

# Rapeseed meal nutraceuticals

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#### Abstract

Many components of human diet previously overlooked are now considered to play an important role in the prevention and treatment of important diseases. Over the last decade disease preventing food and ingredients (called nutraceuticals) have written some success stories. Today the most important nutraceutical formulations contain vitamins, with antioxidant properties, oligosaccharides, bioactive peptides, phenolic compounds, glucosinolates and other products. Studies showed that cruciferous seed meals that include rapeseed exerted protective effects against tumour formation and growth. Rapeseed is reported to have a broad range of therapeutic effects, including antioxidant, anti-inflammatory, antitumour and anticancer effect. The medicinal properties of rapeseed are due to presence of variety of phytochemicals imparting the effect. In the laboratory, rapeseeds have been found to have very strong antioxidant activity, inhibiting cancer cell proliferation, decreasing lipid oxidation, and lowering cholesterol. While extensive research have been done, a literature review for health benefits of rapeseed meal and their phytochemicals has not been compiled to summarize the investigations to provide a baseline for considering rapeseed meal in isolating nutraceuticals and developing functional food. This review attempts to report the medicinal properties of individual bioactive component, with the recent updates in vivo and in vitro research to explore the possibility of utilizing rapeseed meal in development of functional food. The consideration of all the bioactive components of rapeseed meal is beyond the purview of this review. Hence, only extensively studied major bioactive components viz. rapeseed phenolics, glucosinolates, dithiolthiones, phytates, phytosterols and bioactive peptides are considered.

Keywords: rapeseed meal, nutraceuticals, phytochemicals, bioactive components, functional food

#### Introduction

Specific dietary recommendations for treating or preventing various types of illness have been documented in since Vedic era (Anon, 2000). Hippocrates correctly emphasized "Let food be your medicine and medicine be your food", about 2000 years ago (Hasler, 2002). Presently, functional food is the topic of interest in the food and nutrition industry. There are various terms in market for explaining foods with health benefits, a few of them being bioactive compounds, functional ingredients, natural health products, designer foods, novel foods, phyto-nutrients, medical foods, dietary supplements, etc. However, the terms nutraceuticals and functional foods are most common. Among all these foods with health benefits, there seems to be a thin dividing line in their interchangeable usage by different people on different occasions. *"Functional food"* can be defined as the food or beverages consumed as part of the usual diet, which have physiological benefits beyond basic nutrition and/or reduce the risk of chronic disease. Hence, functional foods are similar in appearance to a conventional food and have demonstrated physiological benefits, and/or reduce the risk of chronic disease beyond basic nutritional functions. While as *"nutraceuticals"* are isolated active ingredient in a medicinal form such as pill or powder and/or reduces the risk of chronic disease (Anon., 2003). The definition of functional foods still varies among countries for reasons that are historical, cultural and regulatory. In its broadest use, functional foods are food-derived products that, in addition to their nutritional value, enhance normal physiological or cognitive functions or prevent the abnormal function that underlies disease.

Global nutraceutical food market is likely to cross US\$187 billion in sales by 2010. Along with the growing healthcare industry, in India there is an emerging trend in 'Fast Moving Healthcare Goods (FMHG)'. According to Cygnus estimates, nutraceuticals market in 2007 was INR18.75 billion and was expected to grow at 20% CAGR to achieve a market size of INR27 billion in 2009 in India. There is, thus, a proliferation of these value-added products aimed at not only keeping oneself healthy but also prevention/treatment of various ailments ranging from heart diseases to cancer.

India's share in global nutraceutical market is only 0.9% (FICCI, 2009). Considering the huge growth potential of the Indian Nutraceutical industry, India is reeling under the burden of nutrition deficiencies with one-fifth of the Indian population lacking purchasing power to even consume a diet sufficient in calories, let alone nutrients; and an astounding 570 million consuming sufficient or excess calories, but lacking adequate intake of nutrients. India can capitalize on consumers' interest in functional food by considering the major research in this area and developing the functional food from the sources which are cheaply available, out of which rapeseed meal could be a boon and seek efforts in exploring its strength as a source of nutraceuticals. Nevertheless, functional foods contain larger profit margins than conventional foods (30 to 500 percent higher).

Oilseed crops in India accounted for almost 5% of Gross National Product (GNP) in 2006 and 10% of the value of agricultural products (Anon., 2006). Rapeseed-mustard (*Brassica*) is a group of crops that contributes 32% of total oilseed production in India and it is the second largest indigenous oilseed crop (Meena *et al.*, 2010). Mustard research and promotion consortium provided an advanced estimate of production of rapeseed in 2008 at 4.81 m t with production area of 5.02 m ha.

Under the name rapeseed-mustard, seven important annual oilseeds belonging to the Brassicaceae (*Cruciferae*) are grown in India. They are Indian mustard (*Brassica juncea* [L.] Czern. & Coss.), commonly called rai (raya or laha), the three ecotypes of Indian rape, *B. rapa* L. ssp. *oleifera* viz., *toria*, *brown sarson* (*lotni* and tora types) and yellow sarson, Swede rape or gobhi sarson (*B. napus L.*), Ethiopian mustard or karan rai (*B. carinata* Braun.) and taramira or tara (*Eruca sativa Mill.*). On the Indian subcontinent *B. juncea* is the dominant species grown and along with *B. rapa* (syn. *B. rapa* L.) and *B. napus* L. are the important sources of edible oil in India. These species are regarded as of Asiatic origin.

Rapeseed has been consumed by humans as a condiment for about 3000 years. The original use of rapeseed-mustard was to mask the taste of degraded perishables (Hemmingway, 1993). The spiciness of rapeseed-mustard is caused by a group of compounds called isothiocyanates. When mustard seeds are crushed and exposed to liquid, an enzyme called myrosinase (thioglucoside glucohydrolase) hydrolyses the glucosinolates to release isothiocyanates. The differences in mustard flavours are due to the structural changes of the released isothiocyanates. For example, oriental and brown mustard (Brassica juncea) release a volatile compound allyl isothiocyanate (AIT), which produces a sharp taste sensation and pungent aroma similar to horseradish. Yellow mustard (Sinapis alba) releases a non-volatile compound, 4-hydroxybenzyl isothiocyanate (PHBIT), which provides a hot mouth feel in condiments (Cui, 1997).

Rapeseed/Mustard oil is generally used as cooking medium in northern states of India, while there are also other uses like lamp oil, in soap making, plastic manufacturing and as a high temperature, tenacious high-erucic acid lubricating oil. Rapeseed-mustard does not get a good price in the international market due to presence of high erucic acid content. However, with the sustained efforts of the society for rapeseed-mustard research, rapeseed varieties are now available for cultivation with lower erucic acid content (Arvind Kumar et al., 2009). Rapeseed-mustard oil is rich in alpha-linolenic acid. It contains high amount of mono-unsaturated fatty acids and a good ratio of polyunsaturated fatty acids, which is good for heart. It contains the least amount of saturated fatty acids, making it safe for heart patients. The relatively high level of oleic acid and the favourable balance between linolenic and linoleic acids is present in mustard oil. It may be said that it is the safest oil and is as good as any other edible oil (Anon., 2008). Rapeseed meal, in spite of having high protein content and nutritional value, is still underutilized and worth minimum price gain due to its use as fertilizer. Numerous research and reviews are available in justifying utilization of rapeseed oil as functional food. However, less emphasis was given in utilizing the rapeseed meal in developing health food products and it is hard to find information on potential benefits of minor components of rapeseed meal having medicinal value. Hence, this review attempts to highlight the potential of rapeseed meal as a source of nutraceuticals in developing highly valued functional food products. The information summarized in the review highlights medicinal properties of individual bioactive component from rapeseed meal viz. phenolics, glucosinolates, dithiolthiones, phytates, phytosterols and bioactive peptides, with the recent updates in in vivo and in vitro research, to present a broad range of therapeutic properties, including antioxidant, anti-inflammatory, antitumour, anticancer effects, etc.

## **Rapeseed Phenolics**

Several plant-derived medicines, which can prevent or cure diseases, are rich in phenolic compounds (Scalbert, 1993). In particular, phenolic compounds have been shown to exhibit protection against coronary heart disease and carcinogenesis (Hertog *et al.*, 1995). Rapeseed contains more phenolic compounds than any other oilseed plant (Nowak *et al.*, 1992). Rapeseed phenolics include esterified phenolic acids, free phenolic acids, and insoluble-bound phenolic acids (Krygier *et al.*, 1982). Rapeseed phenolic compounds are potent antioxidants in various environments relevant to food products. The most phenolics remain in the meal when the oil is pressed from the seeds. But some phenolics are also found in crude rapeseed oil (Koski et al., 2003). The most significant of these phenolic compounds in rapeseed is *sinapine*, the *choline* ester of sinapic acid (ca. 80% of the total phenolic compounds) (Kozlowska et al., 1990). Sinapic acid in rapeseed also exists as the glucosidic ester, glucopyranosyl sinapate (Amarowicz and Shahidi, 1994). Rapeseed, especially rapeseed meal, is rich in phenolic compounds. The main phenolic compounds in rapeseed meal were *sinapine* (the choline ester of sinapic acid) and sinapic acid while the sinapic acid was also found with one-tenth that of sinapine (Naczk et al., 1998; Cai and Arntfield, 2001). Typically the amount of sinapic acid derivatives in rapeseed meal varies between 6390 and 18370 µg/g depending on the variety of oilseed plant and the oil processing method (Kozlowska et al., 1990).

The total phenolic content rapeseed oil is around 439  $\mu$ g/g. The main phenolic compound in crude post-expelled rapeseed oil was vinylsyringol (i.e. 245-700ug/g) (Koski *et al.*, 2003), a decarboxylation product of sinapic acid, followed by sinapine and sinapic acid. Amongst nonpolar phenolic compounds tocopherols is predominant. Sinapic acid content of crude rapeseed oil falls in the range of 23 – 34 ug/g.

Food tannins are polyphenolic compounds, which are widely distributed in plants. They can be classified as condensed or hydrolyzable tannins. Most of the tannins in rapeseed are *condensed tannins*, formed by polymerization of flavan-3-ols or flavan-3,4-diols. The amount of tannins in rapeseed depends on the variety, the degree of maturation and extraction method, and varies from 0.2 to 3% of defatted rapeseed meal (Naczk *et al.*, 1998).

Antioxidant Properties : In biological systems, an antioxidant can be defined as any substance that, in low concentration compared with the oxidizable substrate, significantly delays or prevents oxidation of that substrate. The substrate, i.e. the oxidizable compound, is usually a lipid, but can also be a protein, DNA, or carbohydrate. In the case of lipid oxidation, the main mechanism of antioxidants is to act as radical chain-breakers. Another mechanism is to act as preventive antioxidant oxygen scavenging or blocking the pro-oxidant effects by binding proteins that contain catalytic metal sites (Frankel and Meyer, 2000).

Some earlier studies have been made on the antioxidant activity of rapeseed phenolics. Several studies of these have shown that phenolic compounds have antioxidant properties. The effect of rapeseed phenolics on radical scavenging has been investigated by Amarowicz et al. (2000) and Matthäus (2002). Wanasundara and Shahidi (1994) found that the antioxidant activity of ethanolic (95%) extract of rapeseed meal toward the oxidation of rapeseed oil was better than that of some widely used synthetic antioxidants, butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT) and the combination of BHA, BHT, and monoglyceride citrate (BHA/BHT/MGC). Phenolic compounds present in crude rapeseed oil have also shown antioxidant properties. Koski et al. (2003) fractionated crude rapeseed oil and found vinylsyringol-containing fraction to be the most effective antioxidant in bulk and emulsified methyl linoleate and lecithin-liposome systems.

Rapeseed phenolic extracts with sinapine as the main phenolic compound showed excellent antioxidant activities toward oxidation of liposomes and low density lipid (LDL) particles. This indicates that sinapine is the major contributor to the antioxidant activity. However, the total phenolic content is not the only thing affecting the antioxidant activity and other compounds also contribute to overall antioxidant activity of rapeseeds (Wanasundara *et al.*, 1996).

Antimicrobial activity: In a test of the antimicrobial activity of rapeseed phenolic fractions Nowak *et al.* (1992) found the fraction of free phenolic acids (FFA) and the sinapic acid (SA) fraction isolated from the ethanolic extract to be highly effective against the growth of gram-negative (*Escherichia coli*, *Enterobacter aerogens*, *Pseudomonas fluorescens*) and gram-positive (*Bacillus subtilis*, *Bacillus cereus*, *Streptococcus lactis*, and *Streptococcus cremoris*) bacteria. The SA fraction found to be more effective then FFA fraction was almost effective. Antimutagenic properties : Kuwahara et al. (2004) tested the antimutagenic properties of canolol (i.e. *vinylsyringol*) isolated from crude rapeseed oil, and found it to have antimutagenic properties when Salmonella typhimurium TA 102 was present. The effects of rapeseed meal extract as the phenolic extract of crude rapeseed oil on the antimutagenicity was tested and it was shown that without metabolic activation all tested samples showed antimutagenic properties to prokaryotic cells. However, *there was no antimutagenic* property against eukaryotic cells (Kuwahara et al., 2004).

Anti-inflammatory properties : Crude rapeseed oil extract with vinylsyringol as the main phenolic compound has anti-inflammatory properties. These results indicate that vinylsyringol was responsible for the anti-inflammatory effect of crude rapeseed oil while sinapic acid in the rapeseed meal had no effect (Karonen *et al.*, 2004). The studies showed that phenolics from crude rapeseed oil had anti-inflammatory properties as they inhibited the formation of NO and PGE<sub>2</sub>. Rapeseed meal phenolics had no anti-inflammatory property which indicates that *vinylsyringol* is responsible for the anti-inflammatory effect.

*Cell permeability*: The crude rapeseed oil phenolic extract had no significant effect on the permeability of the model drugs. Rapeseed meal phenolics enhanced the permeability of verapamil and ketoprofen indicating that they may have an impact on drugs and other components being actively transported across the cell membrane (Satu *et al.*, 2005). Rapeseed meal phenolics enhanced the permeability of verapamil and ketoprofen, while the oil phenolics had no effect.

### **Rapeseed Glucosinolates**

Glucosinolates are amino acid-derived secondary plant metabolites found exclusively in cruciferous plants. Majority of cultivated plants that contain glucosinolates belong to the family of *Brassiceae*. Glucosinolates and their breakdown products are of particular interest in food research because of their nutritive and antinutritional properties, the adverse effects of some glucosinolates on health, their anticarcinogenic properties and because they are responsible for the characteristic ûavour and odour of many vegetables (Mithen et al., 2000). There are currently about 120 different glucosinolates characterized, of which only a limited number have been investigated thoroughly. Glucosinolates themselves are chemically stable and biologically inactive while they remain separated within sub-cellular compartments throughout the plant. However, tissue damage caused by pests, harvesting, food processing or chewing initiates contact with the endogenous enzyme myrosinase, which leads to hydrolysis releasing a broad range of biologically active products such as isothiocyanates (ITCs), organic cyanides, oxazolidinethiones and ionic thiocyanate on enzymatic degradation by myrosinase in the presence of water. The anticarcinogenic mechanisms by which these compounds may act include the induction of detoxiûcation enzymes and the inhibition of the activation of promutagens/ procarcinogens (Dragsted et al., 1993; Jongen, 1996; Mithen et al., 2000).

Anticarcinogenicity : The most extensively studied mechanism for inhibition of carcinogenesis by glucosinolate-derived hydrolysis products are the modulation of the antioxidant potential, enhancement of detoxiûcation mechanisms and the induction of apoptosis in undifferentiated cells (Mithen *et al.*, 2000).

Isothiocyanates that arise in plants as a result of enzymatic cleavage of glucosinolates by the endogenous enzyme myrosinase are attracting increasing attention as chemical and dietary protectors against cancer. Their anticarcinogenic activities have been demonstrated in rodents (mice and rats) with a wide variety of chemical carcinogens (Talalay and Zhang, 1996).

The anticarcinogenic effects of ITCs can be explained by two different mechanisms. The ûrst, a blocking effect, involves induction of Phase II enzymes, including quinone reductase in the small intestinal mucosaandliver (Zhang et al., 1992; Talalay and Zhang, 1996). These enzymes are involved in the detoxiûcation in the body of foreign compounds (xenobiotics). Increased activity will therefore block exposure of target tissues to DNA damage (Fimognari and Hrelia, 2007). The second mechanism, a suppressing effect, involves suppression of tumour development via deletion of damaged cells from colonic mucosal crypts through the induction of programmed cell death (apoptosis). Smith et al. (1996) showed that dietary supplementation with the glucosinolate sinigrin, or its breakdown product allyl isothiocyanate, can protect against chemically-induced colorectal carcinogenesis by stimulation of apoptosis. The evidence for anticarcinogenic effects of Brassica vegetables in humans is strongly supported by evidence obtained from epidemiological and human intervention studies as well as with experimental animals. Verhoeven et al. (1996) reviewed seven cohort studies, 87 case-control studies and showed an inverse correlation between the consumption of individual Brassica and the risk of lung, stomach and second primary cancers.

#### **Rapeseed Dithiolethiones**

Several dithiolethiones are now found to be potent inhibitors of chemical-induced tumours in multiple tissues and have positive chemopreventive efficacy. Oltipraz is the best-characterized member of the dithiolethione class of cancer chemopreventive agents (Wattenberg and Bueding, 1986). Monicah et al. (2000) examined the induction of hepatic antioxidant genes in rats treated with 3H-1, 2-dithiole-3-thione (D3T) and concluded that antioxidant enzyme regulation may be altered during carcinogenesis. Induction of these genes could provide a potentially important mechanism of action of chemoprotective 1, 2-dithiole-3-thiones. D3T is clearly more potent than oltipraz and also serves as a useful molecular probe for determining the key events associated with protection by this class of agents Roebucck et al. (2003).

There are several studies describing the antimutagenic and anticarcinogenic activity of dithiolethiones (Moolky *et al.*, 1992). Dithiolethiones mediate carcinogen detoxification and suppressed the multiplicity of benzo [a] pyrene-induced pulmonary adenomas and fore-stomach tumours (Ansher *et al.*, 1983).

Studies on effect of *Brassica nigra* on benzo [[alpha]] pyrene mutagenicity reported that even 1 per cent of seed extract exhibited strong antimutagenic effect, which remains same with increase in dosages (Polasa *et al.*, 1994). Cui (1997) attributed this research as the effect of presence of sulphur-containing compound as dithiolethiones in rapeseed.

Dithiolethiones constitute an important class of chemopreventive agents that enhance the expression of carcinogen detoxication and antioxidant genes. However, characterization, quantification and specificity of mustard dithiolthiones still require further investigations.

### **Rapeseed Phytosterols**

Phytosterols have been known for more than half a century for their LDL-cholesterol-lowering properties (Best *et al.*, 1954). Apart from being essential membrane constituents, regulating their fluidity and permeability, phytosterols also regulate membrane-bound enzyme activities and signal transduction events (Hartmann, 1998). Various studies showed that phytosterols decreases serum total and LDL cholesterol levels (Gylling *et al.*, 1997; Miettinen, 2001).

One of the main mechanisms for cholesterol reduction is prevention of cholesterol absorption by its replacement with phytosterols in the intestinal-micellar phase (Nissinen *et al.*, 2002; Trautwein *et al.*, 2003). These observations have led to the development of a new type of "phytosterols as functional food", including margarine, milk and yoghurt, enriched with phytosterols as bioactive components. Following consumption, phytosterols reduce the absorption of dietary and endogenous cholesterol by about 50% (Law, 2000).

Phytosterols are an important group of minor constituents of rapeseed. With concentrations ranging from 0.5 to 1% of the crude rapeseed oil, rapeseed is one of the richest natural sources of phytosterols (Piironen *et al.*, 2000). In rapeseed, major phytosterols are sitosterol, campesterol, brassicasterol and avenasterol, while stigmasterol and cholesterol occur only in trace amounts (Appelqvist *et al.*, 1981). Sitosterol is the most prominent phytosterol, accounting for 53% of the total phytosterol content, followed by campesterol.

**Sitosterol :** Alone and in combination with similar phytosterols, -sitosterol reduces blood levels of cholesterol, and is sometimes used in treating hypercholesterolemia. -sitosterol inhibits cholesterol absorption in the intestine (Matsuoka *et al.*, 2008).

**Campesterol :** Campesterol can regulate lipoprotein metabolism in the intestine and regulate biliary secretion (Sudhop *et al.*, 2002; Ho and Pal, 2005).

Stigmasterol: Stigmasterols are chemically similar to animal cholesterol. It is used as the precursor of vitamin D3 (Kametani and Furuyama, 1987). Research has indicated that stigmasterol may be useful in prevention of certain cancers, including ovarian, prostate, breast, and colon cancers (Lee et al., 2009). Stigmasterol lowers plasma cholesterol levels, inhibits intestinal cholesterol, plant sterol absorption, and suppresses hepatic cholesterol, classic bile acid synthesis (Ashok et al., 2006). Studies with laboratory animals-fed stigmasterol found it inhibits several pro-inflammatory and matrix degradation mediators typically involved in osteoarthritis-induced cartilage degradation. These promising results justify further investigations with stigmasterol (Berenbaum, 2009). It also possesses potent antioxidant, hypoglycemic and thyroid inhibiting properties (Gabay et al., 2010).

**Brassicasterol :** Brassicasterol is a 28-carbon sterol synthesised by several unicellular algae (phytoplankton) and some terrestrial plants, e.g., oilseed rape. Rapeseed is found to contain 0.55 to 0.73 g/kg (Piironen *et al.*, 2000). Some studies on

brassinosteroids in inhibition of cell growth in a dose dependent manner in cancer cell lines were done. Flow cytometry analysis showed that brassinosteroids treatment inhibited cell growth in a dose dependent manner in cancer cell lines without affecting the growth of normal cells (Jana, 2008). However brassicasterol is yet to find a place as a nutraceutical and brassicasterol activity in cancer prevention as a pure molecule in nutraceutical formations. At present it has got no attention and much research is needed to justify it as a nutraceutical.

## **Rapeseed Phytate**

Phytate is a food component that is considered an antinutrient by virtue of its ability to chelate divalent minerals and prevent their absorption. But it has also been shown to have anticancer activity (Shamsuddin *et al.*, 1997) and antioxidant activity (Graf, 1990). It forms an iron chelate that suppresses lipid peroxidation by blocking iron-driven hydroxyl radical generation.

Rao *et al.* (1991) tested the ability of (*myo*-inositol hexakisphosphate, InsP6) to protect Sprague-Dawley rats from damage caused by experimentally induced ischemia-reperfusion injury. They found that phytate injected intravenously, at concentrations of 7.5 and 15 mg/100 g BW, prior to cardiac excision and testing, protected the myocardium from damage as indicated by markers of muscle damage, heart function, blood flow, and lipid peroxidation.

Porres *et al.* (1999) tested the effect of intrinsic dietary phytate to protect the liver and colon mucosa of pigs from oxidativestress induced by moderately high intakes of iron. Their results indicated that phytate may be protective for some tissues under the stress induced by high dietary intake of iron but not under conditions in which iron intake is normal. Rimbach and Pallauf (1998) cautioned that the negative effects of significant phytate intake on mineral bioavailability might compromise its positive effect as an antioxidant. They found that dietary phytate, at 0.75 and 1.5%, impaired Mg absorption and decreased Mg concentration in the plasma and femur. The livers of the animals fed higher amounts

of phytate. Chang *et al.*, (1990) stated that phytate may also contribute to antioxidant defense by increasing the activity of key enzymes that detoxify Reactive Oxygen Species (ROS).

The literature suggests that if the essential minerals in a diet are present in adequate concentrations and in reasonable ratios with respect to one another, and to phytate, no reason exists for nutritional concern. However, many populations might ingest inadequate quantities of calcium, iron, and zinc and may be marginally deficient in magnesium (Kelsay, 1987). Still others will strive to increase phytate consumption even as a supplement because of the health benefits (Thompson, 1989). Thus, it will be complicated to sort out in the various subpopulations until we have better measures of mineral status as we currently have for iron.

### **Rapeseed Bioactive Peptides**

Primary structure of natural proteins consists of certain amino acid sequences that have the ability to exert physiological benefits in human beings. The amino acid sequences remain inactive when they are present as part of the continuous primary structure of the parent protein. However, when the parent protein is acted upon by an appropriate enzyme (usually a protease), the peptide is released in intact form and can be found in the digest called a protein hydrolysate. If the hydrolysate has adequate quantities of the active intact peptides, the product can be applied as therapeutic tools in the management of chronic diseases. The most studied bioactive peptides are the antihypertensive peptides that modulate the renin-angiotensin system. Angiotensin converting enzyme (ACE) is one of the key enzymes that operate the blood pressure regulating pathway called the renin-angiotensin system. ACE acts by removing a dipeptide (two amino acids linked together) from the C-terminal of angiotensin I to produce angiotensin II, the latter being a very powerful vasoconstrictor (Skeggs et al., 1956; Yang et al., 1970). In disease conditions or as a result of genetic or environmental factors, the level of ACE is up-regulated and the resultant high levels of angiotensin II produces undesirable rates of blood vessel contraction that

leads to the development of high blood pressure (hypertension). Initial clinical treatment of hypertension involved the use of ACE inhibitors such as captopril, lisinopril, enalaprilat, chemical compounds that were modelled after ACE-inhibitory peptides discovered from snake venom (Ondetti et al., 1971). However, some of the drugs have side effects and precludes their use by some parts of the human population (Seseko and Kaneko, 1985; Israaili and Hall, 1992). Therefore, there has been an increased interest in producing natural peptides that can act as ACE inhibitors for the treatment of hypertension. Though less active (on a weight to weight) basis, naturally occurring hypotensive peptides are believed to be safer and cheaper than synthetic drugs as agents for the treatment of hypertension. It is also important to note that food protein-derived ACE-inhibitory peptides have been tested in vitro and in vivo as antihypertensive agents that can be consumed at higher levels since there is no risk associated with overdose (FitzGerald et al., 2004; Aluko, 2007).

As rapeseed in rich in protein, it could be utilized in isolating the bioactive peptides for developing functional food. Rapeseed meal is a byproduct of the oil removal process, and is comprised of approximately 40% of protein. Ewa *et al.* (2003) investigated rapeseed protein as a source of new peptides having ACE inhibitory activity and possessing the ability to lower blood pressure after oral administration. Besides its ACE inhibitory activity, rapeseed protein hydrolysates when treated with food-grade endoprotease and alcalase, found to results in formation of two fractions rich in HIV-protease inhibitors (Del Mar Yust *et al.*, 2004).

### Conclusion

Scientific studies on medicinal properties have opened some interesting doors for utilization of rapeseed meal. In numerous epidemiological studies, rapeseed meal phytochemicals has been associated with a decreased risk of chronic diseases such as cardiovascular disease, cancer, and asthma. Major limiting factors in utilization of rapeseed meal as functional food include the poor digestibility of protein with unacceptable sensorial characteristics. Rapeseed meal showed significant antioxidant potential due to presence of phenolics, phytates and phytosterols. High phenolic content of meal could play a role developing functional food with enhancing antioxidant, anti-inflammatory, antimicrobial and antimutagenic properties. It is the richest source of phytosterols and is useful in prevention of certain cancers. It also decreases serum and LDL cholesterol levels. Rapeseed dithiolethiones could have a positive chemopreventive efficacy and it is a potent inhibitors of chemical induced tumours in multiple tissues. Glucosinolates and its derived hydrolysis products have proven its anticarcinogenic and chemopreventive activities on the basis of in vitro and in vivo studies, and rapeseed is the riches source of it. Phytates possess anticancer activity and prevents muscle damage, heart function, blood flow and lipid peroxidation. However concern continues about the impact of dietary phytate upon mineral status. Hence, use of phytates as a functional ingredient needs to be avoided. While bioactive peptide [due to its Angiotensin converting enzymes (ACE) inhibitory effect] is the most potent nutraceutical ingredient among studied phytochemicals in developing functional food and could go a long way in enhancing utilization of rapeseed meal as it comprises more than 40% of protein.

## **Future Trusts**

Quantification and methods for isolation of rapeseed nutraceuticals with least effects of their bioavailability should be the principal area of investigation. Additional research is needed to optimize methods of preparation of functional food with least effect of processing on nutraceutical properties. Further, valuation of rapeseed functional foods should be done through extensive clinical investigations.

## References

Aluko, R.E. 2007. Technology for the production and utilization of food protein- derived antihypertensive peptides: a review. *Recent Patents in Biotechnology.* 1:260-267.

- Amarowicz, R., Naczk, M., and Shahidi, F. 2000. Antioxidant activity of various fractions of nontannin phenolics of canola hulls. *Journal of Agriculture and Food Chemistry*. 48: 2755-2759.
- Anonymous, 2000. Health Canada. Consultation Document: Standards of Evidence for Evaluating Foods with Health Claims: A Proposed Framework. June. (www.hcsc.ca/ food-aliment/english/subjects/health\_claims/ standards\_of\_evidence; seen on 05 Dec 2000).
- Anonymous, 2003. Functional food and nutraceutical food groups. Alberta Crop Industry Development Fund, 3: 1-12.
- Anonymous, 2006. New developments in rapeseed mustard. Meeting Report. *Current Science*, **90**: 1173-1174.
- Ansher, S.S., Dolan, P. and Bueding, E. 1983. Chemoprotective effects of two dithiolthiones and of butylhydroxyanisole against carbon tetrachloride and acetaminophen toxicity. *Hepatology*. 3: 932-935.
- Appelqvist, L.A.D., Kornfeldt, A.K. and Wennerholm, J.E. 1981. Sterols and steryl esters in some brassica and Sinapis Seeds. *Phytochemistry*. **20**: 207-210.
- Arvind Kumar., Pankaj Sharma., Lijo Thomas., Abha Agnihotri and S.S. Banga. 2009. Canola cultivation in India: scenario and future strategy. In: proc. 16th Australian Research Assembly on Brassicas. *Ballarat Victoria*: 1-5.
- Ashok K. Battaab, Guorong Xuab, Akira Hondac, Teruo Miyazakic, Gerald Salena. 2006. Stigmasterol reduces plasma cholesterol levels and inhibits hepatic synthesis and intestinal absorption in the rat. *Metabolism Clinical and Experimental* **55** (3): 292-299.
- Barret, J.E., Klopfenstein, C.F., and Leipold, H.W. 1998. Protective effects of cruciferous seed meal and hulls against colon cancer in mice, *Cancer Lett.* 127: 83–88.

- Berenbaum, F., Breton, M., Chevy, F., Gabay, O., Jacques, C., Nourissat, G., Salvat, C., Sánchez, C., Wolf, C. 2009. The plant sterol stigmasterol inhibits several pro-inflammatory and matrix degradation mediators typically involved in osteoarthritis-induced cartilage degradation. *Osteoarthritis Cartilage*. 2009 Sep 15. PMID: 19786147.
- Best, M.M., Duncan, C.H., van Loon, E.J. and Wathens, J.D. 1954. Lowering of serum cholesterol by the administration of plant sterol. *PubMed Circulation*. 10: 201–206.
- Cai, R., Arntfield, S.D. 2001. A rapid highperformance liquid chromatographic method for the determination of sinapine and sinapic acid in canola seed and meal. *Journal of American Oil Chemistry Society*. **78**: 903-910.
- Chang, M., Burgess, J.R., Scholz, R.W. and Reddy, C.C. 1990. The induction of specific rat liver glutathione S-transferase subunits under inadequate selenium nutrition causes an increase in prostaglandin F2 alpha formation. *J. Biol. Chem.* 265: 5418–5423.
- Cui, W. 1997. Mustard: chemistry and potential as a nutraceutical ingredient. *Canadian Chemical News*: 1-12.
- Del Mar Yust, M., Pedroche, J., Megias, C., GironCalle, J., Alaiz, M., Millan, F., and Vioique, J. 2004. Rapeseed protein hydrolysates: A source of HIV protease peptide inhibitors, *Food Chem.*, 87: 387–392.
- Dragsted, L.O., Strube, M. and Larsen, J.C. 1993. Cancer-protective factors in fruits and vegetables: Biochemical and biological background. *Pharmacology and Toxicology*. 72: 116–135.
- Ewa, D. Marczak., Hachiro, Usui., Hiroyuki, Fujita c., Yanjun, Yang., Megumi, Yokoo., Andrzej, W. Lipkowski., Masaaki, Yoshikawa. 2003. New antihypertensive peptides isolated from rapeseed. *Peptides* **24**: 791–798.
- FICCI. 2009. Nutraceutical Critical supplement for building a healthy India. Ernst and Young Report – Federation of Indian Chambers of Commerce and Industry: 3-30.

- Fimognari, C. and Hrelia, P. 2007. Sulforaphane as a promising molecule for fighting cancer. *Mutation Research.* **635**: 90–104.
- Fitz Gerald, R.J., Murray, B.A., Walsh, D.J. 2004. Hypotensive peptides from milk proteins. *Journal of Nutrition*.**134**: 980S-988S.
- Frankel, E.N., and Meyer, A.S. 2000. The problems of using one-dimensional methods to evaluate multifunctional food and biological antioxidants. *Journal of Science of Food and Agriculture*. 80: 1925-1941.
- Gabay, O., Sanchez, C., Salvat, C., Chevy, F., Breton,
  M., Nourissat, G., Wolf, C., Jacques, C.,
  Berenbaum, F. 2010. Stigmasterol: a phytosterol
  with potential anti-osteoarthritic properties. *Am.*J. Clin. Nutr. 18 (1): 106-116.
- Graf, E. and Eaton, J.W. 1990. Antioxidant functions of phytic acid. *Free Radical Biol. Med.* **8**: 61-69.
- Gylling, H., Radhakrishnan, R., and Miettinen, T.A. 1997. Reduction of serum cholesterol in postmenopausal women with previous myocardial infarction and cholesterol malabsorption induced by dietary sitostanol ester margarine. *Circulation*. **96**: 4226-4231.
- Hartmann, M.A. 1998. Plant sterols and membrane environment. *Trends in Plant Science*. **3**: 170-175.
- Hemmingway, J.S. 1993. Mustard and Condiment Products: Encyclopaedia of Food Science and Technology and Nutrition, Academic Press, New York, pp. 3178-3182.
- Hertog, M.G.L., Kromhout, D., Aravanis, C., Blackburn, H., Buzina, R., Fidanza, F., Giampaoli, S., Jansen, A., Menotti, A. and Nedeljkovic, S. 1995. Flavonoid intake and longterm risk of coronary heart disease and cancer in the seven countries study. *Archives of Internal Medicine*. **155**: 381-386.
- Ho, S.S., Pal, S. 2005. Margarine phytosterols decrease the secretion of atherogenic lipoproteins from Hep G2 liver and Caco 2 intestinal cells. *Atherosclerosis*, **182**: 29-36.

- Israaili, Z.H., Hall, W.D. 1992. Cough and angioneurotic edema associated with angiotensin converting enzyme inhibitor therapy: A review of the literature and pathophysiology, *Annu. Int. Med.* **117**: 234-242.
- Jana Malíková, Jana Swaczynová, Zdenik Koláø, Miroslav Strnad. 2008. Anticancer and antiproliferative activity of natural brassinosteroids. *Phytochemistry* 69 (2): 418-426.
- Jongen, W.M.F. 1996. Glucosinolates in brassica: Occurrence and significance as cancermodulating agents. *Proceedings of the Nutrition Society* 55: 433–446.
- Kametani T, Furuyama H. 1987. Synthesis of vitamin D3 and related compounds. *Med Res Rev.* 7 (2): 147-171.
- Karonen, M., Hämäläinen, M., Nieminen, R., Klika, K.D., Loponen, J., Ovcharenko, V.V., Moilanen, E., and Pihlaja, K. 2004. Phenolic extractives from bark of *Pinus sylvestris* L. and their effects on inflammatory mediators nitric oxide and prostaglandin E2. *Journal of Agriculture and Food Chemistry* 52: 7532-7540.
- Kelsay, J. 1987. Effects of fiber, phytic acid, and oxalic acid in the diet on mineral bioavailability, *Amer. J. Gastroenterol.* 82: 983–986.
- Koski, A., Pekkarinen, S., Hopia, A., Wähälä, K., and Heinonen, M. 2003. Processing of rapeseed oil: effects on sinapic acid derivative content and oxidative stability. *European Food Research Technology.* 217: 110-114.
- Kozlowska, H., Naczk, M., Shahidi, F., and Zadernowski, R. 1990. Phenolic acids and tannins in rapeseed and canola. In: *Canola and Rapeseed. Production, Chemistry, Nutrition and Processing Technology.* Shahidi F (Ed.). Van Nostrand Reinhold, USA, pp. 193-210.
- Krygier, K., Sosulski, F. and Hogge, L. 1982. Free, esterified, and insoluble-bound phenolic acids.
  1. Extraction and purification procedure. *Journal of Agriculture and Food Chemistry*.
  30: 330-334.

- Kuwahara, H., Kanazawa, A., Wakamatu, D., Morimura, S., Kida, K., Akaike, T., and Maeda, H. 2004. Antioxidative and antimutagenic activities of 4-vinyl-2,6-dimethoxyphenol (Canolol) isolated from Canola oil. *Journal of Agriculture and Food Chemistry.* 52: 4380-4387.
- Law, M. 2000. Plant sterol and stanol margarines and health. *British Medical Journal*. **320**: 861-864.
- Lee, D.Y., Lee, S.J., Kwak, H.Y., Jung, L., Heo, J., Hong, S., Kim, G.W., Baek, N.I. 2009. Sterols isolated from Nuruk (Rhizopus oryzae KSD-815) inhibit the migration of cancer cells. *J. Microbiol Biotechnol* **19** (**11**): 1328-32.
- Matsuoka, K., Nakazawa, T., Nakamura, A., Honda, C., Endo, K., Tsukada, M. 2008. Study of thermodynamic parameters for solubilization of plant sterol and stanol in bile salt micelles. *Chem. Phys. Lipids* **154**: 87-93.
- Matthäus, B. 2000. Isolation, fractionation and HPLC analysis of neutral phenolic compounds in rapeseeds. *Nahrung* **42**: 75-80.
- Meena, P.D., Awasthi R.P., Chattopadhyay, C., Kolte S.J. and Arvind Kumar. 2010. Alternaria blight: a chronic disease in rapeseed-mustard. *Journal of Oilseed Brassica*. **1** (1): 1-11.
- Miettinen, T.A., Puska, P., Gylling, H., Vanhanen, H., Vartiainen, H. 1995. Reduction of serum cholesterol with sitostanol-ester margarine in a mildly hypercholesterolemic population. *The New England Journal of Medicine* **333**: 1308-1312.
- Mithen, R.F., Dekker, M., Verkerk, R., Rabot, S. and Johnson, I.T. 2000. The nutritional significance, biosynthesis and bioavailability of glucosinolates in human foods. *Journal of the Science of Food and Agriculture*. **80**: 967– 984.
- Monicah, A. Otieno., Thomas, W. Kensler., and Kathryn, Z. Guyton. 2000. Chemoprotective 3H-1,2-dithiole-3-thione induces antioxidant genes in vivo. *Free Radical Biology and Medicine*. 28 (6): 944-952.

- Moolky, N., Darren, L., Peter, J. P. and Dennis B.S. 1992. Anticarcinogenic action of diallyl sulfide in hamster buccal pouch and forestomach. *Cancer Letters.* **66** (**3**): 207-216.
- Naczk, M., Amarowicz, R., Sullivan, A., and Shahidi, F. 1998. Current research developments on polyphenolics of rapeseed/canola: a review. *Food Chemistry*. 62: 489-502.
- Nissinen, M., Gylling, H., Vuoristo, M., and Miettinen, T.A. 2002. Micellar distribution of cholesterol and phytosterols after duodenal plant stanol ester infusion. *American Journal of Physiology - Gastrointestinal and Liver Physiology*. 282: 1009-1015.
- Nowak, H., Kujava, R., Zadernowski, R., Roczniak, B., and Kozlowska, H. 1992. Antioxidative and bactericidal properties of phenolic compounds in rapeseeds. *Fat Science and Technology*. 94: 149-152.
- Ondetti M.A., Williams, N.J., Sabo, E.F., Pluscec, J., Weaver, E.R., and Kocy, O. 1971. Angiotensin converting enzyme inhibitors from the venom of Bothrops- jararaca: isolation, elucidation of structure and synthesis. *Biochemistry* 10: 4033-4039.
- Piironen, V., Lindsay, D.G., Miettinen, T.A., Toivo,
  J. and Lampi, A.M. 2000. Review Plant sterols:
  Biosynthesis, biological function and their importance to human nutrition. *Journal of the Science of Food and Agriculture* 80: 939-966.
- Polasa, J.A., Kumar, P.U., and Krishnaswamy, K. 1994. Effect of *Brassica nigra* on benzo[[Alpha]]pyrene Mutagenicity. *Food Chemistry and Toxicology.* 32: 777-781.
- Porres, J.M., Stahl, C.H., Cheng,W.H., Fu,Y., Roneker, K.R., Pond,W.G., and Lei, X.G. 1999. Dietary intrinsic phytate protects colon from lipid peroxidation in pigs with a moderately high dietary iron intake. *Proc. Soc. Exp. Biol. Med.* 221: 80–86.
- Rao, P.S., Liu, X., Das, D.K., Weinstein, G.S., and Tyras, D.H. 1991. Protection of ischemic heart from reperfusion injury by myo-inositol hexaphosphate, a natural antioxidant. *Ann. Thorac. Surgery.* 52: 908–912.

- Rimbach, G and Pallauf, J. 1998. Phytic acid inhibits free radical formation in vitro but does not affect liver oxidant or antioxidant status in growing rats. *Journal of Nutrition* **128**: 1950–1955.
- Roebuck B.D., Thomas J. Curphey., Yuan, Li., Karen, J.Baumgartner., Sridevi, Bodreddigari., Jian, Yan., Stephen, J. Gange., Thomas, W. Kensler., and Thomas, R. Sutter. 2003. Evaluation of the cancer chemopreventive potency of dithiolethione analogs of Oltipraz. *Carcinogenesis* 24 (12): 1919-1928.
- Satu, Vuorela., Kari, Kreander., Maarit, Karonen,, Riina, Nieminen., Mari, Hämäläinen., Anna, Galkin., Leena, Laitinen., Juha-Pekka, Salminen., Eeva, Moilanen., Kalevi, Pihlaja., Heikki, Vuorela., Pia, Vuorela., and Marina, Heinonen. 2005. Preclinical Evaluation of Rapeseed, Raspberry, and Pine Bark Phenolics for Health Related Effects. Journal of Agricultural and Food Chemistry. 53 (5): 5922–5931.
- Scalbert, A. 1993. Introduction. In: *Polyphenolic phenomena*. Scalbert, A. Ed. INRA Editions, *Versailles Cedex, France*: 5-16.
- Seseko, S., and Kaneko, Y. 1985. Cough associated with the use of captopril. *Arch. Intern. Med.* **145**: 1524-1529.
- Shamsuddin, A.M., Vucenik, I. and Cole, K.E. 1997. IP6: a novel anti-cancer agent. *Life Sci.* **61**: 343–354.
- Skeggs, L.T., Kahn, J.R., and Shumway, N.P. 1956. The Preparation and function of the angiotensinconverting enzyme. *Journal of Experimental Medicine*. 103: 259-299.
- Smith, T.K., Musk, S.R.R. and Johnson, I.T. 1996. Allyl isothiocyanate selectively kills undifferentiated HT 29 cells in vitro and suppresses aberrant crypt foci in the colonic mucosa of rats. *Biochemical Society Transactions.* 24 (3): 381.
- Sudhop T, Sahin Y, Lindenthal B, Hahn C, Luers C, Berthold HK, Von Bergmann K. 2002.
  Comparison of the hepatic clearances of campesterol, sitosterol, and cholesterol in healthy subjects suggests that efflux transporters controlling intestinal sterol absorption also regulate biliary secretion. *Gut.* 51: 860-863.

- Talalay, P. and Zhang, Y. 1996. Chemoprotection against cancer by isothiocyanates and glucosinolates. *Biochemical Society Transactions.* 24: 806–810.
- Thompson, L.U. 1989. Nutritional and physiological effects of phytic acid. In: *Food Proteins*. J.E. Kensella and V.G. Soucie, Eds. Champaign, IL: American Oil Chemists Society, pp. 410–431.
- Trautwein, E.A., Duchateau, G., Lin, Y., Melcnikov, S.M., Molhuizen, H., and Ntanios, F.Y. 2003. Proposed mechanisms of cholesterol-lowering action of plant sterols. *European Journal of Lipid Science and Technology*. **105**: 171-185.
- Verhoeven, D.T.H., Goldbohm, R.A., van Poppel,
  G., Verhagen, H. and van den Brandt, P.A. 1996.
  Epidemiological studies on Brassica vegetables
  and cancer risk. *Cancer Epidemiol Biomarkers and Prevention*, 5: 733–751.
- Wanasundara, U.N., Amarowicz, R., and Shahidi, F. 1996. Partial characterization of natural antioxidants in canola meal. *Food Research International.* 28: 525-530.
- Wanasundara, U.N., and Shahidi, F. 1994. Canola extract as an alternative antioxidant for canola oil. *Journal of American Oil Chemists Society.* 71: 817-822.
- Wattenberg, L.W. and Bueding, E. 1986. Inhibitory effects of 5-(2 pyrazinyl)-4-methyl-1,2-dithiole-3-thione (oltipraz) on carcinogenesis induced by benzo[a]pyrene, diethylnitrosamine and uracil mustard. *Carcinogenesis*. **7**: 1379-1381.
- Yang, H.Y.T., Erdos, E.G., and Levin, Y. 1970. A dipeptidyl carboxypeptidase that converts angiotensin II and inactivates bradykinin. *Biochem. Biophys. Acta* **214**: 374-376.
- Zhang, Y., Talalay, P., Cho, C.G. and Posner, G.H. 1992. A major inducer of anticarcinogenic protective enzymes from broccoli: Isolation and elucidation of structure. *Proceedings of the National Academy of Sciences of the USA*. 89: 2399–2403.