Bio.Clear™ Oestro

Synergistic support for oestrogen metabolism | Suitable for men and women

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Hormonal imbalances can heavily impact women's metabolism, mood, reproduction, stress regulation and weight management. Poor biotransformation of oestrogen has been linked with diseases such as endometriosis, hormone dependant cancers, PCOS, infertility issues to name a few [1–3].

Oestrogen is the name for the collective group of metabolites which play a major function in women's reproductive function, and a minor one in males. There are three major forms of oestrogen – oestrone (E1 or acyclic oestrogen), oestradiol (E2) which is the most active oestrogen and oestriol (E3) which is a weaker form on route to excretion.

Oestrogen production is dependent on a complex negative feedback system in menstruating females. In men and post-menopausal women, it is mainly produced from the aromatisation of androgens. Oestrogen is metabolised through the liver before excretion. There are three different stages to liver detoxification of oestrogen, and every stage can vary slightly, resulting in a myriad of different metabolites that can have slightly differing effects on the body.

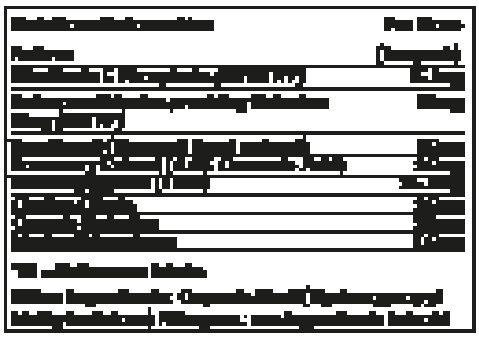
The totality of all the metabolites of oestrogen contribute to the overall oestrogenic load. Dependent on enzymatic production, which can be affected by genes and environmental factors, one could end up with a varying amount of toxic oestrogen metabolites, which may increase the risk of DNA damage and the risks that brings [4].

→ for more information on oestrogen detoxification, go to articles in the education section on Invivo's website.

Supporting oestrogen metabolism

Bio.Clear™ Oestro has been developed to support healthy oestrogen metabolism by using plants and nutrients that encourage the production of the enzymes involved in the oestrogen detoxification process, and by supporting a healthy oestrobolome.

Many of the enzymes required for biotransformation are also used in the clearance of xenoestrogens and other toxins.



Bioactives

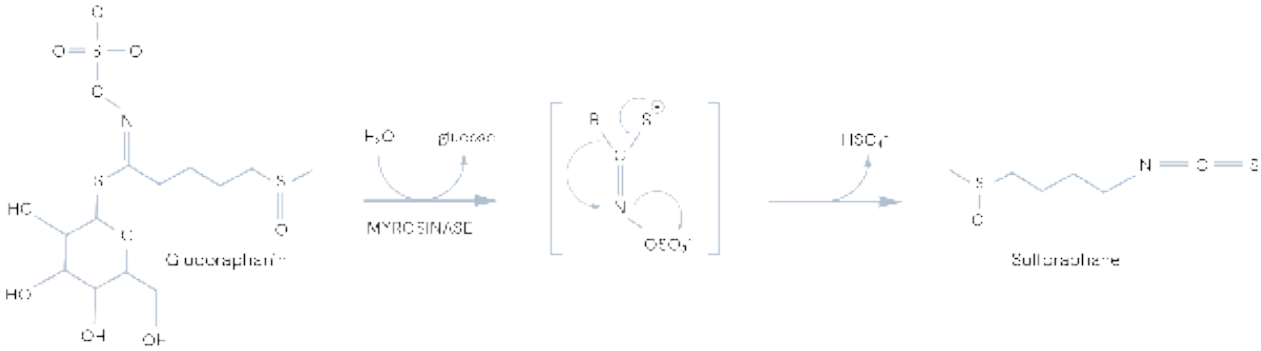
Broccoli seed extract (TrueBroc®)

Cruciferous vegetables, such as broccoli, cabbage, and kale, are rich sources of natural sulphur-containing compounds called glucosinolates. Glucosinolates are hydrolysed to form smaller, active metabolites, which are used locally or absorbed into the blood stream. These metabolites are known collectively as isothiocyanates. One important isothiocyanate is glucoraphanin, which is then metabolised into the well-researched molecule, sulforaphane.

Glucoraphanin and their metabolites are known for their ability to induce xenobiotic-metabolising enzymes that protect DNA from damage [5] and they are frequently being investigated for their antioxidant, chemoprotective, anti-inflammatory, neuroprotective and anti-cancer effects, due to their ability to induce detoxification enzymes [6–8].

The pathway for sulforaphane production is dependent on an enzyme known

as myrosinase, which is stored within plant cells separately from the glucosinolate molecules. When plant cells are damaged by cutting or chewing, myrosinase is released and comes into contact with the glucosinolates, hydrolysing them into their more active versions, such as sulforaphane [9]. Thioglucosidase enzymes in the microbiota of the human colon can also undertake this task, but as the microbiota varies between individuals, so does the production of glucosinates [5,6]. Studies have shown that combining an endogenous source of myrosinase with broccoli seed extract can increase the bioavailability of the sulforaphane production and absorption by 3-4 fold [10].



Conversion of glucoraphanin to sulforaphane (7)

TrueBroc® is a water extract of sprouted broccoli seeds. The seed variety is picked especially for its high glucoraphanin content, which is tested both pre- and post-manufacture. Ingestion of broccoli seed extract can impact the production of enzymes up to 72 hours after ingestion, giving it a long half-life. To ensure that the glucoraphanin content can be metabolised to sulforaphane, we have combined it with **mustard seed myrosinase** to improve the conversion [10].

In the human body, sulforaphane has multiple mechanisms of action. It possesses anti-carcinogenic activity through encouraging changes in the activities of drug-metabolising enzymes, by inducing cell cycle arrest and apoptosis, by preventing inhibition of angiogenesis and metastasis, and by causing changes in histone acetylation status.

It also shows antioxidant, anti-inflammatory and immunomodulatory activities, through induction of different endogenous antioxidant systems [11,12]. It possesses these abilities by increasing the production of an overriding nuclear transcription signalling protein known as nuclear factor (erythroid-derived 2)-like 2, or for short: Nrf2.

Nrf2's role is to signal the cell nucleus to increase the production of intracellular REDOX capacity and phase II detoxification enzymes such as NAD(P)H quinone oxidoreductase 1 (NQO1) and glutathione S-transferases (GST). Isothiocyanates may also induce a NRF2-independent modulation of inflammasomes, reducing inflammation and providing a chemoprotective effect. Sulforaphane inhibits histone deacetylase in cancer cells [13,14].

It is through sulforaphane's ability to induce cytochrome enzymes that it plays an important role in oestrogen metabolism.

Multiple population group studies show a correlation with cruciferous vegetable intake and lower rates of breast and prostatic cancers, amongst others [15]. Oestrogen is known for two mechanisms of cancer promotion: direct action on oestrogen receptors (ER), causing proliferation of cells, and through oxidative catechol metabolites and quinone conjugates, which react with DNA to cause damage [16]. Increased levels of oestrogen-quinone conjugates and DNA adducts have also been detected in urine of women at increased risk for, and with, breast cancer [17]. Oestrogen-quinone conjugates often come through the CYP1B1 pathway, where they end up as 4-hydroxyestrone (4-OH), which is the form more likely to create the quinolated conjugates.

Sulforaphane and broccoli seed extract potently induce quinone reductase activity in cultured prostate cells as well as increasing intracellular glutathione levels, thereby preventing generation of highly reactive semiquinones that can go on to do DNA oxidative damage [18]. Sulforaphane has been shown to induce detoxification enzymes that alter oestrogen metabolism and protect against oestrogen mediated DNA damage in postmenopausal women [19]. Cultures of human cell lines exposed to sulforaphane showed CYP1B1 protein expression was significantly inhibited by approximately 50% with a 2.5-fold significant increase in COMT protein levels. Both of these factors alone would favour the 2-methoxyestrone production over the 4-OH form [17]. Sulforaphane has been shown to reduce the growth of four different breast cancer cell lines in vitro [20].

Actions of sulforaphane: (6)

Direct Actions

Release of Nrf2 from the gatekeeper Keap1/Cul3/Rbx1/E3/Nrf2 complex.
Recruitment of intracellular glucose to the NADPH-producing pentose phosphate pathway.
Increased cellular uptake of glucose.
Attenuation of bacterial lipopolysaccharide-induced expression of:

- Inducible nitric oxide synthase
- Cyclooxygenase-2

Attenuation of bacterial lipopolysaccharide-induced synthesis and secretion of proinflammatory cytokines.
Attenuation of interleukin-1β.
Potentiation of the expression of:

- ADAMTS4 (aggrecanase-1)
- ADAMTS5 (aggrecanase-2)
- MMP1
- MMP13

Actions that may be direct or indirect:

- Attenuation of the cytotoxicity of a number of oxidisers, including:
  - Menadione
  - *tert*-butyl hydroperoxide
  - 4-hydroxynonenal
  - Dexamethasone
  - Peroxynitrile
  - Gentamycin
  - Amyloid-β25-35

Up-regulation of the expression of phase-2 antioxidant enzymes, including:

- Superoxide dismutase
- Catalase
- Quinone reductase
- Thioredoxin reductase 1
- Heme-oxygenase-1
- NADPH: quinine oxidoreductase 1
- Glutathione-*S*-transferase
- Glutathione peroxidase-2

Up-regulation of the expression of the endogenous antioxidant, glutathione.
Down-regulation of the expression phase-1 enzymes, including:

- CYP, family 3, subfamily A, polypeptide 4 (CYP3 A4)
- CYP, family 4, subfamily A isozymes (CYP4 A isozymes)
- Epoxide hydrolase

ADAMTS, A disintegrin and metalloproteinase with thrombospondin motifs; CYP, cytochrome P450; MMP, matrix metalloproteinase; NADPH, nicotinamide adenine dinucleotide phosphate

Sulforaphane has also been shown to improve liver function. In a randomised controlled trial of male subjects with fatty liver, three month supplementation with a broccoli extract (containing 30mg of glucoraphanin) lowered liver function markers of aspartate and alanine aminotransferases and γ-glutamyl transpeptidase, as well as markers of oxidative stress [21].

Broccoli seed extract provides glucoraphanin, which acts through epigenetic modification of numerous pathways involved in detoxification, biotransformation, reduction-oxidation reactions and inflammatory pathways to optimise oestrogen metabolism.

Rosemary extract

Rosemary has been traditionally used for the treatment of a variety of disorders such as memory and cognition, as well as a digestive tonic. It is well-established for its anti-microbial, anti-inflammatory and antioxidant properties. It has been shown to have ant-icancer activity in different in vitro and in vivo models [22]. Rosemary contains a variety of phytochemicals including carnosol, carnosic acid, rosmanol, rosmarinic acid. Carnosic acid is a potent natural antioxidant, often used in the preservation of food materials. Both rosmarinic and carnosic acid give rosemary its strong antioxidant, anti-inflammatory and anti-microbial properties [23,24].

Rosemary has been shown to be an inducer of Nrf2, and it is thought this mechanism gives it anti-proliferative effects on cancer cell lines [25]. It has been shown to induce detoxifying enzyme genes, as well as increase expression of three genes involved in glutathione synthesis (CYP4F3, GCLC) and transport (SLC7A11) as well as increase reduced glutathione (GSH) levels and superoxide dismutase (SOD). In addition, rosemary upregulated anti-inflammatory genes, inhibited COX-2 formation, and decreased IL-6 production [22,26]. The essential oils have been shown to have a hepatoprotective effect [27].

Rosemary reduces the expression of oestrogen receptors (ER) and androgen receptors in prostate cancer cell lines, and reduces the proliferation of ovarian cancer cell lines [26,28–30]. It is also been shown to be a potent anti-microbial, especially to multi drug resistant *E. coli* [31].

Rosemary induces detoxification enzymes and antioxidant enzymes, possesses hepatoprotective effects, down regulates oestrogen receptors and reduces cellular proliferation in hormone-dominant cancers. Its anti-microbial effects also may have an effect on the oestrobolome by preventing the growth of bacteria that produce beta-glucuronidase [31–33].

Methylation support - Betaine, Choline, Riboflavin

The oxidative metabolism of oestrone and oestradiol to the catechol oestrogens (2-OHE2, 4-OHE2, 2-OHE1, and 4-OHE1) and oestrogen quinones has been postulated to be a factor in carcinogenesis of the breast tissue.

The enzyme Catechol-O-methyltransferase (COMT) catalyses the methylation of the catechol oestrogens to methoxy oestrogens, which simultaneously lowers the potential for DNA damage and increases the concentration of 2-methoxyestradiol which is an antiproliferative metabolite.

Some individuals may have epigenetic variants in their enzymes that contribute to inadequate production of methylation co-factors, which may be why some individuals are more prone to the toxic metabolic by-products of oestrogen [17,34].

Betaine (trimethylglycine/TMG) is primarily found in seafood, wheatgerm and spinach. It plays an important role in the liver and kidneys of humans by acting as a methyl donor in the methionine cycle. Inadequate dietary intake of methyl groups can lead to hypomethylation in many important pathways, such as decreasing methionine concentrations and production of S-Adenosyl methionine (SAMe) which is a principle co-factor for many methylation reactions. An insufficiency of methylation can lead to altered fat metabolism in the liver, with resultant hepatic fat accumulation and dyslipidaemia [35].

Dietary intake of choline can modulate methylation via betaine homocysteine methyltransferase (BHMT), this nutrient (and its metabolite, betaine) regulate the concentrations of S-adenosylhomocysteine (SAH) and SAMe, which are the main methyl donors for methylation reactions [36,37].

Low choline concentrations in women with low oestrogen, have been shown to be at a higher risk of fatty liver disease [37] and ample levels of choline are required in pregnancy and lactation to promote healthy neurodevelopment [38].

Choline, betaine and riboflavin help support adequate methylation, even when there are possible polymorphisms on some of the methylation production enzymes.

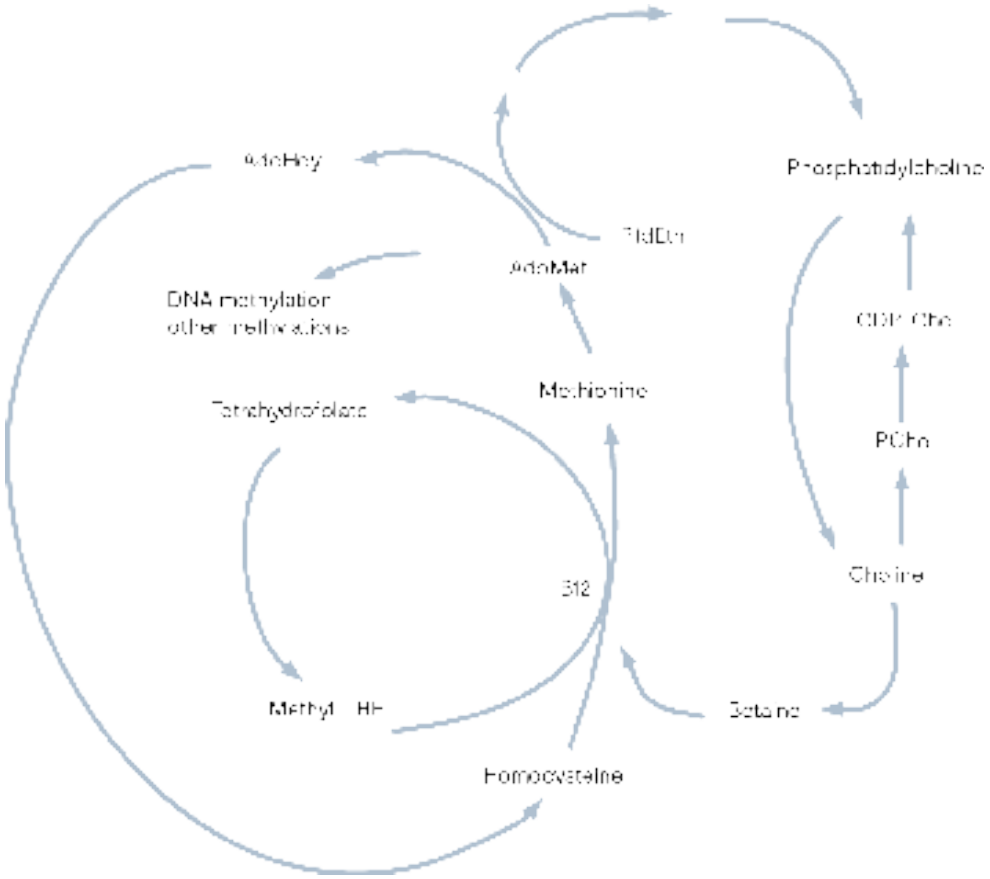


Figure 2 (39): Choline, folate, and methionine metabolism are closely interrelated. CDP-Cho, cytidine diphoshocholine; PCho, phosphocholine; PtdEtn, phosphatidylethanomine; AdoHcy, S-adeno-sylhomocysteine; AdoMet, S-adenosylmethionine; B12, vitamin B

Selenium

Selenium is an essential micronutrient, which plays a major role in twenty-five selenoproteins essential for many antioxidant systems in the body. It is essential for five glutathione peroxidases, three thioredoxin reductases, three iodothyronine deiodinases, and one methionine sulfoxide reductase B1 enzyme system. These enzymes play a large role in detoxification, anti-inflammatory and antioxidant-systems, as well as thyroid metabolism. Low selenium status has been associated with increased risk of mortality, poor immune function, and cognitive decline. Higher selenium status or selenium supplementation has anti-viral effects, is essential for successful male and female reproduction, and reduces the risk of autoimmune thyroid disease [40].

Selenium has been added to this formula to support the function of the enzymes induced by the glucoraphanins.

	Function or health effect	Health effects associated with polymorphisms (or haplotypes) in the selenoprotein*
Glutathione peroxidases (GPxs)	Family of antioxidant enzymes: remove hydrogen peroxide, lipid hydroperoxides, and (GPx4) phospholipid and cholesterol hydroperoxides [4]	--
GPx1 (cytosolic)	Reduces retroviral virulence by preventing viral mutations; [5] deficiency causes cardiomyopathy [5,6]	Cardiometabolic effects: metabolic syndrome, CVD, CAD, blood pressure, restenosis, coronary-artery calcium score, intimamedia thickness, peripheral vascular disease, thoracic aortic aneurysm, intracerebral haemorrhage. Cancer: lung, prostate, bladder, primary liver; Keshan disease, GPx1-198Leu carriers had low blood selenium and low GPx1 activity; Kashin-Beck disease, GPx activity lower in GPx1-198Ley carriers; autism
GPx2 (gastrointestinal)	Antiapoptotic function in colon crypts; helps to maintain intestinal mucosal integrity [7]	--
GPx3 (plasma)	Antioxidant in extracellular fluids; kidney is source of GPx3 in plasma; [4,8] Thyroid protection from from hydrogen peroxide in the thyrocytes and follicular lumen [9]	Ischaemic stroke; differentiated thyroid cancer
GPx4 (phospholipid)	Membrane-associated; present at high concentrations in the testis, where it is essential for sperm mobility and viability [10–12]	Adenomatous polyps, colorectal adenocarcinomas; colorectal cancer; breast cancer survival
Iodothyronine deiodinases	Production of active thyroid hormone T3 and reverse T3(rT3) [13]	--
Dio1 (thyroid, liver, kidney, etc)	Production of T3 in the thyroid and peripheral tissues [13]	Free IGF-1 concentrations, muscle strength, lean body mass
Dio2 (brain, pituitary, muscle, BAT, ear, heart, etc)	T3 production in peripheral tissues [13]	Type-2 diabetes and insulin resistance; osteoarthritis and bone-mineral density; mental retardation (in iodine deficient areas)
Dio3 (cerebral cortex, skin, placenta, pregnant uterus)	Production of rT3; prevents overexposure of foetus to T3 [13]	Osteoarthritis
Selenoprotein P (SEPP1)	Contains 10 selenocysteine residues; major contributor to plasma selenium from the liver via the plasma: brain, testis, and kidney have special receptors; [14] needed for the brain; deficiency causes spasticity, abnormal movements, and spontaneous seizures in mice; [14,15] essential for male fertility; deficiency causes infertility with kinked and hypomotile spermatozoa in mice; [14,16] correlated with fasting plasma glucose; [17,18] may serve as heavy-metal (eg, mercury) chelator [4]	Prostate cancer; affects selenium status (plasma selenium and plasma SEPP) and expression of other selenoproteins; colorectal adenoma, colorectal cancer
Thioredoxin reductases (TrxR)	Redox active with a wide range of substrates, notably thioredoxin, required for DNA synthesis [4]	--
TrxR1 (cytoplasmic/ nuclear)	Controls activity of transcription factors, cell proliferation, apoptosis; reduction of expression leads to slower tumour-cell growth [4]	Advanced colorectal adenoma; familial amyotrophic lateral sclerosis
TrxR2 (mitochondrial)	Indispensable for cardiomyocyte viability [4]	Gastric cancer: a SNP in TrxR2 interacts with GPx1 (Pro/Leu) to affect risk
TrxR3 (testis-specific)	--	--
Selenoprotein S (SEPS1)	Anti-inflammatory, [19] located in the ER; [4] might protect cells from ER stress-induced apoptosis; [4] linked to glucose metabolism and insulin sensitivity [20]	Risk of pre-eclampsia; risk of CHD, ischaemic stroke; W:H ratio, BMI; gastric, colorectal, and rectal cancers
15kDa selenoprotein (SEP15)	Located in the ER; may affect glycoprotein folding [4]	Prostate cancer mortality; lung cancer; rectal cancer
Selenoprotein N (SelN)	Located in the ER; may regulate calcium mobilisation required for early muscle development; mutations cause myopathies including multiminicore disease [4]	--

CVD = cardiovascular disease. CAD = coronary artery disease. IGF-1 = insulin-like growth factor 1. BAT = brown adipose tissue. SNP = single nucleotide polymorphism. ER - endoplasmic reticulum. CHD = coronary heart disease. W:H - waist to hip. BMI = body-mass index. *Appendix pp 6–12 shows table with full list of references.

Table 1: A selection of selenoproteins with known functions relevant to health effects [(40)]. Please see referece 40 for full list of in-table references.

References

1. Rižner TL. Estrogen metabolism and action in endometriosis. Mol Cell Endocrinol. 2009;307(1–2):8–18.

2. Samavat H, Kurzer MS. Estrogen metabolism and breast cancer. Cancer Lett. 2015;356(2):231–43.

3. Thomas MP, Potter BVL, Warwick E, Cassidy A, Hanley B, Jouni ZE, et al. Estrogen metabolism and autoimmunity. J Natl Cancer Inst [Internet]. 2012;307(6):231–43. Available from: http://dx.doi.org/10.1016/j.autrev.2011.11.014

4. Brown KA, Simpson ER. Obesity and breast cancer: The role of dysregulated estrogen metabolism. Obes Breast Cancer Role Dysregulated Estrogen Metab. 2014;1–56.

5. Moreno DA, Carvajal M, López-Berenguer C, García-Viguera C. Chemical and biological characterisation of nutraceutical compounds of broccoli. J Pharm Biomed Anal. 2006;41(5):1508–22.

6. Glade MJ, Meguid MM. A Glance at… Broccoli, glucoraphanin, and sulforaphane. Nutrition.

7. Kensler TW, Chen JG, Egner PA, Fahey JW, Jacobson LP, Stephenson KK, et al. Effects of glucosinolate-rich broccoli sprouts on urinary levels of aflatoxin-DNA adducts and phenanthrene tetraols in a randomized clinical trial in He Zuo Township, Qidong, People's Republic of China. Cancer Epidemiol Biomarkers Prev. 2005;14(11 I):2605–13.

8. Kim JK, Park SU. Current potential health benefits of sulforaphane. EXCLI J. 2016;15:571–7.

9. Tiwari A. Sulforaphane salads. Pharmacogn Mag. 2018;14(53):3.

10. Wehage SL, Wade KL, Stephenson KK, Holtzclaw WD, Talalay P, Fahey JW. Sulforaphane Bioavailability from Glucoraphanin-Rich Broccoli: Control by Active Endogenous Myrosinase. PLoS One. 2015;10(11):e0140963.

11. Dinkova-Kostova AT, Kostov R V. Glucosinolates and isothiocyanates in health and disease. Trends Mol Med [Internet]. 2012;18(6):337–47. Available from: http://dx.doi.org/10.1016/j.molmed.2012.04.003

12. Kaufman-Szymczyk A, Majewski G, Lubecka-Pietruszewska K, Fabianowska-Majewska K. The role of sulforaphane in epigenetic mechanisms, including interdependence between histone modification and DNA methylation. Int J Mol Sci. 2015;16(12):29732–43.

13. Charron CS, Vinyard BT, Ross SA, Seifried HE, Jeffery EH, Novotny JA, et al. Sulphoraphane, a naturally occurring isothiocyanate induces apoptosis in breast cancer cells by targeting heat shock proteins. Eur J Nutr [Internet]. 2012;307(12):571–7. Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC1222573/pdf/11988104.pdf

14. Charron CS, Vinyard BT, Ross SA, Seifried HE, Jeffery EH, Novotny JA. Absorption and metabolism of isothiocyanates formed from broccoli glucosinolates: effects of BMI and daily consumption in a randomised clinical trial. Br J Nutr. 2018;120(12):1370–9.

15. Higdon J V, Delage B, Williams DE, Dashwood RH. NIH Public Access. Pharmacol Res. 2007;55(3):224–36.

16. Cavalieri E, Rogan E. The molecular etiology and prevention of estrogen-initiated cancers. Mol Aspects Med. 2013;36:1–55.

17. Yager JD. Mechanisms of Estrogen Carcinogenesis: The Role of E2/E1- Quinone Metabolites Suggests New Approaches to Preventive Intervention – A Review. Steroids. 2015;99(0 0):56–60.

18. Brooks JD, Paton VG. Potent Induction of Phase 2 Enzymes in Human Prostate. Cancer Epidemiol Biomarkers Prev. 2001;10(September):949–54.

19. Yang L, Zahid M, Liao Y, Rogan E, Cavalieri E, Davidson NE, et al. Reduced formation of depurinating estrogen-DNA adducts by sulforaphane or KEAP1 disruption in human mammary epithelial MCF-10A cells. Carcinogenesis. 2013;34(11):2587–92.

20. Pawlik A, Wiczk A, Kaczyńska A, Antosiewicz J, Herman-Antosiewicz A. Sulforaphane inhibits growth of phenotypically different breast cancer cells. Eur J Nutr. 2013;52(8):1949–58.

21. Kikuchi M, Ushida Y, Shiozawa H, Umeda R, Tsuruya K, Aoki Y, et al. Sulforaphane-rich broccoli sprout extract improves hepatic abnormalities in male subjects. World J Gastroenterol. 2015;21(43):12457–67.

22. Petiwala SM, Johnson JJ. Diterpenes from rosemary (Rosmarinus officinalis): Defining their potential for anti-cancer activity. Cancer Lett [Internet]. 2015;367(2):93–102. Available from: http://dx.doi.org/10.1016/j.canlet.2015.07.005

23. Attokaran M. Rosemary. In: Natural Food Flavors and Colorants. John Wiley & Sons Ltd; 2017. p. 321–4.

24. Erkan N, Ayranci G, Ayranci E. Antioxidant activities of rosemary (Rosmarinus Officinalis L.) extract, blackseed (Nigella sativa L.) essential oil, carnosic acid, rosmarinic acid and sesamol. Food Chem. 2008;110(1):76–82.

25. Pérez-Sánchez A, Sánchez-Marzo N, Herranz-López M, Barrajón-Catalán E, Micol V. Rosemary (Rosmarinus officinalis L) extract increases ROS and modulates Nrf2 pathway in human colon cancer cell lines. Free Radic Biol Med [Internet]. 2017;108:S79. Available from: http://dx.doi.org/10.1016/j.freeradbiomed.2017.04.263

26. Petiwala SM, Berhe S, Li G, Puthenveetil AG, Rahman O, Nonn L, et al. Rosemary (Rosmarinus officinalis) extract modulates CHOP/GADD153 to promote androgen receptor degradation and decreases xenograft tumor growth. PLoS One. 2014;9(3).

27. Rašković A, Milanović I, Pavlović N, Ćebović T, Vukmirović S, Mikov M. Antioxidant activity of rosemary (Rosmarinus officinalis L.) essential oil and its hepatoprotective potential. BMC Complement Altern Med. 2014;14:1–9.

28. Johnson JJ, Syed DN, Suh Y, Heren CR, Saleem M, Siddiqui IA, et al. Disruption of androgen and estrogen receptor activity in prostate cancer by a novel dietary diterpene carnosol: Implications for chemoprevention. Cancer Prev Res. 2010;3(9):1112–23.

29. Lee WH, Chen HM, Yang SF, Liang C, Peng CY, Lin FM, et al. Bacterial alterations in salivary microbiota and their association in oral cancer. Sci Rep [Internet]. 2017;7(1):1–11. Available from: http://dx.doi.org/10.1038/s41598-017-16418-x

30. Tai J, Cheung S, Wu M, Hasman D. Antiproliferation effect of Rosemary (Rosmarinus officinalis) on human ovarian cancer cells in vitro. Phytomedicine [Internet]. 2012;19(5):436–43. Available from: http://dx.doi.org/10.1016/j.phymed.2011.12.012

31. Sienkiewicz M, Łysakowska M, Pastuszka M, Bienias W, Kowalczyk E. The potential of use basil and rosemary essential oils as effective antibacterial agents. Molecules. 2013;18(8):9334–51.

32. Taylor MR, Flannigan KL, Rahim H, Mohamud A, Lewis IA, Hirota SA, et al. Vancomycin Relieves Mycophenolate Mofetil-Induced Gastrointestinal Toxicity by Eliminating Gut Bacterial ß-Glucuronidase Activity. bioRxiv. 2019;1–38.

33. Gonçalves GA, Corrêa RCG, Barros L, Dias MI, Calhelha RC, Correa VG, et al. Effects of in vitro gastrointestinal digestion and colonic fermentation on a rosemary (Rosmarinus officinalis L) extract rich in rosmarinic acid. Food Chem [Internet]. 2019;271(May 2018):393–400. Available from: https://doi.org/10.1016/j.foodchem.2018.07.132

34. Dawling S, Roodi N, Mernaugh RL, Wang X, Parl FF. Catechol-O-methyltransferase (COMT)-mediated metabolism of catechol estrogens: Comparison of wild-type and variant COMT isoforms. Cancer Res. 2001;61(18):6716–22.

35. Stuart AS Craig. Betaine in human nutrition. Am J Clin Nutr. 2004;80:539–49.

36. Zeisel, Steven H. and da Costa K-A. Choline: An Essential Nutrient for Public Health. Nutr Rev. 2009;67(11):615–23.

37. Fischer LM, Costa K, Kwock L, Galanko J, Zeisel SH. Dietary choline requirements of women : effects of estrogen and. Clin Lab. 2010;(C):1–7.

38. Wallace TC, Blusztajn JK, Caudill MA, Klatt KC, Natker E, Zeisel SH, et al. The underconsumed and underappreciated essential nutrient. Nutr Today. 2018;53(6):240–53.

39. Zeisel SH, Blusztajn JK. Choline and Human Nutrition. Annu Rev Nutr. 2003;14(1):269–96.

40. Rayman MP. Selenium and human health. Lancet [Internet]. 2012;379(9822):1256–68. Available from: http://dx.doi.org/10.1016/S0140-6736(11)61452-9