiosteoporosis, Enfermedades óseas, Extractos vegetales, Osteoporosis, Productos naturales.

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cising; early menopause, and endocrinopathy. The diagnosis is made on the basis of the clinical history, physical examination, besides complementary tests such as: blood cell count, VHS, calcium and phosphorus dosing, 24-h calciuria, bone formation markers (bone alkaline phosphatase, osteocalcin, and pro-collagen type I C-Terminal peptide), markers of bone reabsorption (hydroxyproline, pyridinoline, desoxipyridinoline and Ntx), besides X-ray image diagnostic and mainly bone densitometry, which is used for serial study and may determine bone loss extension, and verify the efficacy of prevention or treatment [1].

According to the World Health Organization, 1/3 of white women over 65 are affected by osteoporosis. It also may occur in men. It is estimated that a white man over 60 has a 25% possibilility of suffering an osteoporotic fracture [2].

There is no doubt that the main treatment of osteoporosis is prevention, for which bone mass peak and the prevention of postmenopausal reabsorption are critical elements. Several kinds of treatment can be used such as: calcium consumption as calcium carbonate, D vitamin supplements, and hormone reposition [3], the use of calcitonine to modulate serum levels of calcium and phosphorus [4], use of bisphosphonate, mainly alendronates [2], use of ipriflavone (bone reabsorption inhibitor), and sodium fluoride (increases trabecular bone mineralization), besides physical activity to strengthen muscles, stimulate osteoblast formation, and prevent reabsorption [1].

Considering the broad spectrum effect of osteoporosis in the medical system, currently an increasing demand is sought in the alternative system of medicine to design strategies to prevent and cure this devastating ailment.

In this work we have reviewed the literature related with natural products which act specifically in the treatment or prevention of osteoporosis.

The search was carred out on Biological Abstracts, Chemical Abstracts and the data bank of The University of Illinois in Chicago - NAPRALERT (Acronym for Natural Products ALERT), updated to December 2001, using osteoporosis plus plant. The references found in the search were later consulted.

The search for data from different sources led in the elaboration of a list of natural products evaluated specifically for osteoporosis (Tables 1 and 2). It should be noted that most of the references cited are not first hand observations, but compilations copied from other sources. The original references should be consulted for details on the models or mechanism based bioassays used for testing plant extracts and pure compounds against osteoporosis.

PLANT EXTRACTS INHIBITING OSTEOPOROSIS

Both estrogen and dietary calcium deficiencies are important risk factors in the pathogenesis of osteoporosis. Post-menopausal osteoporosis is considered to result from ovarian exhaustion. Age related bone loss is greatly accelerated in women after the menopause and women lose approximately 30% of their cortical bone during their life time. Mitra *et al.* [5] created an experimental model of menopause using ovariectomized rats. These animals developed bone changes similar to those seen in osteoporotic women as indicated by decrease in bone mineral content. OST-6 treatment (includes extracts of *Comiphora mukul, Terminalia arjuna* and *Withania somnifera)* demonstrated a dose dependent increase in the bone mineral content and bone mass in ovariectomized rats.

Chae *et al.* [6] evaluated the efficacy of a famous Korean prescription, "Dae-Bo-Won-Chun" DBWC for the treatment of osteoporosis. This product consists of extracts which contain a mixture of eight Korean traditional drugs, *viz, Angelica gigas, Cornus officinalis, Dioscorea batatas, Eucommia ulmoides, Glycyrrhiza uralensis, Lycium chinense, Panax ginseng* and *Rehmannia glutinosa.* The results obtained in the study provide evidence that DBWC contributes importantly to the prevention or treatment of the development of bone loss induced by ovariectomy in rats.

Kim *et al.* [7] studied the effects of five herbal medicine on travecular bone area using ovariectomized rats as an animal model of Type I osteoporosis and SAM P6 as that of Type II. Each is a traditional boiling water extract of *Achyrathes bidentata, Astragalus membranaceus, Cornus officinalis, Psoralea corylifolia* and *Rehmannia glutinosa* and was given 5 g/kg/day, p.o., for 30 days in a group of 4-5 ovariectomized rats. The traditional hot water extract of Cervi parvum cornu (Cervi) was given in the same dose as described above for 14 days in a group of 10 SAM P6 mice. Travecular bone area was measured 5 mm decalcified and stained thin bone shoe by image analysis using a digitalizer. In Type I, any administration of herbal medicine used in this study did not elevate travecular bone area significantly except *Cornus offici-*

Plant	Family	Part Used	Extract	Model	Reference
Achyranthes bidentata	Amaranthaceae	Root	Aqueous ext	Ovariectomized rat	7,15
Adenophora triphylla	Campanulaceae	Part not specified (**)	Type ext not stated (**)	PTH-induced calcium	16
Agrimonia pilosa	Rosaceae	Part not specified (**)	Polyphenol-rich aq. ext.	Human adult	17
Angelica archangelica	Apiaceae	Root	Part used powdered	Rat treated with GnRH agonist Ovariectomized rat	18 19
Angelica gigas	Apiaceae	Root	Aqueous ext	Ovariectomized rat	6
Astragalus membranaceus	Leguminosae	Root	Aqueous ext	Ovariectomized rat	7,15
Astragalus specie	Leguminosae	Root	Part used powdered	Rat treated with GnRH agonist Ovariectomized rat	18 19
Atractylodes lanceae	Compositae	Rhizome	Part used powdered	Rat treated with GnRH agonist Ovariectomized rat	18,20 19
Bauhinia forficata	Leguminosae	Part not specified (**)	Type ext not stated (**)	PTH-induced calcium	16
Belamcanda sinensis	Iridaceae	Part not specified (**)	Type ext not stated (**)	Ovariectomized rat	21
Boswellia serrata	Burseraceae	Stem fresh	Juice	Human adult	22
Bupleurum chinense	Umbelliferae	Root	Part used powdered	Rat treated with GnRH agonist Ovariectomized rat	18 19
Bushen Jiangu (*)	Not specified (**)	Part not specified (**)	Type ext not stated (**)	Human adult	23
Callicarpa japonica	Verbenaceae	Part not specified (**)	Type ext not stated (**)	PTH-induced calcium	16
Camellia sinensis	Theaceae	Part not specified (**)	Polyphenol-rich aq. ext.	Human adult	17
Cassia mimosoides	Leguminosae	Part not specified (**)	Type ext not stated (**)	Human adult	24
Cimicifuga foetida	Ranunculaceae	Rhizome	Part used powdered	Rat treated with GnRH agonist	18
			EtOAc ext	Ovariectomized rat	25
Cimicifuga heracleifolia	Ranunculaceae	Rhizome	EtOAc ext	Ovariectomized rat	25
Cimicifuga specie	Ranunculaceae	Rhizome	Type ext not stated (**)	Human adult	26
Cinnamomum zeylanicum	Lauraceae	Part not specified (**)	Type ext not stated (**)	PTH-induced calcium	16
Cistanche salsa	Orobanchaceae	Aerial parts	EtOH ext	Ovariectomized rat	13
Citrus aurantium	Rutaceae	Pericarp	Part used powdered	Ovariectomized rat	18,19

(Cont.)

Plant	Family	Part Used	Extract	Model	Reference
Clerodendrum trichotomum	Verbenaceae	Part not specified (**)	Type ext not stated (**)	PTH-induced calcium	16
Cnidium monnieri	Apiaceae	Fruit	Coumarin fraction	Ovariectomized rat	27,28
		Part not specified (**)	Type ext not stated (**)	Human adult	29
Coleus specie	Labiatae	Part not specified (**)	Type ext not stated (**)	PTH-induced calcium	16
Commiphora mukul	Burseraceae	Gum resin	Type ext not stated (**)	Ovariectomized rat	5
Cornus officinalis	Cornaceae	Fruit	Aqueous ext	Ovariectomized rat	6,7,15
Curcuma longa	Zingiberaceae	Rhizome fresh	Juice	Hulman adult	22
Cynomorium specie	Cynomoriaceae	Part not specified (**)	Type ext not stated (**)	Ovariectomized rat	30
Dalbergia subcymosa	Leguminosae	Part not specified (**)	Type ext not stated (**)	PTH-induced calcium	16
Drynaria baronii	Polypodiaceae	Rhizome	EtOH ext	Osteoblast-like UMR 106 cells	31
Drynaria specie	Polypodiaceae	Part not specified (**)	Type ext not stated (**)	Ovariectomized rat	30
Dioscorea batatas	Dioscoreaceae	Rhizome	Aqueous ext	Ovariectomized rat	6
Dioscorea specie	Dioscoreaceae	Rhizome	Part used powdered	Ovariectomized rat	32
Epimedium leptorrhizum	Berberidaceae	Steam	Part used powdered	Ovariectomized rat	33
Epimedium sagittalum	Berberidaceae	Aerial parts	Aqueous ext	Ovariectomized rat	34
Epimedium specie	Berberidaceae	Aerial parts	Part used powdered	Ovariectomized rat	20,30
Eucommia ulmoides	Eucommiaceae	Bark	Aqueous ext	Ovariectomized rat	6,35
Glycine max	Fabaceae	Seed	Protein fraction	Ovariectomized rat	36, 37,38,39
Glycyrrhiza glabra	Leguminosae	Root	Part used powdered	Ovariectomized rat	18,19
Glycyrrhiza uralensis	Leguminosae	Root	Aqueous ext	Ovariectomized rat	6
Gonystylus keithii	Thymelliaceae	Leaf	MeOH ext	PTH-induced calcium	40
Iris tectorum	Iridaceae	Part not specified (**)	Type ext not stated (**)	Ovariectomized rat	21
Kanggusong (*)	Not specified (**)	Part not specified (**)	Type ext not stated (**)	Ovariectomized rat	41
Liuwei Dihuang Wan (*)	Not specified (**)	Part not specified (**)	Type ext not stated (**)	Ovariectomized rat	42
Lycium chinense	Solanaceae	Fruit	Aqueous ext	Ovariectomized rat	6
Morus albus	Moraceae	Part not specified (**)	Polyphenol-rich aq. ext.	Human adult	17
Notopterygium forbesii	Umbelliferae	Root	MeOH ext	Human adult	43

(Cont.)

Notopterygium franchetii	Umbelliferae	Root	MeOH ext	Human adult	43
Panax ginseng	Araliaceae	Root	Part used powdered	Rat treated with GnRH agonist	18
			Aqueous ext	Ovariectomized rat	6
Persea specie	Lauraceae	Fruit	Oil unsaponified fract.	Human adult	44
Piper methysticum	Piperaceae	Part not specified (**)	Kava ext commercial	Human adult	45
Podophyllum emodi	Berberidaceae	Root	MeOH ext	Human adult	43
Podophyllum peltatum	Berberidaceae	Root	MeOH ext	Human adult	43
Polygonum multiflori	Polygonaceae	Root	Part used powdered	Ovariectomized rat	46
Psoralea corylifolia	Leguminosae	Root	Aqueous ext	Ovariectomized rat	7,15
Ptychopetalum specie	Olacaceae	Part not specified (**)	Type ext not stated (**)	PTH-induced calcium	16
Punica granatum	Lythraceae	Part not specified (**)	Type ext not stated (**)	PTH-induced calcium	16
Rehmannia glutinosa	Scrophulariaceae	Root	Aqueous ext	Ovariectomized rat	7,15
Rosa sp	Rosaceae	Part not specified (**)	EtOH (95%) ext.	Human adult	47
Rubus suavissimus	Rosaceae	Part not specified (**)	Polyphenol-rich aq. ext.	Human adult	17
Sambucus nigra	Caprifoliaceae	Stem and branch	EtOH 50%	Human adult	48
Sambucus sieboldiana	Caprifoliaceae	Stem	EtOAc ext	Ovariectomized rat	12
Sambucus williamsii	Caprifoliaceae	Stem and branch	EtOH 50%	Human adult	48
Sanguisorba officinalis	Rosaceae	Part not specified (**)	Polyphenol-rich aq. ext.	Human adult	17
Solanum paniculatum	Solanaceae	Part not specified (**)	Type ext not stated (**)	PTH-induced calcium	16
Terminalia arjuna	Combretaceae	Bark	Type ext not stated (**)	Ovariectomized rat	5
Thujopsis dolabrata	Cupressaceae	Root	MeOH ext	Human adult	43
Tripterygium wilfordii	Celastraceae	Part not specified (**)	Type ext not stated (**)	Human adult	49,50
Withania somnifera	Solanaceae	Root dried	Type ext not stated (**)	Ovariectomized rat	5
		Root fresh	Juice	Human adult	22
Zingiber officinale	Zingiberaceae	Rhizome	Part used powdered	Rat treated with GnRH agonist	18
Ziziphus specie	Rhamnaceae	Fruit	Part used powdered	Rat treated with GnRH agonist	18

Table 1. Plant extract inhibiting osteoporosis. (*) Effect of a traditional chinese herbal medicine. (**) Data incomplete, derived from an abstract

Chemical name	Class	Source	Model	Reference
Angelicain	Chromone	*Cimicifuga* specie	Human adult	26
Antibiotic F-10463-A	Alkaloid	*Dasyscyphus* specie	Human adult	51
Arginine, L	Aminoacid	Commercial	Osteoblast culture of cells	52
Berberine	Alkaloid	Kampo formula (Tsu-kan-gan)	PTH-induced calcium	14
Camelliatannin	Tannin	*Camellia japonica*	Ovariectomized rat	53
Catechin	Flavonoid	Found in many plants	Human adult	17
Catechin gallate	Flavonoid	Found in many plants	Human adult	17
Coniferyl alcohol	Phenylpropanoid	*Sambucus sieboldiana*	Ovariectomized rat	12
Curcumin	Diferuloylmethane	*Curcuma longa*	Human adult	54
Cyclosporin A	Oligopeptide	*Trichoderma polysporum*	Measured by radioimmunoassay	55
Daidzin	Flavonoid	*Glycine max*	Ovariectomized rat	37,56,57
Epicatechin	Flavonoid	Found in many plants	Human adult	17
Escin, α	Saponin	*Aesculus hippocastanum*	Ovariectomized rat	58
Gallic acid	Benzenoid	Found in many plants	Human adult	17
Genistein	Flavonoid	*Glycine max*	Effect in the femoral tissue of rat Ovariectomized rat Human adult	59 37 60
Genistin	Flavonoid	*Glycine max*	Ovariectomized rat	56
(2E, 6R)-8-Hydroxy-2,6-dimethyl-2-octenoic acid	Terpene	*Cistanche salsa*	Ovariectomized rat	13
Icariin	Flavonoid	*Epimedium* specie	Human adult	61
Isoferulic acid	Phenylpropanoid	*Cimicifuga* specie	Human adult	26
Isoimperatorin	Coumarin	*Cimicifuga* specie	Human adult	26
Lysine, L	Aminoacid	Commercial	Osteoblast culture of cells	52
Neocucurbitacin A	Terpene	*Luffa operculata*	Human osteoblast cell	62

(Cont.)

Norbisunadin	Terpene	*Cimicifuga* specie	Human adult	26
Osthole	Coumarin	*Cnidium* sp	Ovariectomized rat	63
Oxopodopyrone, 9´	Pyrone	*Gonystylus heithii*	Ovariectomized rat	40
Oxopodopyrone, 10´	Pyrone	*Gonystylus heithii*	Ovariectomized rat	40
Oxopodopyrone, 8-methyl-9´	Pyrone	*Gonystylus heithii*	Ovariectomized rat	40
Oxopodopyrone, 8-methyl-10´	Pyrone	*Gonystylus heithii*	Ovariectomized rat	40
Peltatin, α	Lignan	*Podophylum peltatum*	Human adult	64
Peltatin, β	Lignan	*Podophylum peltatum*	Human adult	64
Podophyllotoxin	Lignan	*Podophylum peltatum*	Human adult	64
Podophyllotoxin, 4´-demethyl	Lignan	*Podophylum peltatum*	Human adult	64
Podophyllotoxin, desoxy	Lignan	*Podophylum peltatum*	Human adult	64
Resveratrol	Stilbene	Found in many plants	Human adult	65
Rutin	Flavonoid	Found in many plants	Ovariectomized rat	66
Shinflavanone	Flavonoid	*Glycyrrhiza glabra*	Osteoclast cell culture *in vitro*	67
Tectorigenin	Flavonoid	*Iris tectorum*	Ovariectomized rat	21
Triptolide	Terpene	*Tripterygium wilfordii*	Osteoblasts detected by MTT methods	68
Vanillic acid	Benzenoid	*Sambucus sieboldiana*	Ovariectomized rat	12
Vanillin	Benzenoid	*Sambucus sieboldiana*	Ovariectomized rat	12
Vitamin C	Vitamin	Commercial	Human adult	54
Vitamin D	Vitamin	Commercial	Ovariectomized rat	69
Vitamin E	Vitamin	Commercial	Orchidectomized rat Human adult	70 54
Vitamin K	Vitamin	Commercial	Ovariectomized rat	69
WF14861	Aminoacid	*Colletotrichum* specie	Low-calcium-diet-fed mouse model	71
Xylitol	Carbohydrate	Commercial	Streptozotocin-diabetic rat	72

Table 2. Chemically defined molecules inhibiting osteoporosis.

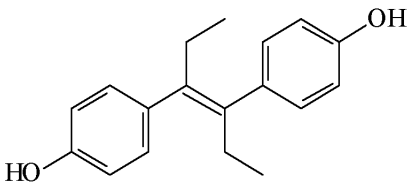
nalis, that showed a trend of increase in trabecular bone area. However Type II, Cervi and *Astragalus membranaceus* increased both mean and total trabecular bone area. Thus, there are significant differences in response of herbal medicine in different types of osteoporosis.

To investigate the bioactivities of soyabean *(Glycine max),* which act on bone metabolism, Choi *et al.* [8] studied the effect of a soyabean ethanol extract on the activity of osteoblast MC3T3-E1 cells. Soya extract (0.01-0.1 g/l) dose-dependently increased survival ($P < 0.05$) and DNA synthesis ($P < 0.05$) of MC3T3-E1 cells. In addition, soya extract (0.05 g/l) increased alkaline phosphatase activity ($P < 0.05$) and collagen synthesis ($P < 0.05$) of MC3T3-E1 cells. Moreover, the synthetic classic anti-oestrogen tamoxifen eliminated the stimulation of MC3T3-E1 cells on the proliferation, ALP activity and collagen synthesis by soy extract, indicating that the main action of the soya extract on osteoblastic MC3T3-E1 cells is similar to the effects of oestrogen. Treatment with soya extract prevented apoptosis, as assessed by a one-step sandwich immunoassay and DNA gel electrophoresis studies. This effect may be associated with the activation of the oestrogen receptor, since it was shown that soya extract-mediated survival against apoptosis was blocked by the oestrogen receptor antagonist tamoxifen in cells, further supporting a receptor-mediated mechanism of cell survival. These results suggest that osteoblast function is promoted by soya extract and that the oestrogen receptor is involved in the response, thereby playing an important role in bone remodeling. The authors conclude that soya extract has a direct stimulatory effect on bone formation in cultured osteoblastic cells *in vitro.* Presumably, dietary soya products are useful in the prevention of osteoporosis.

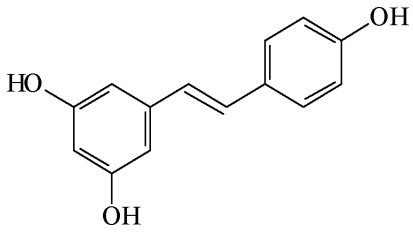
The results of the literature survey are presented in Table 1, which lists 69 extracts of plants used in osteoporosis. The plants are listed in alphabetical order of their scientific names followed by the plant family, part used, type of extract, model tested, and references.

PLANT DERIVED-COMPOUNDS INHIBITING OSTEOPOROSIS

Estrogen replacement therapy (ERT) is recommended for postmenopausal women primarily for reduction of menopausal symptoms and prevention of osteoporosis. However, only 35% to 40% of women ever start ERT, and many do not continue it. One of the reasons women are reluctant to receive postmenopausal ERT is that they perceive prescription estrogens as being "unnatural" [9]. Because of this, there is increasing interest in the use of plant-derived estrogens, also known as phytoestrogens. Conventional ERT drugs, especially diethylstilbestrol (I), have been shown to cause serious side effects including stroke, gallbladder disease and certain types of cancer. Resveratrol (II) is a powerful phytoestrogen found in grape skin and other plant foods as well as wine. Studies have shown that resveratrol enhances estrogen metabolism through the formation of a complex with estrogen receptors, and can help women maintain normal estrogenic activity, reduce hot flushes, balance mood swings, maintain healthy bone density, promote cardiovascular health and prevent the effects of premature aging [10].



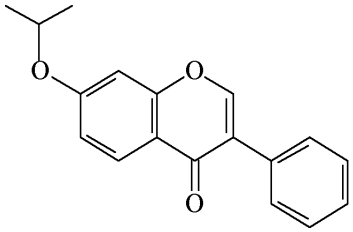
(I)



(II)

Genistein (IV) is an isoflavone abundantly present in soybeans *(Glycine max),* which shows a structural similarity to the synthetic oestrogen ipriflavone (III), suggesting that it may act as a phytoestrogen. Morita et al. 11 investigated the effect of genistein on the expression of type I collagen (COL I), alkaline phosphatase (AP), osteopontin (OP) and osteocalcin (OC) genes that have been associated with bone formation in mouse osteoblastic cells (MC3T3-E1 cells) and OVX mice. In MC3T3-E1 cells, genistein as well as oestrogen increased the amount of OP mRNA. No significant differences were observed in the levels of COL I, AP and OC mR-

NAs. In OVX mice, the weight of the uterus was significantly decreased compared with the sham-operated controls. Oestrogen completely restored the weight of the uterus in the OVX mice, whereas genistein had no effect. The levels of the four transcripts in the bone were markedly decreased in the OVX mice. Oestrogen increased the levels of COL I mRNA, but not those of AP, OP or OC mRNA. Similar findings were observed in the genistein-treated OVX animals. These results indicate that genistein exhibits oestrogenic action in the bones of OVX animals without oestrogenic action in the uterus. Thus, these results suggest that soyabean containing genistein may be a useful nutritional source in the prevention of osteoporosis.



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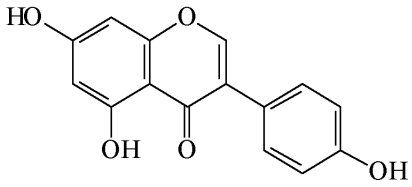
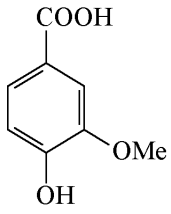


(IV)

A methanolic extract of the stems of *Sambucus sieboldiana,* found to inhibit bone resorption in organ culture, was subjected to further fractionation guided by the activity towards bone resorption stimulated by parathyroid hormone (PTH) *in vitro* [12]. The ethyl acetate fraction (EtOAc-fr) of the methanolic extract inhibited PTH-stimulated bone resorption of neonatal mouse bones, and the inhibitory activity was more potent than those of other fractions. Oral administration of the EtOAc-fr (50 or 100 mg/kg per day) to ovariectomized (OVX) rats prevented the decrease in bone mineral density (BMD) of the lumbar (L2-4) vertebrae, indicating that the EtOAc-fr is effective *in vivo.* EtOAc-fr (50, 100 or 150 mg/kg per day) decreased the serum calcium level elevated in rats with insufficient dietary calcium. The phenolic constituents of EtOAc-fr were examined for their inhibitory effect on bone resorption stimulated by PTH in neonatal mouse bone. Among them, vanillic acid (V), vanillin (VI) and coniferyl alcohol (VII) showed significant inhibitory effects on bone resorption. Of the compounds examined, vanillic acid was found to have a significant inhibitory effect on the decrease of BMD in OVX mice. Thus the EtOAc fraction of *S. sieboldiana* showed a suppressive effect on bone resorption both in vitro and *in vivo;* these inhibitory effects may be at least partly due to the inhibitory action of vanillic acid.



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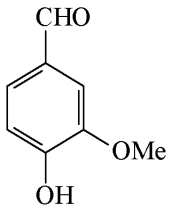

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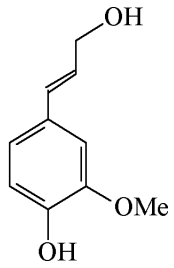
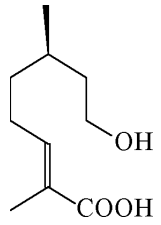


(VII)

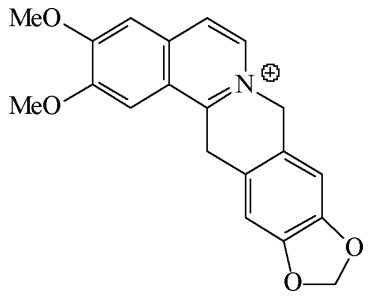
(2E,6R)-8-Hydroxy-2,6-dimethyl-2-octenoic acid [(R)-VIII], a novel monoterpene from *Cistanche salsa,* a Chinese herb, was found to be an anti-osteoporotic compound. The extract of *C. salsa* significantly suppressed the bone weight loss in ovariectomized mice, a postmenopausal osteoporosis model. The active substance was then purified by using this osteoporotic model and the chemical structure was determined. The active compound from *C. salsa,* (R)-VIII, supressed the decrease of bone weight and the mechanical strength in the ovariectomized mice. Furthemore, (R)- and (S)-VIII were synthesized and the activity of each

was evaluated. (R)-VIII suppressed the bone weight loss, although (S)-VIII did not show any activity [13].



Structure of (R)-VIII

Berberine (IX) is an alkaloid previously isolated from aqueous extracts of Tsu-kan-gan, a Kampo formula used for the treatment of osteoporosis. Li *et al.* [14] verified that oral administration of berberine (daily dose of 30 or 50 mg/kg) to ovariectomized rats prevented a decrease in bone mineral density (BMD) of the lumbar vertebra without affecting the weight of the uterus and plasma concentration of estradiol. The results suggest that berberine prevents a decrease in BMD *in vivo* by inhibiting osteoclastic bone resorption.



(IX)

Forty six chemically defined natural molecules have been reported in the literature as useful for the treatment or prevention of osteopososis (see Table 2). Of the 46 active compounds, ten are flavonoids; five lignans; four pyrones, terpenes and vitamins; three aminoacids and benzenoids; two alkaloids, coumarins and phenylpropanoids; one carbohydrate, chroman, diferruloilmetane, peptide, saponin, stilbene and tannin.

CONCLUSION

Osteoporosis is a progressive disease characterized by the decrease in bone mass which has major consequences for the patient. Considering there is no effective means to make up for the lost bone, there is no doubt that the main strategy at present is prevention, along with basic care to prevent fractures and mainly the elimination of risk factors such as: tobacco, alcohol, coffee, sedentariness, and an inadequate diet with low calcium intake. In the case of family history of osteoporosis, peri- and postmenopausal, yearly control must be made with bone dosimetry, followed with hormone reposition and calcium and D vitamin supplementation if necessary.

Due to the large incidence of this pathology throughout the world and taking into consideration work published in this area on the use of natural products in osteoporosis control, we have found it appropriate to organize this information. Furthermore, there is a large variety of plants in the world not studied yet, and thus, there is a potential to reach a stage in the future when osteoporosis will no longer be a threat.

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