Adjunctive Nutritional Management in Cancer (Research Reviews)

James Meschino DC, MS, ROHP

The information contained in these notes is designed exclusively to accompany Dr. James Meschino's (DC, MS, ROHP) educational program on the Adjunctive Nutritional Management Of Cancer. This information is not intended to replace medical treatment, but rather to provide health care professionals with evidence-based scientific information from which to make clinical decisions, as to the appropriateness of specific dietary and nutritional supplementation decisions regarding patients who have had cancer, are undergoing cancer treatment or who are seeking advice as to proactive practices to reduce risk of cancer development.
In recent years researchers have disclosed vital information regarding the effects of signal transduction and its relationship to cancer at the cellular level and have developed specific biological targeted therapies that affect some of these signal transduction pathways as new methods of treating cancer. Research also reveals that many constituents of food and natural medicines can also affect certain signal transduction pathways that are related to cancer development and therefore, this program will emphasize many of these new developments, which involve the effect of natural medicine, diet and other supplements on signal transduction and cancer.

Course Overview:

1. **Mechanisms Involved In Cancer Development**
2. **Biological Targets For Cancer Prevention and Adjunctive Management**
3. **Natural Agents Shown To Be Useful In Adjunctive Cancer Management and Prevention**
4. **Controversy Pertaining To The Concurrent Use Of Chemotherapy, Radiation With Nutritional Supplements**
5. **Adjunctive Cancer Management Protocols To Consider For Various Types Of Cancer**

Part I Mechanisme Involved in Cancer Development And Biological Targets In Cancer Prevention And Adjunctive Management

Signal Transduction and Cancer

What Is Signal Transduction

Signal transduction refers to any process by which a cell converts one kind of signal or stimulus into another, most often involving ordered sequences of biochemical reactions inside the cell, which are carried out by enzymes and linked through second messengers

resulting in a “second messenger pathway". Such processes are usually rapid, lasting in the order of milliseconds in the case of ion flux, to minutes for the activation of protein and lipid mediated kinase cascades. In many signal transduction processes, the number of proteins and other molecules participating in these events increases as the process eminates from the initial stimulus, resulting in a signal cascade and often results in a relatively small stimulus eliciting a large response.

Signal transduction usually involves the binding of small extracellular signaling molecules to receptors that face outwards from the plasma membrane and trigger events inside the cell. However, steroids (e.g. testosterone, estrogen, cortisone etc) as well as vitamin A and vitamin D represent an example of extracellular signalling molecules that may cross the plasma membrane due to their lipophilic or hydrophobic nature. Many steroids, but not all, have receptors within the cytoplasm and usually act by stimulating the binding of their receptors to the promoter region of steriod responsive genes.

Activation of genes, alterations in metabolism, the continued proliferation and death of the cell and the stimulation or suppression of locomotion, are some of the cellular responses to extracellular stimulation that require signal transduction. Gene activation leads to further cellular effects, since the protein products of many of the responding genes include enzymes and transcription factors themselves. Transcription factors produced as a result of a signal transduction cascade can in turn activate yet more genes. Therefore, an initial stimulus can trigger the expression of an entire cohort of genes, and this in turn can lead to the activation of any number of complex physiological events (Li E et al, 2006)

The total number of scientific papers related to signal transduction published since 1st Jan 1977 up to the 31st December 2007 was 48,377 of which only 11,211 were reviews of other papers.

Epidermal Growth Factor Receptors: Cell Proliferation and Cancer Mediated Via Signal Transduction

Epidermal growth factor receptor (EGFR) consists of a family of four receptors, HER1 (ErbB1), ErbB2 (HER2/neu), ErbB3 (HER3), and ErbB4 (HER4). These receptors exist singularly but form pairs when activated. Ligand pairing (dimerization) activates intracellular tyrosine kinase, which in turn, interacts with the ras protein. Ras protein plays a crucial role in regulating cell growth and has been associated with mutated cellular proliferation. The ras protein then activates the mitogen-activated protein kinase (MAPK). MAPK delivers the signal to the nucleus where it interacts with the cyclin D molecule. MAPK interference results in accumulation of cyclin D. This excess of cyclin D allows the nucleus to override its resting phase and move into uncontrolled growth. As such, over expression of EGFR results in excess accumulation of cyclin D, with a tendency for uncontrolled cellular proliferation.

Investigators have found overexpressed EGFR in a variety of solid tumors. Overexpressed EGFR is found in many human malignancies, including non-small cell lung cancer and breast, head and neck, gastric, colorectal, esophageal, prostate, bladder, renal, pancreatic, and ovarian cancers. Overexpression of EGFR in these cancers is also

associated with poor prognosis, including time to treatment failure and shorter overall and progression-free survival.

Overexpression of EGFR-mediated signaling is also responsible for other key cellular activities associated with malignancy. Although the exact mechanism remains under study, researcher suggests that EGFR-mediated signaling results in reduced apoptosis, the normal process by which dysfunctional cells are destroyed, thereby leading to increased volume of malignant cells.

It is also believed that EGFR-mediated signaling plays a role in angiogenesis, the creation of blood vessels in tumors, by increasing angiogenic factors such as vascular endothelial growth factor. The mechanism remains unknown, but overexpression of EGFR is associated with increased incidence of metastases.

How The EGFR Receptors Work And Effect Downstream Proteins and Cellular Response

At present, ten ligands have been identified to bind HER-1, HER-3 and HER-4:

epidermal growth factor (EGF)
transforming growth factor alpha (TGF-α)
amphiregulin
heparin-binding epidermal growth factor (HB-EGF)
betacellulin
epiregulin
neuregulins 1,2,3 and 4

HER-2 is considered to be an 'orphan' receptor, since no specific direct ligand has been identified as yet, and increasing evidence suggests that it acts mainly as a co-receptor, increasing the affinity of ligand binding to the dimeric receptor complex. Tyrosine kinase (TK) activity, which is required for receptor mediated cellular signaling, is present in all receptors except HER-3. Instead, HER-3 has six intracellular binding sites for phosphatidylinositol 3-kinase (PI3K), which is therefore a potent activator of this enzyme.

The binding of ligands induces dimerization of two identical (homodimer) or different (heterodimer) receptors. The dimerization partner has an important impact on the type and number of downstream effectors activated and also on the downregulation mechanism of the ligand bound receptors. Signaling through HER-2 and -3 requires heterodimerization since, as stated, HER-2 has no known ligand and HER-3 lacks TK activity. Importantly, HER-2 is the preferred dimerization and signaling partner for all other HER receptors. Upon dimerization, intracellular TK domains are phosphorylated, which in turn provide docking sites for several adaptor proteins and signaling enzymes. These proteins link upstream membrane receptor kinases to downstream intracellular protein kinases, which control a wide variety of cellular processes, including:

Cellular proliferation
Apoptosis
Angiogenesis.

Not surprisingly, the involvement of multiple ligands, paired combinations of four receptors and plenty of intracellular downstream effectors (signal processing units) result in extensive diversification of EGFR signals mediated by the EGFR family of receptors.

The EGFR signaling pathway in Breast Cancer

The EGFR family of receptors is involved in the regulation of normal breast development. Preclinical data suggest that all compartments of EGFR signaling network including ligands, receptors and downstream effectors are implicated in the development and progression of breast cancer. Overexpression of HER-2 in transgenic mouse mammary glands has been shown to promote oncogenic transformation and development of malignant phenotype. In line with this data, HER-2 activation has been reported to increase metastatic/invasive potential and to induce progression of cell cycle by disrupting the delicate balance between cyclins and the endogenous CDK inhibitors in BC cell lines.

As such, HER-2 overexpression is associated with aggressive disease biology and reduced survival in breast cancer patients. These preclinical findings are further supported clinically by the success of trastuzumab, a monoclonal antibody against HER-2, which led to improved survival, longer time to progression, higher response rate and longer duration of response in breast cancer patients with HER-2 overexpressing tumors.

Numerous studies have also reported that EGFR (HER-1) overexpression is a poor prognostic factor in BC, often associated with advanced disease. HER-2 is overexpressed in 20–25% of breast tumors and is associated with a poor prognosis (The EGFR expression rate in breast cancer is in the range of 14–91%, depending on the method of assessment, and is almost always caused by increased receptor synthesis). While HER-2 overexpression is shown to be an early event in breast cancer development, HER-1 seems to be involved in later stages. The expression of both receptors is inversely correlated with the ER status of the tumor and HER-1/2 heterodimers have been shown to increase metastatic potential of breast cancer cell lines. Accumulating data suggest that EGFR and HER-2 also predict for a poor response to endocrine therapy, and that the mutual interaction between ER and growth factor pathways seems to hotwire the events inducing endocrine resistance in ER-positive breast tumors. Acquisition of resistance to endocrine therapy was shown to be associated with upregulation of EGFR signaling in breast cancer cell lines. ErbB receptors enhance ER signaling either by directly activating ER or through activation of **MAPK and Akt.** So far, preclinical data have shown that agents blocking HER-1 or HER-2 driven signaling pathways can restore hormone sensitivity in endocrine-resistant, HER-2 overexpressing breast tumors, delay the development of endocrine resistance and enhance the antiproliferative effect of endocrine agents.

These effects are partly mediated by the inhibition of downstream signaling cascades involved in the development of endocrine resistance. The expression of MAPK or Akt has been reported to predict a worse clinical outcome in patients treated with endocrine therapy. Both MAPK and Akt are capable of activating ER in a ligand independent manner, and activated MAPK can also enhance ER signaling by recruiting nuclear receptor co-activators.

The impact of EGFR downstream effectors on cell cycle progression may also indirectly mediate endocrine resistance. For example, Akt and MAPK phosphorylate the cell cycle inhibitor p27, which is required for the G_1 arrest mediated by anti-estrogens. In addition, phosphorylation by MAPK leads to destabilization and proteasome-mediated degradation of p27 in breast cancer cell lines, therefore disturbing its inhibitory effect on cell cycle

progression. Importantly, Akt-induced cytoplasmic sequestration of p27 in human breast cancer specimens has recently been shown to be associated with reduced patient survival.

Endogenous Akt, either constitutively activated via PTEN mutations and overexpression of ErbB receptors or induced by therapeutic agents, has been shown to promote breast cancer cell survival. Induction of Akt activity in response to commonly used therapeutic modalities in breast cancer is considered to be a potential mechanism of therapeutic resistance. In fact, inhibition of Akt by a targeted agent has been shown to potentiate trastuzumab-, doxorubicin-, paclitaxel- and tamoxifen-mediated apoptosis in BC cell lines.

EGFR signaling also seems to be vital for the survival of ER-negative breast cancer. Akt-3 is overexpressed in ER-negative breast cell lines and in tissue samples taken from patients. In addition, EGF-induced activation of PKC (one of the downstream substrates of PI3K) and NF-κB (nuclear factor – kappa beta) has been shown to mediate cellular proliferation in ER-negative BC cells.

Regarding the other receptors of the EGFR family, HER-3, the kinase deficient member, has also been shown to be expressed in breast cancer, while the expression of HER-4 is uncommon in breast carcinomas compared with normal breast epithelium and is associated with favorable prognostic factors. Of note, HER-2/HER-3 heterodimers have the highest mitogenic potential and are constitutively activated in breast cancer cells with HER-2 gene amplification.

Consequently, all these data render the EGFR family of receptors, their ligands and their downstream effectors rational targets for the development of novel antitumor strategies in breast cancer. (To this end natural substances such as curcumin represent interesting prophylactic measures in the prevention of breast cancer, as curcumin has been shown to be a pan Erb inhibitor, in that it induces inhibition of all EGFR receptors (1-4)).

Monoclonal Antibodies And Other New Cancer Treatments Target Signal Transduction Pathways In New Regiment Targeted Biological Therapies Offered By Medicine

Two new drug treatment methods for cancer are being developed, which target the EGFR and associated kinase second messengers involved in uncontrolled cellular proliferation:

1. Monoclonal antibodies are made by fusing the spleen cells from a mouse that has been immunized with the desired antigen with human multiple myeloma cells. One problem in medical applications is that the standard procedure of producing monoclonal antibodies yields mouse antibodies. Although murine antibodies are very similar to human ones there are differences. The human immune system hence recognizes mouse antibodies as foreign, rapidly removing them from circulation and causing systemic inflammatory effects. Monoclonal antibodies have been generated and approved to treat: cancer, cardiovascular disease, inflammatory diseases, macular degeneration, transplant rejection, multiple

sclerosis, and viral infection. In August 2006 the Pharmaceutical Research and Manufacturers of America reported that U.S. companies had 160 different monoclonal antibodies in clinical trials or awaiting approval by the Food and Drug Administration.

Herpectin is a monoclonal antibody drug that targets one of the EGFR's in breast cancer treatment. It may also have application as preventive intervention in women who overexpress the Erb2 receptor, which carries a high risk for future breast cancer development.Monoclonal antibodies target the external ligand-binding domain. The monoclonal antibodies, Herceptin® for the treatment of breast cancer and Gleevec® (imatinib mesylate, STI-571) for the treatment of chronic myeloid leukemia have provided the most prominent initial proof that targeted cancer therapy can translate into improved clinical outcomes.

Trastuzumab represents the only biological targeted therapy for breast cancer in routine clinical practice today. The establishment of the unique role of HER-2 in malignant transformation in preclinical models, coupled with the biological significance of HER-2 overexpression in breast cancer and the preclinical demonstration of the antitumor activity of monoclonal antibodies (mAbs) directed against HER-2, encouraged the development of this drug Other monoclonal antibodies that currently are in development and are directed specifically at EGFR include IMC-225, bevacizumab, and ABX-EGF.

2. The generation of active immune response to HER-2 represents an attractive alternative to passive immunity provided by monoclonal antibodies. Under development are vaccines that, for example, stimulate the patient's own immune system to induce a strong killer cell (CTL) response against HER-2-overexpressing tumor cells.

3. Small molecules inhibit EGFR via the intracellular tyrosine kinase pathway. Some examples of EGFR tyrosine kinase inhibitor agents currently in development are GW572016, OSI-774, and CI-1033. They possess the unique ability to inhibit all members of the EGFR family.

   In studies with a variety of small molecule agents, investigators report rashes, most often maculopapular or acneiform in nature, as the most common adverse effect. In addition, some patients experienced disease responses without evidence of rash. Other small molecule adverse events include nausea, vomiting, diarrhea, and asthenia.

4. Tyrosine kinase inhibitors - Another mechanism by which one can target HER-2 is the inhibition of its TK activity. The safety, tolerability and pharmacokinetics of a new selective HER-2 tyrosine kinase inhibitor (TKI), TAK-165, is currently being evaluated in a multicenter phase I trial in HER-2 expressing breast cancer patients.

5. Curcumin, Vitamin D and emodin Inhibit HER-2 and Tyrosine Kinase and Enhance Effects of Chemotherpy Drugs - Two natural compounds derived from plants, were also shown to possess anti-TK activity in preclinical experiments. Emodin ,a purgative resin, from rhubarb and the buckthorn, inhibits HER-2 TK activity, suppresses the

transformation of HER-2 overexpressing breast cancer and **sensitizes them to chemotherapeutic agents, including cisplatin, doxorubicin, etoposide and paclitaxel**.

Curcumin inhibits HER-2 TK and depletes HER-2/neu protein in HER-2-expressing breast cancer cell lines.

Vitamin D (1,25 dihydroxycholecalciferol) has also been shown to decrease the number of EGF receptors on human breast cancer cells, an effect that partially account for the anti-cancer effects associated with vitamin D.(Koga M Cancer Research May 1988)

6. Heat Shock Protein Inhibitors - Heat shock proteins have emerged as attractive targets for new anticancer drugs because these molecules modulate the signal transduction pathways controlling tumor cell growth and survival. These molecules are not mutated in cancer, but they facilitate malignant transformation by **enhancing** the activity of many oncogenic growth factor receptors, kinases and transcription factors. Heat Shock Protein 90 (HSP90) is required for the refolding of proteins in response to environmental stress and the conformational maturation of several signaling proteins. The function of Heat Shock Proteins can be inhibited by certain compounds, such as 17-AAG (17-allylamino-17-demathoxygeldanamycin), which is currently in phase I clinical trials in advanced cancer patients. HER-2 TK is one of the most sensitive targets for HSP90 inhibitors.

7. Ligand-based strategies - Toxin conjugated ligands are generated by conjugating a potent cellular toxin with one of the ligands of the HER family receptor. For example, the TGF-α–pseudomonas exotoxin A conjugate has been evaluated in a phase I trial. The toxins conjugated to the ligand can destroy the receptor bringing about decreased concentrations of specific receptors on the cell surface and thus, down-regulating signal transduction from this class of receptors.

8. Agents targeting RAS–RAF–MAPK pathway - In approximately 30% of human cancers, mutated Ras genes produce abnormal proteins that remain locked in the activated state, thereby relaying uncontrolled proliferative signals. Ras mutations are not common in BC (<5%). However, the fact that Ras can be activated by multiple stimuli, including EGFR family TKs, renders Ras a promising therapeutic target in breast cancer and other cancers. Only a few agents targeting the Ras–Raf–MAPK pathway are in clinical stages of development in breast cancer, but many of these agents are in the late phases of clinical development in other cancer types

9. Targeting Ras. Farnesyl transferase (FTase) – Activation of this pathway catalyzes a critical step in Ras activation and represents an important target for drug development. However, the initial early clinical studies with farnesyl transferase inhibitors (FTIs) focusing on colorectal cancer (CRC), non small cell lung cancer (NSCLC) and pancreatic cancers, known to have a high incidence of K-ras mutations, did not satisfy the high expectations of therapeutic success.

10. Targeting MEK - CI 1040 (PD 1843220), a small molecule MEK inhibitor, has shown low toxicity and moderate efficacy in phase I trials. Based on this promising phase I data, this agent is moving into phase II clinical trials.

11. Agents targeting PI3K/Akt pathway - Targeting mTOR**.** The mTOR pathway is a key growth factor-mediated signal transduction pathway that regulates cell growth, closely related to the PI3K/Akt pathway. mTOR-dependent growth factor signaling include estrogen, HER-2/neu and IGF-1, all of which can be inhibited by mTOR inhibition, resulting in G_1 arrest of the cell cycle. CCI-779 is an m-TOR inhibitor with activity seen in breast cancer cells, both in vitro and in vivo, and is the only agent of this category in later stages of clinical development in breast cancer. Side-effects were generally mild and the most frequently occurring grade 3–4 toxicities were asthenia, depression, g glutamyl transpeptidase increase, hypercholestrolemia, leucopenia, mucositis and somnolence. Based on this preliminary clinical activity, further evaluation of this agent in combination with other treatment modalities is planned in breast cancer patients

12. Targeting Akt - 17-AAG, an indirect inhibitor of Akt activity, is an ansamycin antibiotic which has shown antitumor effects in HER-2-overexpressing breast cancer cell lines in several preclinical experiments. It inhibits the chaperone function of Heat Shock Protein 90, which in turn depletes several key signaling proteins involved in cell cycle control, hormone signaling and growth factor pathways, including HER-2, Akt and Raf-1. The preliminary findings from phase I trials in advanced cancer patients reported disease stabilization with acceptable toxicity.

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Brief Description And Function Of Receptors and Second Messengers In Signal Transduction Pathways Associated With Cancer

PI3 Kinase (Phosphatidylinositol 3-Kinase)- PI3K is responsible for phosphorylation of the inositol ring to generate a potent second messenger required for survival signaling, and insulin action. PI3 Kinase is also activated by tyrosine kinase, Ras, and by the β:γ subunits of heterotrimeric G-proteins. Akt is the major known effector of the PI3 Kinase pathway. Generation of PIP3 results in the activation of PDK1, which phosphorylates Akt.

Akt- Akt phosphorylates Bad, resulting in protection from apoptosis. Bad, or "Bcl-2". Unphosphorylated Bad dimerizes with Bcl-2 and Bcl-XL, neutralizing their anti-

apoptotic activity. Activation of the PI 3-Kinase pathway leads to activation of Akt which phosphorylates Bad, releasing Bcl-2 and Bcl-XL to block apoptosis.

Apoptosis/survival is regulated by the relative balance of pro-apoptotic (Bax, BclXS, Bak, Bad, Bar) and anti-apoptotic (Bcl2, BclXL, Mcl-1, Dad-1) family members, and their heterodimerization.

Cytochome c - Cytochrome c, released from the mitochondria, along with (apoptosis protease-activating factor 1) APAF1 complex in the cytosol activates caspase 9, as part of the apoptotic pathway.

Apaf -1 - Apaf-1 (apoptosis protease-activating factor 1) binds to cytochrome c and caspase-9 which leads to caspase-9 activation. The release of cytochrome c from the mitochondria provides a mechanism for apoptosis through Apaf-1-induced activation of caspases as well as necrosis due to the cessation of electron chain-transport. It has been proposed that Bcl-2 may interfere with the mitochondria-dependent apoptotic pathways at two points; preventing cytochrome c release from the mitochondria and directly interfering with Apaf-1.

Caspase Enzymes and Apoptosis – Caspase - Cysteine aspartyl proteases related to the C. elegans CED-3 death protein comprise the caspase family. All are expressed as proenzymes which are activated by proteolysis. With respect to their roles in apoptosis, Caspases can be subdivided into initiator (Caspases 8, 9, 10) and effector (Caspases 3, 6, 7) caspases, depending on whether they are activated by receptor clustering (initiator) or by mitochondrial permeability transition (effector).
Effector caspases, most notably Caspase 3, cleave numerous substrates to effect the morphological changes associated with apoptosis..
Mitochondria play an essential role in the apoptotic pathway of many cells by releasing apoptogenic proteins into the cytosol.
Mitochondrial proteins known as SMACs (second mitochondria-derived activator of caspases) are released into the cytosol following an increase in permeability. SMAC binds to inhibitor of apoptosis proteins (IAPs) and inhibits them, preventing the IAPs from arresting the apoptotic process and therefore allowing apoptosis to proceed. IAP also normally suppresses the activity of a group of caspases, which carry out the degradation of the cell, therefore the actual degradation enzymes can be seen to be indirectly regulated by mitochondrial permeability.
Cytochrome c is also released from mitochondria due to increased permeability of the outer mitochondrial membrane, and serves a regulatory function as it precedes morphological change associated with apoptosis. Once cytochrome c is released it binds with Apaf-1 and ATP, which then bind to pro-caspase-9 to create a protein complex known as an apoptosome. The apoptosome cleaves the pro-caspase to its active form of caspase-9, which in turn activates the effector caspase-3, ultimately causing apoptosis to occur.

PARP - Poly(ADP-Ribose) Polymerase 1 (PARP-1) - when activated by breaks in DNA, poly ADP-ribosylates nuclear proteins, result in NAD+ depletion and **cell death by apoptosis**. During apoptosis, caspase 3 cleaves PARP into signature 85 and 24 kDa

fragments. The 24 kDa fragment irreversibly binds to the broken ends of DNA, preventing access of repair enzymes to DNA, and ensuring the irreversibility of apoptosis**. PARP cleavage is commonly used as a marker to "prove" cell death by apoptosis versus necrosis.**

Apoptosis (programmed cell death) - Between 50 billion and 70 billion cells die each day due to apoptosis in the average human adult. For an average child between the ages of 8 to 14, approximately 20 billion to 30 billion cells die a day. In a year, this amounts to the proliferation and subsequent destruction of a mass of cells equal to an individual's body weight. Excessive apoptosis causes hypotrophy, such as in ischemic damage, whereas an insufficient amount results in uncontrolled cell proliferation, such as cancer.

Apoptosis and Cancer:

Two important signal transduction pathways leading to apoptotic mechanisms in mammals include the *TNF*-induced (tumour necrosis factor) model and the Fas-Fas ligand--mediated model, both involving receptors of the TNF receptor (TNFR) family coupled to extrinsic signals.

TNF is a cytokine produced mainly by activated macrophages, and is the major extrinsic mediator of apoptosis. Most cells in the human body have two receptors for TNF: *TNF-R1* and *TNF-R2*. The binding of TNF to *TNF-R1* has been shown to initiate the pathway that leads to caspase activation via the intermediate membrane proteins TNF receptor-associated death domain (TRADD) and Fas-associated death domain protein (FADD). Binding of this receptor can also indirectly lead to the activation of transcription factors involved in cell survival and inflammatory responses.

The Fas receptor (also known as Apo-1 or CD95) binds the Fas ligand (FasL), a transmembrane protein part of the TNF family. The interaction between Fas and FasL results in the formation of the death-inducing signaling complex (DISC), which contains the FADD, caspase-8 and caspase-10. In some types of cells, processed caspase-8 directly activates other members of the caspase family, and triggers the execution of apoptosis. In other types of cells, the Fas-DISC starts a feedback loop that spirals into increasing release of pro-apoptotic factors from mitochondria and the amplified activation of caspase-8.

Following *TNF-R1* and *Fas* activation in mammalian cells a balance between pro-apoptotic (BAX, BID, BAK, or BAD) and anti-apoptotic (Bcl-Xl and Bcl-2) members of the Bcl-2 family is established. This balance is the proportion of pro-apoptotic homodimers that form in the outer-membrane of the mitochondrion. The pro-apoptotic homodimers are required to make the mitochondrial membrane permeable for the release of caspase activators such as cytochrome c and SMAC.

Apoptosis can occur when a cell is damaged beyond repair, infected with a virus, or undergoing stress conditions such as starvation. DNA damage from ionizing radiation or toxic chemicals can also induce apoptosis via the actions of the tumour-suppressing gene p53. The "decision" for apoptosis can come from the cell itself, from the surrounding

tissue, or from a cell that is part of the immune system. In these cases, apoptosis functions to remove the damaged cell, preventing it from sapping further nutrients from the organism, or to prevent the spread of viral infection.

Apoptosis also plays a role in preventing cancer; if a cell is unable to undergo apoptosis, due to mutation or biochemical inhibition, it can continue dividing and develop into a tumour. For example, infection by papillomaviruses causes a viral gene to interfere with the cell's p53 protein, an important member of the apoptotic pathway. This interference in the apoptotic capability of the cell plays a critical role in the development of cervical cancer.

In the adult organism, the number of cells is kept relatively constant through cell death and division. Cells must be replaced when they become diseased or malfunctioning; but proliferation must be compensated by cell death. This balancing process is part of the homeostasis required by living organisms to maintain their internal states within certain limits.

The Immune System and Apoptosis - Cytotoxic T-cells are able to directly induce apoptosis in cells by opening up pores in the target's membrane and releasing chemicals which bypass the normal apoptotic pathway. The pores are created by the action of secreted perforin, and the granules contain granzyme B, a serine protease which activates a variety of caspases which induce apoptosis. Interferon, sometimes used in cancer therapy, is a protein made by immune cells that upregulates the function of cytotoxic T-cells (natural killer cells), helping them stimulate apoptosis of cancer cells.

FADD Death Receptors - FADD (Fas-Associated, Death Domain) binds to the cytoplasmic portion of Fas, and recruits caspase 8 to the Fas receptor. As such, it acts as an adapter protein to transduce apoptotic signals from receptor into the cell.

Fas Death Receptor - Fas is a receptor of the (Tumor Necrosis Factor) TNF Receptor family involved in initiation of apoptosis of lymphoid cells. Fas ligand binds to Fas receptors and causes clustering of Fas molecules. Adapter proteins such as FADD recruit caspase 8 to Fas, which becomes activated and induces the apoptotic cascade.

Tissue Necrosis Factor Receptors - Tissue Necrosis Factor Receptors (TNFRs), which are members of a superfamily of proteins known as Death Receptors, resulting in receptor aggregation and recruitment of adapters (i.e., TRAF [TNF Receptor-Associated Factor] proteins) to the intracellular portion of the complex. TNFR complex formation activates caspase 8, the SAP Kinase pathway (dependent on recruitment of Daxx), and the NFκB pathway (which controls apoptosis versus proliferation).

Transforming Growth Factor – Beta (TGF-β) - TGF-β is a multifunctional peptide that controls proliferation, differentiation, and other functions in many cell types. TGF-β acts synergistically with TGF-α in inducing cellular transformation. It also acts as a negative autocrine growth factor. Specific receptors for TGF-β activation trigger apoptosis when activated. Many cells synthesize TGF-β and almost all of them have specific receptors for this peptide.TGF-β1, TGF-β2, and TGF-β3 all function through the same receptor signaling systems.

TGF beta induces apoptosis in numerous cell types. TGF beta can induce apoptosis in two ways: The SMAD pathway or the DAXX pathway.

The SMAD pathway is the classical signaling pathway that TGF beta family members signal through. In this pathway TGF beta dimers binds to a type II receptor which recruits and phosphorylates a type I receptor. The type I receptor then recruits and phosphorylates a receptor regulated SMAD (R-SMAD). SMAD3, an R-SMAD, is implicated in inducing apoptosis. The R-SMAD then binds to the common SMAD (coSMAD) SMAD4 and forms a heteromeric complex. This complex then enters the cell nucleus where it activates the Mitogen-activated protein kinase 8 pathway. In turn, this triggers apoptosis.

In the DAXX pathway, TGF beta may also trigger apoptosis via the death associated protein 6 (DAXX adapter protein). DAXX has been shown to associate with and bind to the type II TGF beta receptor kinase.

Effects of Insulin - Insulin action stimulates the PI3 Kinase pathway, resulting in Akt activation, which phosphorylates and inactivates Glycogen Synthase Kinase 3 (GSK3). Glycogen Synthase is then rapidly dephosphorylated, and activated. In cancer, the Akt signaling system becomes dysregulated promoting uncontrolled cell growth and inhibition of apoptosis via activation of Bcl-2

Insuling-like Growth Factor - Insulin-like Growth Factor - The IGFs are involved in skeletal growth, and are essential for prevention of apoptosis. Serum levels of free IGFs are kept low by the action of IGF binding proteins (IGFBPs), which sequester the IGFs. Overexpression of IGFBPs may induce apoptosis, presumably by reduction of free IGF. IGFBP levels are also altered in some cancers. The IGF-I Receptor is not as mitogenic as some other growth factor receptors, but its ability to activate the PI3 Kinase pathway, through the Insulin Receptor Substrate (IRS) proteins, is critical for mediating cell survival

p53 gene tumor suppressor gene - The p53 gene is mutated in approximately half of all human cancers. It is involved in the cellular response to cytotoxic stresses, and together with p19ARF, induces expression of p21Cip1, to cause cell cycle arrest.
In addition, p53 is able to induce apoptosis, both by transcriptional and non-transcriptional mechanisms.

Nuclear Factor – kappa beta (NFκB)- NFκB signaling is a critical regulator not only of immune function, but also of proliferation versus apoptosis in response to various stimuli. It is regulated by an inhibitory protein, IκB. Various stimuli lead to activation of IKK (IκB Kinase), which phosphorylates IκB, marking it for ubiquitination and degradation. **Once IκB is degraded, NFκB is able to initiate transcription**. Persistent activation of NF-kappa beta is common feature of cancer cells, helping them escape apoptosis.

Vascular Endothelial Growth Factor (VEGF) - VEGF is a dimeric ligand, and is among the most potent angiogenic mitogens. VEGF is secreted by tumor cells and other cells exposed to hypoxia. The VEGF receptors stimulate the Ras-MAP kinase pathway,

suggesting that it signals as a conventional receptor tyrosine kinase. In turn, vascular endothelial cells proliferate, forming new blood vessels to supply tumors.

Interleukins (ILs)- Interleukins constitute a broad family of cytokines, primarily of hematopoietic cell origin. IL-1, IL-6 and IL-8 are pro-inflammatory cytokines. IL-2 is a T-cell growth factor, while IL-7 is a B-cell growth factor. IL-15 shares many of its properties with IL-2. In fact, IL-2, IL-4, IL-7 and IL-15 signaling involves a common IL-2R- gamma subunit, a common theme in immune cell signaling, where receptors are multi-component, and some components are shared between multiple receptors.

T-cell Receptor (TCR) - The T-cell Receptor is found on the surface of T-cells and is responsible for recognition of foreign antigen. Antigen binding to the TCR results in T-cell activation and proliferation (clonal selection). The TCR initiates a signaling cascade that begins with the CD3 complex and proceeds through Lck to LAT and so on to Ras and then ERK, or through Fyn and ZAP-70 and so on to Rac and then JNK.

FAK (Focal Adhesion Kinase) - FAK is a tyrosine kinase which localizes to focal adhesions. It autophosphorylates on tyrosine during adhesion and spreading, and is thought to be a critical transducer of adhesion-dependent growth and survival. FAK is a substrate of Src, and phosphorylation of FAK may be the mechanism by which Src-transformed cells achieve adhesion-independent growth. FAK is not known to be overexpressed in human cancer, but the activation state of FAK in cancer has not been thoroughly addressed.

Raf – Raf, in addition to its role in mitogenesis, Raf-1 may play a role in regulation of apoptosis and cell cycle progression. Activating mutations of B-Raf that disrupt its auto-inhibition loop have been implicated in a number of cancers, including melanoma and colon cancer. In these cases the raf pathway remains activated prompting increase cellular proliferation and inhibiting apoptosis.

Ras–Raf–MAPK pathway - Ras proteins control a wide variety of cellular processes including growth, differentiation, apoptosis and cytoskeletal organization. Ras proteins are activated in response to a wide variety of intracellular and extracellular stimuli. Activated Ras is rapidly converted to its guanosine diphosphate (GDP)-bound inactive state by hydrolysis of the bound guanosine triphosphate (GTP) by an intrinsic GTPase activity. This mechanism of inactivation is thought to be impaired in mutant Ras, which remains in the activated form and conveys uncontrolled proliferative signals continuously, despite the absence of stimulation. **Ras proteins are mutationally activated in ~30% of human cancers, including breast cancer.**

Ras can be activated by receptor tyrosine kinase. Tyrosine phosphorylated receptors provide binding sites for the adaptor protein Grb2 (growth factor receptor-bound protein 2), which recruits Ras activator protein SOS (son-of-sevenless guanine nucleotide exchange factor).

Upon binding to Grb2, SOS mediates the activation of Ras by facilitating its switch from the inactive GDP-bound from to the GTP-bound active form. Then, GTP-bound Ras can activate several downstream effector pathways.

Raf-1, another protein kinase, is one of these effector molecules. It phosphorylates MEK1 (MAPK/Erk kinase) and MEK2, which in turn phosphorylate the MAPKs, ERK1 and -2 (extracellular signal-regulated kinases). Once activated, Erk1 and -2 translocate into the nucleus, where they phosphorylate a variety of substrates, including nuclear transcription factors, and ultimately lead to transcription of target genes associated with cellular proliferation.

Other substrates for Ras signaling include Rac/Rho (small GTP-binding proteins), PI3K and mitogen-activated protein kinase kinase (MEKK). The Rac–rho pathway is involved in cytoskeletal organization, the PI3K pathway is associated with cell survival, and the MEKK and MAPK pathways mediate cellular proliferation signals. The MAPK pathway is activated preferentially by mitogens, growth factors and tumor promoters, and the other downstream pathways mediated by Ras signaling are stimulated by inflammatory cytokines, hormones and various forms of stress stimuli.

MEKK activates another MAPK family member JNK (Jun N-terminal kinase) via SEK (stress-activated protein kinase), and the activation of this pathway is associated with apoptosis and/or proliferation depending on the dynamic balance between different signal transduction pathways.

JNK activates nuclear transcription factor c-Jun, a major component of activator protein-1 (AP-1). The other component of AP-1, c-fos, is activated by the MAPK pathway mediated through EGFR signaling. The generation of AP-1 is thought to play an important role in the development of endocrine resistance in breast cancer as one of the critical intersection points between estrogen receptor (ER) and EGFR signaling pathways. Many signal transduction pathways impinge on Ras-mediated signaling at different levels, so that the net effect of a given stimulus reflects merely the balance of crosstalk among these pathways.

PI3K–Akt pathway - The PI3K (phoshpatidylinositol phosphate) family of enzymes has been linked to many aspects of malignant transformation, including increased proliferation, growth, motility, invasiveness, metastases, angiogenesis and cell survival and represent a good target for biological therapies (monoclonal antibodies and/or small molecules). PI3K is activated by a diverse set of stimuli including cell surface receptors (G-protein-coupled receptors, receptors with TK activity), tyrosine phosphorylated proteins (e.g. insulin-regulated substrate, IRS) and small G proteins (e.g. Ras). EGFR (ErbB-1), in contrast to other members of the HER family of receptors, induces activation of the PI3K pathway relatively weakly, since this receptor has no intracellular binding sites for PI3K.

Upon activation, PI3K recruit several proteins to the cell membrane such as PKB also known as Akt (protein kinase B, also known as Akt), PDK1 (3-phosphoinositide-dependent protein kinase) and PDK2, small proteins of the Ras family, enabling these signaling proteins to come into close contact with their substrates. PKB/Akt, is thought to be the major player in the PI3K pathway; however, it can be activated by other molecules independent of the PI3K pathway, such as PKA (protein kinase A), the calmodulin-dependent kinase pathway, and cellular stresses like heat shock and hyperosmolarity.

Among three Akt proteins, Akt2 and Akt3 were shown to be activated in human breast cancer samples.

The binding of PKB to PI3K is followed by its phosphorylation and activation by PDKs. Once activated, Akt regulates a number of proteins involved in the regulation of the cell cycle (cyclin D and E2F), apoptosis and cell survival [p21, p27, nuclear factor κB (NF-κB), Forkhead proteins, caspases and BAD]. **Akt also phosphorylates a number of other proteins, the functions of which are closely related to breast cancer, such as BRCA1 and ER. It has been shown that alpha estrogen receptors can bind PI3K and activate PI3K/Akt pathway in an estrogen-independent manner**. This explains, in part, the increase risk of breast cancer associated with hormone replacement therapy and elevated levels of potent estrogens, estradiol and 16-hydroxy estrone.

Many other signal transduction pathways converge on the PI3K pathway at the level of Akt. For example, Ras activates the PI3K pathway, while Akt also regulates the activity of the Ras–Raf–MAPK pathway through phosphorylation and inactivation of Raf.

PI3K signaling can be inhibited by protein phosphatases, the prototype of which is the tumor suppressor gene product PTEN (phosphatase and tensin homolog).

Loss of PTEN activity due to either mutations or gene deletions is associated with malignancies such as Cowden's syndrome. mTOR (mammalian target of rapamycin) kinases are among the downstream targets of Akt, and they are thought to link mitogenic stimulation to protein synthesis and cell cycle regulation. The inhibition of mTOR blocks the progression of the cell cycle at the G_1 phase. Tumor cells without PTEN activity are considered to be more sensitive to mTOR inhibitors due to their increased level of Akt activity.

Oncogenes, Proto-oncogenes and Cancer

Oncogenes and Cancer - An oncogene is a modified gene, or a set of nucleotides that codes for a protein and is believed to cause cancer. Genetic mutations resulting in the activation of oncogenes increase the chance that a normal cell will develop into a tumor cell. Oncogenes are figuratively thought to be in a perpetual tug-of-war with tumor suppressor genes (e.g, p53 and p21 tumor suppressor genes), which act to prevent DNA damage and keep the cell's activities under control. There is much evidence to support the notion that loss of tumor suppressors or gain of oncogenes can lead to cancer. Oncogenes are mutant forms of normal functional genes (called proto-oncogenes) that have a role in normal cell proliferation. Oncogenes are found in tumours and in retroviruses; in the latter case, having been picked up along with retroviral genes during retroviral replication in the host. Once incorporated as part of the retroviral genome, they are activated at inappropriate times and places by retroviral promoters, thereby becoming oncogenes.

Proto-oncogenes - A proto-oncogene is a normal gene that can become an oncogene due to mutations or increased expression. Proto-oncogenes code for proteins that help to regulate cell growth and differentiation. Proto-oncogenes are often involved in signal transductionand execution of mitogenic signals, usually through their protein products. Upon activation, a proto-oncogene (or its product) becomes a tumor inducing agent, an

oncogene. Examples of proto-oncogenes include RAS, WNT, MYC, and ERK. For instance, proteins in the Ras family are very important molecular switches for a wide variety of signal pathways that control such processes as cytoskeletal integrity, proliferation, cell adhesion, apoptosis, and cell migration. Ras and ras-related proteins are often deregulated in cancers, leading to increased invasion and metastasis, and decreased apoptosis. RAS activates a number of pathways but an especially important one seems to be the mitogen-activated protein (MAP) kinases, which themselves transmit signals downstream to other protein kinases and gene regulatory proteins.

The proto-oncogene can become an oncogene by a relatively small modification of its original function. There are three basic activation types:

1. A mutation within a proto-oncogene can cause a change in the protein structure, causing an increase in protein (enzyme) activity or a loss of regulation.
2. An increase in protein concentration, caused by
    a. an increase of protein expression (through misregulation)
    b. an increase of protein stability, prolonging its existence and thus its activity in the cell
    c. a gene duplication (one type of chromosome abnormality), resulting in an increased amount of protein in the cell.
3. Mutations in microRNAs can lead to activation of oncogenes. Research indicates that small RNAs 21-25 nucleotides in length called microRNAs (miRNAs) can control expression of these genes by down-regulating them, a function that is lost with microRNA mutation.

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Tumor Suppressor Genes

Tumor suppressor genes, or more precisely, the proteins for which they code either have a dampening or repressive effect on the regulation of the cell cycle or promote apoptosis, and sometimes do both. The functions of tumor suppressor proteins fall into several categories:

1. Repression of genes that are essential for the continuing of the cell cycle. If these genes are not expressed, the cell cycle will not continue, effectively inhibiting cell division.
2. Coupling the cell cycle to DNA damage. As long as there is damaged DNA in the cell, it should not divide. If the damage can be repaired, the cell cycle can continue.
3. If the damage can not be repaired, the cell should initiate apoptosis, or programmed cell death, to remove the threat it poses for the greater good of the organism.

4. Some proteins involved in cell adhesion prevent tumor cells from dispersing, block loss of contact inhibition, and inhibit metastasis.

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Some Common Abbreviations Used In Signal Transduction And Cancer

The abbreviations used are: PG, prostaglandin; COX, cyclooxygenase; LO, lipoxygenase; LT, leukotriene; HETE, hydroxyeicosatetraenoic acid; PKC, protein kinase C; EGF, epidermal growth factor; FGF, fibroblast growth factor; PDGF, platelet-derived growth factor; IL, interleukin; PLA_2, phospholipase A_2; FLAP, 5-LO activating protein; PMN, polymorphonuclear neutrophil; HODE, hydroxyoctadecadienoic acid; GRP, gastrin-releasing peptide; IGF, insulin-like growth factor; NDGA, nordihydroguaiaretic acid; BHPP, *N*-benzyl-*N*-hydroxy-5-phenylpentanamide; TGF, transforming growth factor; MNU, *N*-nitrosomethylurea; DMBA, 7,12-dimethylbenz(*a*)anthracene.

Part II Evidence-Based Nutrients Of Importance To Cancer Prevention And Adjunctive Management

1. **Curcumin**
2. **Chinese Scullcap**
3. **Essential Fatty Acids**
4. **Reishi Mushroom Extract**
5. **Astragalus**
6. **Vitamin E Succinate**
7. **Selenium**
8. **Vitamin C**
9. **Vitamin D**
10. **Beta-Carotene**
11. **Parthenolide**
12. **Flaxseed Lignans**
13. **Isoflavones**
14. **Lycopene**

15. **Induribin**
16. **Indole-3 Carbinol and Cruciferous Vegetables**
17. **Other Natural Health Products That Inhibit Angiogenesis Of Cancer Cells**

1. Curcumin

Curcumin And Cancer

Several lipoxygenase inhibitors are known, including plant and mammalian lipoxygenases, but only a few of them are known inhibitors of the 12-lipoxygenase enzyme pathway, which is often involved in cancer development and progression. Curcumin is one of these lipoxygenase inhibitors. (1)

Breast Cancer - Curcumin has also been shown to inhibit the tyrosine kinase activity of ErbB-2 and induces apoptosis in mammary carcinoma cells. Curcumin is also reported to dissociate heat shock GRP94 from ERbB-2, another mechanism through which it may reduce breast cancer in cells prone to malignant activity. **(2)**

Colon Cancer - It has been established that over-expression of Epidermal Growth Factor Receptors (EGFR) and other members of the tyrosine kinase family are frequently indicated in epithelial cancers, including colon cancer.
The epidermal growth factor (EGF) family of receptor tyrosine kinases consists of four receptors, EGF-R (ErbB1), ErbB2 (Neu), ErbB3, and ErbB4.
In response to these discoveries pharmaceutical companies have produced a number of drugs that inhibit the activation of specific receptors of the EGFR series (EGFR inhibitor drugs). However, in general these drugs have had limited success because cancer cells usually possess more than one type of EGFR receptor. As such, researchers conclude that what is needed to help prevent colon cancer, as well to help treat colon cancer, is a broad-spectrum EGFR receptor inhibitor that inhibits signal transduction for all EGFR cell membrane receptors. (pan-erb signal transduction inhibitors). To this end there is a naturally-occurring pan-erb signal transduction inhibitor that is showing promise in experimental and animal studies, known as EGFR Related Protein. This protein occurs naturally and thus, its use as targeted therapeutic agent is unlikely to produce toxic side effects. Curcumin, the active ingredient in the spice turmeric, also acts as a powerful inhibitor of EGFR receptors. Experimental studies, animal studies and a recent Phase I clinical trial, have shown that curcumin inhibits the growth of colon cancer cells and reduces tumor incidence in high risk human subjects. Curcumin inhibits the EGFR receptor, which in turn inhibits the propagation of metabolic reactions (e.g. decreased synthesis of the tumor promoting messenger NF-kB) leading to inhibition of cell replication of cancer cells and preneoplastic cells.
Curcumin also exerts anti-inflammatory effects on cells by inhibiting synthesis of pro-inflammatory eicosanoids as noted previously.
Researchers examined whether curcumin together with ERRP will cause a greater inhibition of growth of colon cancer cells than either agent alone and the mechanisms of

this inhibition. Results showned that the cell growth inhibition, and stimulation of apoptosis, in response to the combinatorial treatment was significantly greater than that caused by either agent alone.
These changes were associated with decreased activation **(tyrosine phosphorylation**) of **EGFR, ErbB-2, ErbB-3, and/or IGF-1R.** Whereas curcumin inhibited constitutive activation of both EGFR and IGF-1R, ERRP decreased activation of EGFR, ErbB-2, and ErbB-3 but had no effect on IGF-1R. Further, the combination therapy caused a greater attenuation of downstream effectors such as **NF-κB, Akt and BAD** activation, and down-regulation of **procaspase-3** than that noted with either agent alone. The superior effects of the combinatorial treatment could partly be attributed to inhibition of constitutive activation of EGFRs and IGF-1R signaling pathways **(3)**

Familial adenomatous polyposis (FAP) - FAP is an autosomal-dominant disorder characterized by the development of hundreds of colorectal adenomas and eventual colorectal cancer. Regression of adenomas in this syndrome occurs with treatment of anti-inflammatory drugs and cyclo-oxygenase inhibitors. However, these compounds can have considerable side effects. In a 2006 study researchers alternatively used 480 mg of curcumin and 20 mg of quercetin, three times per day in five patients with FAP, who had undergone previous colectomy (4 with retained rectum and 1 an anal pouch). After 6 months all five patients had decreased polyp number and size from baseline assessment, with the mean percentage decrease in number and size from baseline measurements of 60.4% and 50.9%, respectively. No serious or significant side effects were reported. As the dosage of quercetin was not much higher than that which is attainable from daily food sources, researchers suggest that growth retardation effects shown in this study were primarily due to curcumin supplementation **(4)**

Prostate Cancer – Curcumin has been shown to decrease the proliferative potential and induce the apoptosis potential of both androgen-dependent and androgen-independent prostate cancer cells in vitro, largely by modulating the apoptosis suppressor proteins and by interfering with the growth factor receptor signaling pathways as exemplified by the EGF-receptor. Further observations showed that in vitro treatment with curcumin, caused a marked decrease in the extent of cell proliferation the growth of LNCaP cells as implanted tumors in nude mice was followed. **(5)**

There is also increasing evidence that the selective pressure imposed by androgen ablation therapy (via castration or anti-androgen drugs) on the residual prostate cancer cells may actually accelerate the development of the hormone refractory and bone metastatic phenotype. The propensity of prostate cancer to establish osseous metastases is very likely mediated by the osteomimetic properties of the prostate cancer cells. Prostate cancer cells acquire these "bone-like" properties in order to survive in the bony microenvironment. This process is facilitated by common growth factor trophisms between the bone stromal cells, osteoblasts, and the prostate cancer cells wherein a number of growth factors and their receptors are involved.
As such, a general inhibition of the tyrosine kinase signaling pathways may have a therapeutic advantage in interfering with the metastatic potential of these prostate cancer cells. A study conducted with curcumin on bone like properties of C4-2B, a highly metastatic derivative of LNCaP prostate cancer cell line showed that curcumin inhibited the ligand-stimulated autophosphorylation of EGF-R and CSF1-R that were crucially

involved in the development of osteomimetic properties of C4-2B cells. When C4-2B cells were grown under promineralization conditions, curcumin prevented the formation of the mineralized nodules. It also inhibited the expression of the core-binding factor a-1 in C4-2B cells which was responsible for the expression of several bone-specific proteins. The IKK activity was severely impaired, showing marked NF-kappa B inhibition. The experiments indicate that curcumin can also interfere with the development of the osteoblast and the osteoclast-like properties by these prostate cancer cells, thus exhibiting a potential to prevent the establishment of bony metastases. **(6)**

Curcumin and Angionesis – Curcumin has also beeen shown to inhibit angiogenesis in cancer cells via a number of pathways (see Angiogenesis)

Curcumin Can Aid Chemotherapy and Radiation Treatment - Curcumin also interacts with cancer cells at a number of levels and can enhance the tumoricidal efficacy of cytotoxic chemotherapy and radiotherapy. Its anti-invasive effects are partly mediated by downregulation of matrix metalloproteinase-2 (*MMP2*) and upregulation of tissue inhibitor of metalloproteinase-1 (*TIMP1*). These enzymes are involved in the regulation of tumour cell invasion. **(7)**

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Curcumin and Other Anti-Inflammatory Herbs In Cancer Prevention and Adjunctive Management

James Meschino D.C., M.S., N.D.

Evidence suggests that 70-90% of colon cancer cases are caused by faulty nutrition and other factors associated with lifestyle. Colon cancer continues to be the second leading cause of cancer death in North America with about 140,000 new cases diagnosed each year in the United States. Despite recent advances in medicine, mortality remains unacceptably high. It is noteworthy however, that periodic colonoscopy assessment after age 50 years of age (or earlier in individuals with a strong family history) is associated with a 15-33% reduction in risk of colon cancer death. Thus, early detection of colon cancer via colonoscopy screening in high-risk individuals and those over 50 years of age is a prudent primary prevention step.

With respect to preventing colon cancer development dietary modifications linked to reduced colon cancer incidence include consuming a low animal fat diet with minimum intake of trans and hydrogenated fats, consuming more fiber, fruits and vegetables, especially cruciferous vegetables, as well as allium-containing foods (garlic and onions) and omega-3 fats, minimizing alcohol consumption and attaining optimal levels of folic acid, calcium, vitamin D, selenium, antioxidants and several other micronutrients. It is also best to minimize intake of barbequed foods (the charring of such foods results in the formation of highly carcinogenic chemical compounds), and sodium nitrite found in many processed foods.

Cell Membrane Receptors and Cancer

In recent years cancer researchers have discovered that an important part of the puzzle in colon cancer development lies in the expression of certain cell membrane receptors and their stimulation by various chemical agents (ligands). Stimulation of various cell membrane receptors by specific ligands produce profound effects on cellular proliferation, cell growth, cell differentiation and apoptosis (programmed cell death). As an example, the binding of vitamin D to the vitamin D receptor on the cell membrane, triggers a series of reactions (known as signal transduction) that ultimately promotes the induction of intracellular messengers, which slow the rate of cell division, and promotes cell maturation; two outcomes linked to reduction of cancer development.

On the other hand the over-expression of Epidermal Growth Factor Receptors (EGFR) and other members of the tyrosine kinase family are frequently indicated in epithelial cancers, including colon cancer.

The epidermal growth factor (EGF) family of receptor tyrosine kinases consists of four receptors, EGF-R (ErbB1), ErbB2 (Neu), ErbB3, and ErbB4.
In response to these discoveries pharmaceutical companies have produced a number of drugs that inhibit the activation of specific receptors of the EGFR series (EGFR inhibitor drugs). However, in general these drugs have had limited success because cancer cells usually possess more than one type of EGFR receptor. As such, researchers conclude that what is needed to help prevent colon cancer, as well to help treat colon cancer, is a broad-spectrum EGFR receptor inhibitor that inhibits signal transduction for all EGFR cell membrane receptors. (pan-erb signal transduction inhibitors). To this end there is a naturally-occurring pan-erb signal transduction inhibitor that is showing promise in experimental and animal studies, known as EGFR Related Protein. This protein occurs naturally and thus, its use as targeted therapeutic agent is unlikely to produce toxic side effects.

Curcumin Is A Natural Pan-erb Signal Transduction Inhibitor In Cancer Prevention

With respect to natural medicine, it is well documented that curcumin, the active ingredient in the spice turmeric, also acts as a powerful inhibitor of EGFR receptors. Experimental studies, animal studies and a recent Phase I clinical trial, have shown that curcumin inhibits the growth of colon cancer cells and reduces tumor incidence in high risk human subjects. Curcumin inhibits the EGFR receptor, which in turn inhibits the propagation of metabolic reactions (e.g. decreased synthesis of the tumor promoting messenger NF-kB) leading to inhibition of cell replication of cancer cells and preneoplastic cells.

Curcumin also exerts anti-inflammatory effects on cells by inhibiting synthesis of pro-inflammatory prostaglandins. Inhibiting pro-inflammatory prostaglandins has also been shown to reduce cancer risk of colon cancer as demonstrated by studies linking the effects of aspirin and other non-steroidal anti-inflammatory drugs to reduced incidence of colon cancer. However, unlike aspirin, curcumin does not cause gastrointestinal erosion leading to ulceration and bleeding disorders.
As such, daily supplementation with curcumin may be viewed as part of a chemoprevention strategy to help reduce risk of colon cancer development.

Prostaglandin series-2 and it is metabolites have been shown to contribute to the cancer processes through one or more of several mechanisms including increased proliferation, apoptosis, enhanced carcinogen metabolism or modulation of the immune system
Along with curcumin, other anti-inflammatory herbs that block the activity of cyclooxygenase and/or lipoxygenase (6 and/or 12 lipoxygenase), inhibiting synthesis of prostaglandin series-2 and its metabolites (related eicosanoids of the PGE-2 series) have also shown promise as agents to reduce risk of cancer, including colon cancer. The herbs of most importance in my view include white willow extract, ginger and boswellia.

Many holistic health practitioners have recognized the value of these herbs as non-toxic, anti-inflammatory supplements that are useful in the

management of inflammatory musculoskeletal conditions (e.g. arthritis, tendonitis, fascitis, bursitis). However, like curcumin, each of these herbs also demonstrates anti-cancer properties as well.

The main ingredients in ginger that have an anti-inflammatory effect, as well as antitumor and antiproliferative properties against tumor cells, are 6-gingerol and 6-paradol, which are found in the oleoresin fraction in ginger. Other constituents of ginger, 8-paradol and 8-shogaol, demonstrate a significant inhibitory effect on the cyclooxygenase enzyme system, which in turn, reduces the synthesis of prostaglandin series-2 metabolites. As such, ginger supplements should be standardized to contain 5% gingerols.

Gummy exudates of the herb boswellia have been traditionally used as anti-arthritic and anti-cancer mediations. Boswellic acid and its acetates isolated from these gummy exudates were found to be inhibitors of topoisomerases and to be non-redox, non-competitive specific inhibitors of 5-lipoxygenase (5-LOX). All of these properties are key factors in preventing and controlling cancer. Experimental evidence has shown that boswellic acid acetates isolated from *Boswellia carterri Birdw* inhibit cell growth and induce apoptosis (programmed cell death) in prostate cancer cells by inhibition of 5-lipoxygenase. Other studies have shown that boswellic constituents exhibited potent cytotoxic activities against three types of human neuroblastoma cells.

White willow bark contains salicin. The role of salicylates in inflammation and pain management is well documented in medicine. Salicin is a potent inhibitor of cyclooxygenase and thus reduces synthesis of prostaglanding series-2 and its metabolites. Ingestion of acetylsalicylic acid (aspirin) is highly associated with reduced risk of colon cancer in human epidemiological studies. However, unlike aspirin, salicylic acid from white willow bark does not cause intestinal erosion or bleeding and does not impair platelet coagulation to an appreciable degree. As such, white willow bark extract has a much higher safety profile than synthetic aspirin. To be effective white willow bark extract should be standardized to contain 15% salicin.

Summary

A strong body of evidence indicates that an important aspect of cancer prevention involves containment of prostaglandin series-2 synthesis and inhibition of cell membrane receptors associated with the receptor tyrosine kinase family (EGFR, ErB-2, ErB-3 and ErB-4). Curcumin, derived from the spice turmeric, has shown significant anti-tumor properties against colon cancer. Curcumin has been shown to inhibit the receptor tyrosine kinase family and decreases synthesis of prostaglandin series-2. The anti-inflammatory herbs ginger, boswellia and white willow bark extract have also been shown to inhibit prostaglandin series-2 synthesis, as well as other pro-inflammatory mediators, and

experimental evidence suggests that their active constituents possess important anti-tumor properties. As such, health practitioners may wish to encourage their patients to ingest a herbal combination supplement product each day containing curcumin, ginger, white willow bark extract and boswellia, as an additional part of a wellness and cancer prevention program. This may have important applications especially in regards to colon cancer, the second leading cause of cancer death.

A suggested combination formula would include taking two capsules per day of the following supplement, to derive sufficient dosages of the active ingredients in these herbs for chemoprevention purposes. Slightly higher dosages would be required to manage moderate to severe inflammatory conditions. I now take this combination myself as part of my cancer prevention campaign.

Curcumin	200 mg
Boswellia (std to 70% boswellic acids)	200 mg
White Willow Extract (std to 15% salicin content)	33 mg
Ginger Root Extract (std to 5% gingerols content)	50 mg

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2. Chinese Scullcap – Baicalein

Baicalein: Anti-Cancer Effects

Deregulation in the fine balances controlling cellular proliferation and cell death is the hallmark of cancer. Many proteins are involved in the process by which cells choose between growth arrest, apoptosis, or survival. The 12-LOX inhibitor, Baicalein, has been shown previously to induce apoptosis in human gastric, colon, hepatoma, and pancreatic cancer cells Platelet-type 12-Lipoxygenase (12-LOX) has been shown to regulate growth, metastasis, and angiogenesis of prostate cancer. Several lines of evidence implicate 12-LOX as a regulator of human cancer development. It is overexpressed in a variety of tumors including breast, colorectal, and prostate cancer, and has been shown to be present in a number of cancer cell lines.

Prostate Cancer

Inhibition Of 12 Lipoxygenase Pathway - The study by Graham P et al (2002) showed that treatment with Baicalein or BHPP resulted in a dose-dependent decrease in cell proliferation in two human prostate cancer cell lines. This growth arrest was due to cell cycle inhibition at G_0/G_1, and was associated with suppression of cyclin D1 and D3 protein levels. PC3 cancer cells also showed a strong decrease in phosphorylated retinoblastoma (pRB) protein. Cyclin D1 is a proto-oncogenic regulator of the G_1-S phase checkpoint that has been implicated in the pathogenesis of several cancers. Cyclin D1 functions upstream of the RB protein by binding to CDK-4 or -6 leading to RB phosphorylation. The phosphorylation of RB in mid-to-late G_1 releases the transcription factors bound by RB, resulting in their subsequent binding to the promoter regions of various genes leading to DNA synthesis. Within 14 h of treatment with either 12-LOX inhibitor, levels of phosphorylated RB were undetectable in PC3 cells. These results indicate the mechanism of G_1 growth arrest induced by 12-LOX inhibition in PC3 cells: reduction in D-type cyclins, which then result in decreased phosphorylation of RB, resulting in the RB protein remaining bound to the transcription factors required for DNA synthesis. The effects of Baicalein treatment on cell cycle proteins were shown to be through 12-LOX inhibition, as addition of 100 ng/ml of the end product 12(S)-HETE partially restored phosphorylated RB levels. In contrast, addition of 5(S)-HETE or 15(S)-HETE did not restore phosphorylated RB levels after Baicalein treatment. As well, treatment of PC3 cells with 12(S)-HETE alone resulted in increased expression of phosphorylated RB, an effect that was not observed when cells were treated with similar amounts of 5(S)- or 15(S)-HETE. As such, the 12 LO pathway appears to be of great importance in prostate cancer development and growth. Therefore, inhibition of 12-LOX is a potential therapeutic agent in the treatment of prostate cancer.

Apoptosis – Evidence suggests that the 12 Lipoxygenase inhibition produced by Baicalein (and other agents) also indirectly over rides the anti-apoptotic behavior of cancer cells. Treatment with either Baicalein or BHPP has also been shown to result in significant apoptosis in both cell lines. The mechanisms involved were decreased phosphorylation of Akt, loss of survivin and subsequent activation of caspase-3 and caspase-7 in each cell line, decreased Bcl-2 and Bcl-X_L expression in DU-145, and a shift in Bcl-2/Bax levels favoring apoptosis in PC-3 cells. These results show that the 12-LOX pathway is a critical regulator of prostate cancer progression and apoptosis, by affecting various proteins regulating these processes.

Studies have examined the mechanisms underlying the induction of apoptosis after 12-LOX inhibition. Results show that the Bcl family of proteins are involved in

apoptosis induced by LOX inhibition in rat Walker-256 carcinoma cells. In that study LOX inhibition resulted in a down-regulation of the antiapoptotic Bcl-2 protein and a dramatic decrease in the Bcl-2:Bax ratio, which could be blocked by Bcl-2 overexpression. Other studies found a similar decrease in Bcl-2 levels in both cell lines after 12-LOX inhibition, and this reduction was coupled to an increased expression of the proapoptotic protein Bax in the PC3 cells. DU-145 cells, on the other hand, did not express Bax; however, treatment with Baicalein resulted in decreased expression of another antiapoptotic protein of the same family, Bcl-X_L, whereas levels of its proapoptotic partner, Bcl-X_S, were unaltered. Therefore, in each cell line, Baicalein altered the expression of different Bcl-family protein members, resulting in a shift in their ratios favoring apoptosis, once again suggesting the broad applicability of these inhibitors to cancer treatment.

Survivin, a member of the inhibitor of apoptosis family, is overexpressed in the majority of human cancers, including cancer of the prostate. In one study treatment with Baicalein resulted in a strong decrease in survivin expression over time, resulting in undetectable levels in both PC3 and DU-145 cells by 14 h. Survivin has been shown to inhibit apoptosis by binding to active caspase-3 and caspase-7

The study by Graham P et al showed that the 12-LOX pathway of arachidonic acid metabolism regulates cell growth, survival, and apoptosis of human prostate cancer cells. Inhibition of 12-LOX led to growth inhibition associated with a specific G_1 arrest, followed by induction of apoptosis through caspase and Bcl-mediated mechanisms. Therefore, the inhibition of 12-LOX is a potential therapeutic approach in the treatment of prostate cancer. A potent inhibitor of this pathway has been shown to be Baicalein, a flavonoid found in Chinese skullcap. **(1)**
Baicalein has also been shown to induce apoptosis in human hepablastoma cell line (Hep G2) via Bcl-2 regulaton and mitochondrial dysfunction. **(2)**

Anti-Angiogenesis – Baicalein has also demonstrated anti-angiogenesis human umbilical vein endothelial cell studies. **(3)**

Parkinson's Disease - one final note of interest is the fact that baicalein has been shown to inhibit the synthesis of synuclein fibrils, which build up as plaque in nerve cells in the course of Parkinson's disease, leading to cell death. Baicalein has been shown to cause disaggregation and fragmentation of the synuclein fibril, inhibiting its formation. **(4)**

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CHINESE SKULLCAP: OVERVIEW OF BIOLOGICAL EFFECTS

James Meschino, D.C., M.S., N.D.

One of the most respected and utilized herbs in Chinese Traditional Medicine is Chinese skullcap. Although less popular among western health practitioners, the bioactive agents (e.g. baicalein flavonoid) in this herb have been the subject of intensive study in regards to their anti-cancer effects and other health benefits. In Asia, Chinese skullcap is incorporated into a number of combination herbal products that have been used in adjunctive cancer treatment. I recently had the opportunity to review the world wide published literature on this potentially important herbal agent and I have summarized it for you below in a clinically relevant format. I believe you will find the data quite intriguing and of great clinical importance.

General Features

Scutellaria baicalensis or Chinese scullcap is a member of the mint family and grows in China and Russia.[1,2] The plant root is a rich source of over 35 flavonoids, giving it a yellow color, and hence its traditional name of golden root or Huang qin, the Chinese term for yellow gold (known as Ogon in Japanese).[3,4] One of the major flavonoids contained in the root of the plant is baicalin, a flavone glycoside (12-17% of all scullcap root flavonoids), which yields the aglycone flavone baicalein, once hydrolyzed by gut bacteria.[3] Baicalein is readily absorbed into the bloodstream where it has been shown to exert a broad spectrum of important biological activities.[5] Baicalein has been incorporated into a number of herbal combinations in Traditional Chinese Medicine that treat a wide array of health conditions, including the widely used Asian herbal remedy, “Sho-saiko-to”.[6,7] Currently, baicalein is being studied for its anti-cancer, anti-inflammatory, anti-viral, anti-bacterial and anti-allergy effects.[8,9,10,11] As explained below, its ability to halt the replication of various human cancer cell lines, via the inhibition of the 12 lipoxygenase enzyme system, is attracting a great deal of attention from the scientific community as a premiere agent in cancer research.

Clinical Application and Mechanism of Action

1. Anti-Cancer Effects

Many experimental studies indicate that baicalein (5,6,7-trihydroxyflavone) prevents and inhibits cancer growth via a number of direct and indirect physiological actions:

a. Inhibits 12-lipoxygenase enzyme and induces apoptosis of cancer cells

Baicalein has been shown to inhibit the 12-lipoxygenase enzyme, which converts arachidonic acid into a specific leukotriene (eicosanoid) that is required for cancer cells to proliferate. Studies demonstrate that by inhibiting the 12- lipoxygenase enzyme baicalein has been shown to inhibit cancer cell proliferation and induce apoptosis (programmed cell death) of human gastric cancer cell lines.[12] Further, activation of arachidonic acid by the 12-lipoxygenase enzyme has been shown to be critical for metastasis of human prostate cancer cells. Experimental evidence demonstrates that after intraprostatic injection of mice with human prostate cancer cells, the prostate cells pre-treated with baicalein failed to metastasize to the lung and failed to express activity of the 12-lipoxygenase enzyme. This evidence suggests that, in the presence of baicalein, various cancer cells can be prevented from multiplying and metastasizing to other tissues and that a primary mechanism through which this occurs is via the inhibition of the 12-lipoxygenase enzyme.[13] The inhibition of cancer cell proliferation also lends itself to increased apoptosis and a better opportunity for immune cells to destroy tumor cells.[12,13] Other studies testing baicalein as an anti-tumor agent have shown that it selectively induces apoptosis in human hepatoma cell lines (liver cancer cells) with minimal influence on non-cancer cells.[14,15,28,29] Its ability to induce apoptosis is under intensive investigation as many studies provide evidence that baicalein exhibits an extraordinary ability to selectively encourage apoptosis of many different human cancer cells. This is likely one of the primary ways in which it has been a useful addition to herbal combinations that have been used in cancer treatment.[16,17,18]

b. Anti-proliferative

Baicalein has also demonstrated anti-proliferative effects on human bladder cancer cell lines and a murine bladder cancer cell line, in vitro. In an in vivo experiment, mice were injected with bladder cancer cells with concurrent oral administration of a high baicalein-yielding supplement in one group, or with no baicalein supplementation in the control group. All the control mice showed a progressive increase in tumor volume over the ensuing days of the study, whereas the mice treated with baicalein (scutellaria) showed a significant inhibition of tumor growth.[19] In other experiments, baicalein has demonstrated anti-proliferative effects on human pancreatic cancer cell lines and T lymphoid leukemia cells. The mechanism of action was shown to be through the reduction of protein tyrosine kinase activity and protein

kinase C activity. Both of these enzyme activities are required for normal cellular proliferation to occur. Thus, in addition to inhibiting the 12-lipoxygenase enzyme pathway and inducing apoptosis of cancer cells, baicalein is also known to reduce cancer cell proliferation by directly or indirectly inhibiting specific enzymes (protein tyrosine kinase and protein kinase C) required for cellular division and proliferation.[28]

c. Anti-oxidant

Baicalein is also a strong antioxidant, and has been shown to protect DNA from undergoing cancerous mutations in challenge studies using potent carcinogens such as benzo{a}pyrene, and toxins such as aflatoxin (AF) B.[1,20,21]

d. Stimulates DNA repair enzymes

In vitro evidence indicates baicalein stimulates recombination and repair of damaged DNA, supporting its traditional inclusion in cancer formulas, and suggesting possible use after sunburn and radiation damage.[22]

e. Inhibits the 5alpha-reductase enzyme

Baicalein has been shown to inhibit the 5alpha-reductase enzyme, which converts testosterone to dihydrotestosterone (DHT). DHT is strongly associated with the development of prostate enlargement (benign prostatic hyperplasia) and prostate cancer. As such, baicalein is reported to be potentially useful for the prevention and/or treatment of androgen-dependent (testosterone-driven) disorders, including prostate enlargement and prostate cancer.[23]

Human Studies Involving Prostate Cancer Patients

Studies on humans have primarily involved patients with advanced prostate cancer, who were shown to be unresponsive to traditional medical drugs and other interventions. In a study performed by researchers from the University of California at San Francisco and Memorial Sloan-Kettering Cancer Center in New York, the oral administration of a herbal combination containing baicalein to patients with metastatic prostate cancer (previously unresponsive to standard treatments) demonstrated reversal and/or stabilization of their condition with significantly improved survival, quality of life scores and other positive outcomes in almost all patients receiving the supplement. After a median treatment period of 57 weeks, 100% of patients with androgen-dependent cancer had a decline of 80% or more in their prostate-specific antigen (PSA) levels, and these levels were undetectable in 81% of the patients. In 31 of 32 patients, the testosterone level declined to that seen in castrated men. Of nine men with elevated prostatic acid phosphatase levels (a marker for cancer progression), all had declines of more than 50%. Of two patients with positive bone scans, one had a complete disappearance of cancerous bone lesions, and the other showed

improvement. One patient had a complete disappearance of a bladder mass, demonstrated by CT scan.

Of 35 patients with androgen-independent prostate cancer 54% showed a drop in their PSA levels by 50% or more, and of 25 patients with metastasis to bone, 2 showed marked improvement, 7 remained stable, 11 progressed, and 5 had no further bone scan follow up due to an increasing trend in their PSA levels.[24] Other studies involving men with known prostate cancer, taking this herbal combination, have demonstrated similar results. In one study PSA levels dropped from 100ng/mL to 24 and from 386ng/mL to 114 within 16 months of supplementation in a two case report of hormone-refractory prostate cancer.[25] Studies following larger groups of men with prostate cancer for up to four years have also shown this herbal formula to significantly reduce PSA levels and improve survival and quality of life scores.[26,27]

Researchers attribute much of the anti-tumor effects of this herbal combination to the presence of baicalein. According to researchers, baicalein has demonstrated impressive anti-cancer effects against androgen-dependent and androgen-independent human prostate cancer cells lines, and many of its anti-cancer mechanisms have been uncovered in recent years, in vitro and in vivo experiments.[16,17,18,24,25,26,27,28] It appears to be of particular benefit in prostate cancer treatment and in combination with other phytonutrients (plant-based nutrients) and micronutrients (vitamins and minerals) may help to prevent the development of clinically important prostate cancer, reducing prostate cancer incidence.[26] Animal studies indicate that baicalein may be effective in the treatment of other cancers as well, with experimental evidence supporting its potential use in stomach, pancreatic, bladder, liver[12, 14, 15,19, 28] and breast cancer. [29, 30, 31]

Hepatoma and Leukemia Patients

Based on its traditional use, and the documentation of its anti-tumor effects against various human cancer cell lines, Baicalein has been used recently by doctors in Asia as a complementary supplemental agent in the treatment of hepatomas and leukemia in human subjects.[29]

2. Anti-viral/Anti-HIV

Baicalein has been shown in experimental studies to inhibit the activity of HIV-1 integrase enzyme, which is an essential enzyme in the life cycle of the HIV-virus. This enzyme is responsible for catalyzing the insertion of the viral genome into the host cell chromosome. It is an attractive target for the design of a HIV antiviral drug because the integrase enzyme has no human counterpart.[32] In a review of all herbal medicines frequently used as an alternative medical therapy by HIV positive patients and AIDS patients, JA Wu et al, emphasized that experimental data suggest that the flavonoid, Baicalein inhibits infectivity and replication of HIV, and shows great promise as an important component to be included in herbal remedies used for HIV treatment.[33]

3. **Asthma and Allergies**

   Baicalein inhibits type I and II hypersensitivity reactions, confirming its traditional use in asthma[34,35] and allergic dermatitis.[3]

4. **Anti-inflammatory**

   Baicalein has shown impressive anti-inflammatory effects that appear to be mediated via repression of leukotriene B4 (inhibition of the 5-lipoxygenase enzyme), inhibition of interleukin-1B, and prostaglandin E2. Studies reveal it may be of benefit in the treatment of gingivitis (gum inflammation).[29, 36, 37]

5. **Alzheimer's and Dementia**

   In a rat model Baicalein has been shown to prevent the build up of amyloid beta peptide (Abeta), which is known to generate free radical damage to brain cells and participate to a significant degree in the development and progression of Alzheimer's disease. Additionally, chronic inflammation occurs in Alzheimer's pathogenesis and Baicalein was shown to inhibit the 12- lipoxygenase enzyme responsible for brain inflammation, to a large degree. When tested against other known 12-lipoxygenase inhibitor agents, only Baicalein was able to attenuate both nerve cell death (apoptosis) and over-expression of Abeta production. As such, Baicalein may help to prevent or forestall the development of Alzheimer's disease by reducing the build up of the toxic brain protein, Abeta and inflammatory mediators, and preventing brain cell death, which has been shown to be triggered by the accumulation of Abeta in affected brain cells.[38] In studies using human neuroblastoma cells, the antioxidant properties of Baicalein were shown to inhibit free radical damage to these nerve cells induced by treatment with hydrogen peroxide (a powerful free radical generating agent). The researchers argue that oxidative (free radical) stress plays an important role in the development of neurodegenerative diseases (e.g. Alzheimer's disease, Multiple Sclerosis, Parkinson's disease) and that antioxidants such as Baicalein and Quercetin (also a flavonoid) may be useful bioactive agents in the prevention and/or management of these conditions, pending further studies. Both of these flavonoids have shown impressive protective effects under experimental conditions.[39]

6. **Inhibition of Xanthine Oxidase (Gout Treatment)**

   Baicalein is one of few bioactive agents that have been shown to inhibit the enzyme xanthine oxidase, which is responsible for the conversion of hypoxanthine to xanthine, and xanthine to uric acid, in the pathway for degradation of purines in the body. High levels of uric acid is a hallmark feature of gout and thus, Baicalein supplementation may be an effective complementary intervention in the long-term management of this condition.[40] The drug Allopurinol is a Xanthine Oxidase inhibitor and is a primary medication prescribed for the treatment of gout.

Dosage

Therapeutic Purposes: The usual doses for therapeutic purposes ranges from 150 to 200 mg per day of Baicalein.[6]

General wellness: Consider 50-100mg per day.

Toxicity and Adverse Side Effects

Baicalein supplementation at therapeutic doses appears to be safe with no reports of significant side effects or toxicity. Baicalein or Chinese scullcap are not known to be contraindicated for any health conditions.[41,42]

Drug-Nutrient Interaction

There are no known drug-nutrient interactions for Baicalein or Chinese skullcap.[42]

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3. Essential Fatty Acids

Lipoxygenase and Cancer: Incriminating Evidence For Dietary Arachidonic and Linoleic Acids

Mounting evidence suggests that lipoxygenase (LO)-catalyzed products have a significant influence on the development and progression of human cancers. Compared with normal tissues, significantly elevated levels of LO metabolites have been found in lung, prostate, breast, colon, and skin cancer cells, as well as in cells from patients with both acute and chronic leukemias. LO-mediated products trigger diverse biological activities required for cancer growth, and influence growth factor and transcription factor activation, oncogene induction, stimulate tumor cell adhesion, and regulate apoptosis.
During the past 10 years, pharmacological agents that specifically inhibit the LO-mediated signaling pathways are now commercially available to treat inflammatory diseases such as asthma, arthritis, and psoriasis. LO inhibitors are now being explored as agents to prevent and treat certain cancers.

Mechanisms Through Which Arachidonic Acid and Leukotrienes Encourage Cancer Development And Growth

Eicosanoids derived from the arachidonic acid cascade have been implicated in the pathogenesis of a variety of human diseases, including cancer, and appera to play important roles in tumor promotion, progression, and metastatic disease. Although most attention has focused on PG_2 and other COX-derived metabolites, mounting evidence suggests that LO-catalyzed products, LTs, and HETEs also exert profound biological effects on the development and progression of human cancers. For example, 12-LO mRNA expression has been well-documented in many types of solid tumor cells, including those of prostate, colon, and epidermoid carcinoma. Also, 12(*S*)-HETE production by some tumor cells, including prostate cells, has been positively correlated to their metastatic potential.

Studies indicate that 12(*S*)-HETE is a critical intracellular signaling molecule, stimulating PKC and eliciting the biological actions of many growth factors and cytokines that regulate transcription factor activation and induction of oncogenes or other gene products needed for cancer growth. Some of these include EGF, FGF, PDGF, tumor necrosis factor, granulocyte-macrophage colony-stimulating factor, and both IL-1 and IL-3. Additionally, PKC activation by 12(*S*)-HETE mediates the release and secretion of cathepsin B, which is involved in tumor metastasis and invasion, particularly in colon cancer cells.
As well, tumor cell synthesis of 12(*S*)-HETE stimulates adhesion by increasing the surface expression of integrin receptors. Besides 12(*S*)-HETE, other LO metabolites, particularly the 5-LO products (5-HETEs), have been implicated in cancer development. In fact, studies show that 5-HETE directly stimulates prostate cancer cell growth.

In general, evidence from studies in human cancer cells shows that the LO pathways are involved in carcinogenesis in several major tissues. More importantly, a few studies in animal carcinogenesis models show that inhibition of the LO pathways also may inhibit carcinogenesis
Two naturally occurring agents, Baicalein and esculetin are among several experimental compounds demonstrating 12-LO inhibitory activity that may show promise as antiproliferative agents. **(1)** These agents will be discussed in the next section

Lipoxygenase and Prostate Cancer

Diets high in fat are associated with an increased risk of prostate cancer, although the molecular mechanism is still unknown. **(2)** The increased risk of prostate cancer associated with high dietary intake of animal fats shown in multiple population-based studies may in part be explained by the rich sources of fatty acids like arachidonate in these diets that favor conversion to proinflammatory eicosanoids. **(4)**

Studies have previously reported that arachidonic acid, an omega-6 fatty acid common in the Western diet, stimulates proliferation of prostate cancer cells through production of the 5-lipoxygenase metabolite, 5-HETE (5-hydroxyeicosatetraenoic acid). Research reveals that 5-HETE is also a potent survival factor for human prostate cancer cells. These cells constitutively produce 5-HETE in serum-free medium with no added stimulus. Exogenous arachidonic acid markedly increases the production of 5-HETE. Inhibition of 5-lipoxygenase by MK886 completely blocks 5-HETE production and induces massive apoptosis in both hormone-responsive (LNCaP) and -nonresponsive (PC3) human prostate cancer cells. Exogenous 5-HETE protects these cells from apoptosis induced by 5-lipoxygenase inhibitors, confirming a critical role of 5-lipoxygenase activity in the survival of these cells. These findings provide a possible molecular mechanism by which dietary fat may influence the progression of prostate cancer. **(2)**

Altered eicosanoid biosynthesis in relation to prostate cancer development has been well documented. Initial studies found dramatically reduced levels of arachidonic acid; 10-fold greater turnover in malignant compared to benign prostatic tissue suggested a possible increase in metabolism via the LO and COX pathways in this tissues. Linoleic

acid stimulated cell growth in experiments conducted in human prostate cancer cells, whereas indomethacin, esculetin, and piroxicam inhibited it, which provides significant proof of the involvement of eicosanoids in prostate cancer cell proliferation. Although attention has focused on COX-derived products, particularly PGE_2, COX inhibitors indomethacin and aspirin failed to reduce human prostate PC3 cell DNA synthesis, although arachidonic acid antagonist eicosatetraynoic acid did reduce synthesis, suggesting that LO products are essential in modulating prostate DNA synthesis. Thus, researchers now recognize that inhibiting or down-regulating the LO synthesis pathway is an important target for prevention and management of certain cancer, including prostate cancer.

Studies show that 5-HETE, particularly the 5-oxo-eicosatetraenoic form, stimulates PC-3 cell growth similarly to arachidonic acid. 5-HETEs also effectively reversed growth inhibition produced by a drug known as MK-886 (Merck), a FLAP inhibitor

In addition to 5-LO products, the 12-LO metabolite 12(*S*)-HETE has been shown to play a critical role in prostate tumor metastasis and invasion . In 122 matched normal and cancerous prostate tissues, 12-LO mRNA expression was confined to prostate epithelial cells and elevated in malignant cells. As well, elevated levels of 12(*S*)-HETE mRNA significantly correlated with advanced stage, poor differentiation, and invasive potential of prostate cancer cells. Indeed, addition of 12(*S*)-HETE to rat Dunning R3327 and human prostate adenocarcinoma cells significantly increased their cellular motility and ability to invade basement membrane and increased their metastatic activity.

Additional biochemical evidence demonstrated that 12(*S*)-HETE stimulates secretion of cathepsin B, which is involved in tumor metastases and integrin expression in other tumor cells, providing support for the importance of LO products in the development and spread of prostate and other human cancers. It has also been shown that 12(*S*)-HETE involves selective activation of membrane-associated PKC, which is known to be involved in cancer growth. **(1)** This information takes on greater significance with the appreciation that cells that have the appearance of prostate cancer can be found in the prostate gland of nearly half of all men over the age of 50. **(4)**

Lipoxygenase and Breast Cancer

Studies have shown that diets rich in linoleic acid, stimulate mammary tumor growth induced by either MNU or DMBA, two frequently used carcinogens in animal used in animal studies. Conversely, dietary supplementation with n-3 polyunsaturated fatty acid, particularly fish oils, has been shown to retard mammary tumor growth and metastasis in a variety of animal tumor models. Several studies have shown that mammary tissue of rats fed 20% menhaden oil produced lower levels of LTB_4 and PGE_2 compared with groups receiving 20% corn oil or primrose oil, supporting the theory that PGs and LTs may be involved in mammary carcinogenesis
Markedly elevated levels of both COX and LO metabolites have been documented in human breast cancer tissue compared with tissue from patients with benign disease.

Other studies have indicated that linoleic acid enhanced the invasive capacity of breast cancer cells and that this effect could be completely blocked by adding esculetin, an

inhibitor of 5- and 12-LO, but not by adding the COX-specific inhibitor piroxicam. This evidence strongly implicates the lipoxygenase pathway in cancer development.

The interaction of growth factors, such as epidermal growth factor (EGF) with their receptors, on breast cancer cells can lead to the hydrolysis of phospholipids and release of fatty acids, such as arachidonic acid, which can be further metabolized by the lipoxygenase (LO) pathway. Several LO products have been shown to stimulate oncogenes and have mitogenic and chemotactic effects.

More specifically, LO products, such as the hydroxyeicosatetraenoic acids (HETEs), have been shown to have actions highly relevant to cellular growth and migration. They have significant mitogenic and chemotactic effects and also can stimulate the expression of several oncogenes and other studies suggest that the biosynthesis of 12(S)-HETE by tumor cells is a determinant of their metastatic potential. The LO products of linoleic acid have been shown to potentiate the mitogenic effects of epidermal growth factor and linoleic acid can stimulate the growth of MCF-7 breast cancer cells.
There is also evidence to show that leukocyte 12-LO mRNA expression is upregulated in breast cancer cells and tissues, compared with their normal counterparts

Results of other studies also suggest that activation of the 12-LO pathway may play a key role in basal and EGF-induced breast cancer cell growth. As such, it appears that 12 LO end products up-regulate epidermal growth factor receptors, which are known to be involved in breast cancer development. In fact, treatment with EGF, a potent breast cell growth factor, caused a significant increase in 12-LO enzyme activity, as well as leukocyte-type 12-LO protein expression in MCF-7 cells. These results suggest that 12-LO expression is enhanced in breast cancer, and this increased expression may be secondary to locally derived growth factors involved in breast cancer cell growth and development. This is further supported by the specific inhibition of the growth of MCF-7 cells by LO inhibitors, but not CO, inhibitors. **(3)**

Lipoxygenase And Other Cancers
12- and 15-HETE are major arachidonic acid metabolites have also been incriminated in squamous epithelial carcinomas of the head and neck, Also, 12(*S*)-HETE is the predominant metabolic product of metastatic B16 melanoma cells and excess LT production, specifically LTC_4, has been documented in cells from patients with both acute and chronic leukemias

Agents Shown To Block The LO and CO Pathways And Inhibit Cancer

Non Steroidal Anti-Inflammatory Drugs - Among the tumor types that have been shown to overexpress COX-2 is prostatic adenocarcinoma. Both selective and nonselective COX-2 inhibitors, such as celecoxib and sulindac, respectively, have been shown to be effective in preventing the progression of premalignant colorectal adenomas in preclinical and clinical studies as well as in preclinical studies in bladder, skin, and mammary gland. It has been suggested that these NSAIDs may be useful for the prevention or therapy of prostate cancer as well. Indeed, both sulindac and celecoxib have been shown to inhibit the growth or induce apoptosis in human prostate cancer cell lines in vitro, and the NSAID flufenamic acid has been shown to inhibit the expression of

the androgen receptor in LNCaP cells. Recently, an inverse association between aspirin or ibuprofen use and prostate cancer development was noted in several case control studies. **(4)** Unfortunatley, the long-term use of NSAIDs is associated with gastrointestinal erosion, ulcers, intestinal bleeding, as well as liver and kidney toxicity. As such, their regular use as prophylactic cancer intervention may produce serious and life-threatening side effects and are clearly contra-indicated in patients with certain co-morbidity issues (e.g. previous intestinal ulcers, kidney disease/dysfunction/transplant etc) **(5,6,7)**

Flavonoids (e.g. Baicalein), Esculetin, Curcumin and Tea Polyphenols - A number of natural occurring agents that affect arachidonate metabolism, either via cyclooxygenase or the alternative 5-lipoxygenase have demonstrated chemoprevention effects against cancer. Some of these include flavonoids, curcumin, and tea polyphenols. **(4)** Others include esculetin and hydroxamic acids.

Esculetin and Baicalein, two naturally occurring agents, have been shown to inhibit lipoxygenase enzymes and modulate cancer development and growth. In vivo studies have shown that esculetin significantly inhibits the development of mammary tumors induced by DMBA in female Sprague Dawley rats fed either a high-fat (20% soybean oil) or low-fat (0.5% soybean oil) diet. As well, esculetin has been effective in reducing the invasive and metastatic activity of malignant tumor cells. Murine melanoma cells pretreated with 50 μM esculetin and injected into syngeneic mice produced significantly lower numbers of melanoma colonies on the lung surface and fewer tumor metastases.

Baicalein, a flavonoid isolated from Scutellaria baicalensis Georgy roots (Chinese scullpcap), is a key component of Japanese herbal medicine Sho-saiko-to, commonly used to treat chronic liver diseases in Japan. Early studies conducted in rat platelets showed that low concentrations of baicalein exert potent 12-LO inhibitory activity, whereas much higher levels are required for COX inhibition. Other studies suggest that baicalein also blocks 5-LO and production of LTC_4.

The antiproliferative effects of baicalein have been reported in several in vitro models. One study showed that baicalein reduced DNA synthesis and inhibited growth in two human hepatoma cell lines. Similar growth-suppressive effects of baicalein have been reported by other investigators in various hepatocellular cancer cell lines. Likewise, treating human breast cancer cells (MCF-7) with baicalen at microgram levels produced potent antiproliferative activity. In human T-lymphoid leukemia cells (CEM) stimulated with 10% FCS, showed reduced cell proliferation with addition of baicalein in a dose dependent fashion, with evidence of reduced activity of messengers known to be of significance in cancer growth. Rat studies showed that baicalein induced apoptosis in carcinosarcoma cells in a dose-dependent manner. Nevertheless, these initial data suggest that baicalein may be an effective chemopreventive agents.

Hydroxamic acids have also shown lipoxygenase inhibition and reduced cancer growth. *N*-benzyl-*N*-hydroxy-5-phenylpentanamide (BHPP), one of a series of hydroxamic acids, originally developed by Rorer, that was found to exert 5-LO inhibitory activity in rat leukocytes. Additional studies demonstrated 12-LO inhibitory activity in Lewis lung carcinoma cells and porcine leukocytes and in murine B16a melanoma cells. Of interest to natural health practitioners is the fact that certain hydroxamic acid are naturally occurring in cernitin pollen extract, often used in the treatment of prostate

enlargement. A cernitin-derived hydroxamic acid has been shown to inhibit replication of certain human prostate cancer cells under experimental conditions. (Fouad K. et al, Prostate. 2006)
Many other experimental studies have provided evidence for the cancer inhibition effects of BHPP via its inhibitory influences on the 5 and 12 lipoxygenase enzymes. **(1)**

Summary

In summary, evidence suggests that tumor cells and several normal cells have LO activity, and both arachidonic acid and linoleic acids are converted to LO products, such as HETEs and hydroxyoctadecadienoic acids (HODEs). The 12-LO product, 12(S)-HETE, has been shown to play an important role in the metastatic process. 12(S)-HETE mediates the adhesion of tumor cells to the subendothelial matrix after endothelial retraction by a protein kinase C-dependent process. LO products, such as 12- and 15-HETE, also have mitogenic effects on endothelial cells.

Research demonstrates that 12-HETE is the predominant arachidonic acid metabolite produced by highly metastatic tumor cells. Furthermore, these highly metastatic cells synthesize much greater amounts of 12-HETE than the low metastatic tumor cells. Thus, an increased concentration of 12-HETE, produced by activated platelets, the tumor cells themselves, leukocytes, or by vascular cells, are thought to be affiliated with the proliferative and metastatic processes

LO metabolites of arachidonic acid have been reported to mediate tumor necrosis factor-induced protooncogene *c-fos* expression . In addition, EGF-induced mitogenic activity has been linked to the formation of LO products of linoleic acid, the HODEs

Thus, a growing body of evidence suggests that specific metabolites of arachidonic and/or linoleic acid serve as central elements in signal pathways necessary for cell mitogenesis, as induced by growth factors or oncogenic transformation. **(3)**

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Essential Fatty Acid Supplementation: Why the combination of flaxseed, borage seed and fish oil is the optimal blend for the heart, brain, joints and the prevention of cancer

James Meschino DC, MS, ND

In Brief

Recent research confirms the findings of previous investigations, indicating that the synergistic action of essential fatty acids, richly supplied by flaxseed oil, borage seeed oil and fish oil, encourage the production of eicosanoids (mini-local hormones) that help defend the body against heart attack, stroke (and other cardiovascular diseases), control blood pressure, reduce joint inflammation, reduce the incidence of age-related dementia, and reduce risk of cancer development. Essentially, the polyunsaturated fatty acids, richly supplied by these oils, provide the body with the building blocks from which our cells make specific prostaglandins, prostacyclins, thromboxanes and leukotrienes (collectively known as eicosanoids, which act like local hormones) that strongly influence tissue behaviour and cellular function.

Whereas the polyunsaturated fatty acids supplied by foods high in animal fat, as well as certain types of vegetable oils that are high in linoleic acid, promote the formation of disease-promoting, inflammation-inducing eicosanoids, a lower animal fat diet, in conjunction with monounsaturated fatty oils (e.g. olive oil) accompanied by daily supplementation with flaxseed, borage seed and fish oil (totalling 2400-3600 mg per day, consisting of equal quantities of each oil) promotes the formation of eicosanoids that reduce risk of many diseases, suppress inflammatory states and help preserve cognitive function as we age.

Introduction

Eicosanoids are molecules produced by most tissues in the body and are known to exert hormone-like effects on the cells that produce them (autocrine effects), as well as on neighboring cells (paracrine effects). Research initiatives over the past 30 years reveal that various types of eicosanoids influence biological steps involved in the prevention and promotion of inflammatory states, cardiovascular disease and cancer. Of the utmost importance is the fact that eicosanoid synthesis is largely dictated by the ingestion of various types of dietary polyunsaturated fats, such that ingestion of certain polyunsaturated fats gives rise to eicosanoids involved in the prevention (or suppression)

of inflammatory states (including arthritis), cardiovascular disease and cancer, whereas the ingestion of other less desirable polyunsaturated fatty acids have been shown to promote inflammation, cardiovascular disease and cancer. As such, specific dietary practices and the use of targeted dietary supplements should be viewed, by individuals and health practitioners, as extremely important interventions in the prevention and management of these prevalent health conditions. **(1-4)**

Important Classes of Eicosanoids: Prostaglandins, Prostacyclins, Thromboxanes and Leukotrienes

Eicosanoids are derived from dietary polyunsaturated fatty acids, which are incorporated as esters into the phospholipids and diacylglycerols found in the cell membrane and nuclear membrane**.** Eicosanoids are not stored within cells, but are synthesized as required, and rapidly inactivated. They are potent in the nanomolar range and have a half-life ranging from seconds to minutes.

The initiation of eicosanoid biosynthesis occurs when a cell is stimulated or influenced by mechanical trauma, cytokines (released by immune cells) growth factors or other stimuli. The stimulus may even be an eicosanoid from a neighboring cell. This promotes the release of a phospholipase enzyme at the cell membrane, which travels to the nuclear membrane, where it catalyzes a reaction (hydrolysis) that releases a 20-carbon polyunsaturared fatty acid. This hydrolysis appears to be the rate-determining step for eicosanoid formation. As such, the activityof phospholipase enzymes play a prominent role in the eicosanoid synthesis, and the disease states influenced by eicosanoids. **(1-4**)

Of interest is the fact that one of the effects of corticosteroid drugs, such as prednisone, is that they inhibit phospholipase A_2 activity, thereby, suppressing the synthesis of pro-inflammatory eicosanoids in the conventional management of a variety of complex inflammatory conditions (e.g. rheumatoid arthritis, other autoimmune conditions etc). Unfortunately, corticosteroid drug administration is associated with a diverse array of significant side effects such as hypertension, thrombophlebitis, thromboembolic events, glucose intolerance and aggravation of diabetic states, hypokalemia, hypocalcemia, bone demineralization, sodium and fluid retention, metabolic alkalosis, weakened immunity, skin eruptions, impaired wound healing, skin thinning, peptic ulcer, pancreatitis, ulcerative colitis, and other problems. For this reason, patient monitoring is imperative during corticosteroid use and medical practitioners are instructed to recommend these drugs with extreme caution. **(5)**

In regards to this matter, it has been shown that eicosapentaenoic acid (EPA) inhibits phospholipase A_2 release of AA from cell membrane, which is one additional means by which omega-3 fats have been shown to reduce the inflammatory response. **(42)**

With respect to eicosanoid synthesis, once released from cell membrane phospholipids by phospholipase A_2 ($cPLA_2$) enzyme, the 20-carbon polyusaturated fatty acids are converted into either prostanoids (prostaglandins, prostacyclins or thromboxanes) by a cyclooxygenase enzyme or into leukotrienes by a lipoxygenase enzyme. The 20-carbon fatty acids that are converted into these classes of eicosanoids include arachidonic acid (an omega-6 fat), dihomo-gamma linolenic acid (an omega-6 fat) and eicosapentaenoic

acid (an omega-3 fat). In the overall scheme of things arachidonic acid (AA) is converted into prostanoids and leukotrienes that that promote inflammation, cardiovascular disease and cancer, whereas dihomo-gamma linolenic acid (DGLA) and especially eicosapentaenoic acid (EPA) are converted into prostanoids and leukotrienes that suppress inflammatory events, cardiovascular disease and the promotion of cancer. **(1-4)**

Dietary and Supplemented Essential Fatty Acids and Eicosanoid Synthesis

Of interest to this discussion is the fact that linoleic acid (LA), an omega-6 fatty acid, commonly found in corn oil, sunflower seed oil, safflower seed oil, mixed vegetable oils and other oils, has been shown to be de-saturated and elongated by enzymes within the human body to form AA. Arachidonic acid (AA) itself is found in high concentrations in high fat animal foods, such as red meat and pork, as well as any milk or yogurt product higher than 1% milk fat and any cheese higher than 3% milk fat. This list would extend of course to ice cream, whipped cream, cream, cream cheese, regular sour cream etc. As such, the North American diet, with its high animal fat content and generous use of oils rich in linoleic acid, provides cells with generous amounts of polyunsaturated fats (AA) from which to produce prostanoids and leukotrienes that promote inflammation, cardiovascular disease and cancer. The key point here is that DGLA and EPA directly compete with AA for conversion to less harmful and more health-promoting prostanoids and leukotrienes, such that higher cellular concentrations of EPA and DGLA and lower cellular concentrations of LA and AA, result in a greater synthesis of health-promoting/ inflammation suppressing eicosanoid production and a reduced synthesis of the dangerous and inflammation-provoking eicosanoids made from AA. This is because AA, DGLA and EPA, all compete with each other for activation by cyclo-oygenase and lipoxygenase enzymes. As such, a simple dietary and supplementation strategy to enrich cellular concentrations of EPA and DGLA involves regular consumption of fatty fish (a rich source of EPA and docosahexaenoic acid – DHA), and supplementation with a combination of flaxseed oil (a rich source of alpha-linolenic acid - ALA, which the body can de-saturate and elongate into EPA), fish oil (a rich source of EPA and DHA) and borage seed oil (a rich source of gamma-linolenic acid – GLA, which the body converts into DGLA).

It should also be noted that ALA competes with LA for the elongase and de-saturase enzymes for its conversion from ALA to EPA. In turn, this blocks the conversion of LA into AA. Studies show that ALA displaces LA from the elongase and desaturase enzymes that produce AA. Thus, flaxseed oil supplementation not only provides raw materials from which the body can make EPA (as well as DHA, which can be made from EPA – DHA is important for vision and brain function throughout life), but ALA also helps to decrease membrane concentrations of AA by blocking the conversion of LA (which is oversupplied by the North American and Western diet) into AA, and thus helps promote the synthesis of health-promoting eicosanoids, and helps suppress the synthesis of disease and inflammation-promoting eicosanoids. **(1,2,3,4,41)**

Eicosanoids of Importance In The Disease Process

Although the body makes numerous types of eicosanoids, (some of which have no known physiological function at this time) there are several which have been shown to significantly impact the prevention or promotion of various health conditions.

Inflammation: In regards to promotion of inflammation, prostaglandin E_2 (PGE-2) – formed from activation of AA by cyclo-oxygenase, acts inside the cell to produce various types and quantities of cytokines, which are pro-inflammatory agents that complete the process by bringing active leukocytes to the injury site. Many leukocytes (white blood cells) convert AA into pro-inflammatory leukotrienes, via 5-lipoxygenase enzyme, such as pro-inflammatory leukotriene B4 (LTB_4), which makes local blood vessels more permeable. In turn, plasma leaks out into the connective tissues causing more swelling. PGE_2 also sensitizes pain nerve endings, increasing the sensation of pain from the inflamed tissues.

In contrast to this, DGLA yields PGE_1, which powerfully counteracts PGE_2, toning down the inflammatory response. The body slowly synthesis DGLA from LA (LA-------GLA-----DGLA), however, as we age this conversion slows down, allowing inflammatory states to occur more easily. In this regard many health experts recommend daily supplementation with 800-1200 mg of borage seed oil (which is 22% GLA compared to only 9% GLA supplied by evening primrose oil). DGLA also yields the leukotriene LTB_5 which counters the inflammatory action of the AA-derived LTB_4.

EPA acts as a precursor for prostaglandin-3 and promotes the synthesis of leukotriene -5 groups, all of which suppress the inflammatory response. It is noteworthy that the body can synthesize EPA from both ALA and DHA.

As noted above EPA also inhibits the action of phospholipase A_2, resulting in less AA being recruited into the pro-inflammatory cascade, and thus providing greater substrate concentration of EPA and DGLA to be activated cyclo-oxygenase and lipoxygenase for conversion into anti-inflammatory eicosanoid mediators. **(42)**

Investigative studies also indicate that although DGLA forms no leukotrienes via the lipoxygenase pathway, derivatives of DGLA have been show to block the conversion of AA to inflammatory leukotrienes, thereby reducing the inflammatory response. **(43)**

In summary, DGLA and EPA participate in biochemical cascades that parallel and compete with the arachidonic acid cascade. EPA provides the most important competing cascade. DGLA provides a third, less prominent cascade. These two parallel cascades (from EPA and DGLA) soften the inflammatory effects of AA and its products. Low dietary intake of these less-inflammatory essential fatty acids, has been linked to several inflammation-related diseases. **(6,7,8)**

Cardiovascular Disease: In regards to cardiovascular disease, a prostanoid synthesized from AA via cyclo-oxygenase, namely thromboxane A_2 increases risk of cardiovascular disease by constricting blood vessels (increasing smooth muscle tone) and by increasing platelet coagulation. Platelet coagulation, forming a plug in the artery wall (in the area of atherosclerosis development), is often the final precipitating event leading to a myocardial infarction, angina or other ischemic vascular event. Thromboxane A_2 is

synthesized with platelets. Interestingly, endothelial cells in the blood vessel wall synthesize an anti-platelet aggregatory protanoid from AA, known as prostacyclin I-2 (PGI_2). However, high tissue concentrations of AA tends to produce a strong vasoconstriction response, and enhances platelet aggregation due to the synthesis of thromboxane A_2, which generally supercedes the anti-aggregatory influence of endothelial-derived PGI_2. However, higher tissue levels of EPA permits the anti-aggregation effects of PGI_2 to be expressed with greater influence, helping to reduce risk of many ischemic cardiovascular events.

EPA inhibits synthesis of thromboxane (TXA_2) and leukotriene B-4 (LTB_4), by platelets and macrophages. Reduction of the pro-aggregatory, vasoconstrictive TXA_2 decreases the thrombotic tendency of platelets, reducing risk of cardiovascular disease. This is augmented by the limited depression of the vasoactive anti-aggregatory prostacyclin (PGI_2) secreted by endothelial cells and the generation of anti-aggregatory prostaglandin I-3 (PGI_3) from EPA. EPA has been shown to reduce blood pressure and blood viscosity and modulate membrane fluidity and associated enzyme and receptor functions. The collective effects of omega-3 fatty acids likely account for the reduction in coronary arterial disease in populations consuming foods rich in omega-3 fatty acids. **(1,2,3,9,10,11,12).**

Additionally, a study published in 2007 showed that supplementation with flaxseed oil significantly lowered blood pressure in patients with dyslipidemia compared to subjects supplemented with safflower seed oil; an oil rich in linoleic acid (LA). This study adds to the body of evidence that ALA (from flaxseed oil) is efficiently converted into EPA within the body, facilitating enhanced production of eicosanoids that dilate blood vessels, which decrease peripheral resistance to blood flow, reducing high blood pressure. **(38).** A previous study (Zhao G et al 2004) also demonstrated that dietary alpha linolenic acid (ALA) reduced inflammatory and lipid cardiovascular risk factors in hypercholesteolemic men and women. **(39)** In conjunction with animal studies these studies further substantiate the level of efficiency in which the body is able to convert ALA into EPA. **(40)**

Cancer:

Abundant evidence suggests that the conversion of AA into various leukotrienes, via the 5 lipoxygenase enzyme and the 12-lipoxygenase enzymes has a profound influence on the development and progression of human cancers. Compared with normal tissues, significantly elevated metabolites of the lipoxygenase pathway (using AA as the essential fatty acid), are common features in lung, prostate, breast, colon, and skin cancers, as well as in cells from patients with both acute and chronic leukemias. Lipoxygenase end-products derived from AA (especially leukotriene A_4 and 5-hydroxyeicosatetraenoic acid) have been shown to elicit diverse biological activities required for neoplastic cell growth. They influence cellular growth factors, transcription factor activation (which up-regulates oncogenes), oncogene induction, help to inhibit programmed cell death (apoptosis), and influence other factors important for cancer cell survival, progression and metastasis. As well, prostanoids derived from AA have also been shown to stimulate cell proliferation, which increases risk for cancer development, and promotes proliferation of existing (possibly latent) cancer cells. **(13)** In recent years it has been

recognized that certain non steroidal anti-inflammatory drugs, such as aspirin, may reduce risk of colon cancer and other cancer by blocking the action of cyclo-oxygenase enzyme. In turn, this inhibits the synthesis of PGE_2 and other prostanoids formed from AA, which are involved in inflammation, platelet aggregation, vasoconstriction and cellular proliferation. In this regard non steroidal anti-inflammatory drugs have been used to reduce inflammation, pain and fever, as well to reduce platelet stickiness in an effort to reduce risk of coronary disease. Experimental and epidemiological studies recognize that these drugs may also offer protection against certain cancers by slowing cellular proliferation. **(14,15).** However, non steroidal anti-inflammatory drugs also increase risk of gastro-intestinal bleeding, ulcers, liver and kidney toxicity and may hasten the progression of cartilage erosion in osteoarthritis. In fact 10,000 to 20,000 individuals die each year in the United States from bleeding disorders (and other complications) induced by the frequent use of non steroidal anti-inflammatory drugs. As such, their application as prophylactic agents in cancer prevention may place individuals at risk for other dire and life-threatening health conditions. **(16,17,18)**

Of interest to natural health practitioners is the fact that certain natural agents such as the flavonoid baicalein (from Chinese skullcap) and curcumin (from the spice turmeric) have been shown to block the 12 lipoxygenase enzyme. Experimental, preclinical and preliminary studies indicate that these natural compounds are able to suppress cancer development and block the recurrence of cancer in colon cancer patients. **(13)** Moreover, other natural agents have been shown to block cyclo-oxygenase without disrupting platelet function or causing bleeding disorders or organ toxicity. Highly effective agents include curcumin, white willow extract, ginger, boswellia, and quercetin. Each of these natural compounds have been used to effectively treat a variety of joint inflammatory conditions, and may hold promise as natural interventions to also help prevent cancer by blocking the conversion of AA to prostanoid metabolites (cyclo-oxygenase inhibition). **(19-31)**

At the same time epidemiological studies, prospective studies and experimental studies suggest that higher tissue concentrations of omega-3 fatty acids and lower tissue concentrations of AA and LA is associated with decreased cancer incidence. **(32-36)** EPA gives rise to metabolites which have shown to inhibit cancer development, including mice with transplantable human breast cancer consisting of the genetic phenotype (HER-2/neu), which is the type that afflicts 15-40% of all human breast cancer patients. EPA has also been shown to compete with AA for activation via the lipoxygenase enzyme system, helping to reduce AA-derived metabolites that spur the growth and spread of cancer. **(4)**

Brain Function

Several recent studies have suggested that higher intake and blood levels of omega-3 fatty acids may help to reduce risk of age-related cognitive decline, dementia and Alzheimer's disease. **(44-47)** Several mechanisms have been proposed to explain how omega-3-fats can reduce nerve cell degeneration associated with these conditions.

Omega-3 fatty acids are known to provide anti-inflammatory effects due to their conversion to anti-inflammatory eicosanoids within the body. The eicosanoids formed from omega-3 fatty acids also improve blood flow by dilating vessels, and decreasing platelet stickiness (anti-thrombotic), and provide other benefits associated with cardiovascular health, such as improving endothelial function and lowering triglyceride blood levels. All of these effects are also associated with prevention of cognitive decline largely via preserved blood flow circulation to brain tissue (lower risk of cerebrovascular disease).

However, omega-3 fatty acids also play a direct role in nerve cell structure and function. Eicosapentaenoic acid (EPA) and docsahexaenoic acid (DHA) have been shown to improve the composition of nerve cell membranes, and stimulate the development, regeneration and function of nerve cells by stimulating synaptic plasticity and increasing neurotransmission, as well as increasing memory abilities. In short, long chain omega-3 fatty acids are structural components of neuronal and other cell membranes, affecting membrane fluidity, nerve transmission and nerve cell function in a positive way. They also modulate the production of eicosanoids and inflammatory cytokines and help preserve blood flow to the brain.

There is also the suggestion that oxidative stress (from oxygen and other free radicals), significantly contributes to neuronal damage seen in cases of cognitive impairment and Alzheimer's disease, by depleting the brain of vulnerable highly unsaturated fatty acids (e.g. EPA and DHA). Some researchers suggest that by replenishing brain cells with EPA and DHA via higher intake levels, individuals may help protect themselves against cognitive decline to a significant degree. **(48,49,50)**

Adding support to the epidemiological and experimental studies that suggest that omega-3 fatty acids can reduce risk of cognitive decline, the April 2007 issue of the *American Journal of Clinical Nutrition* contained the findings of two large prospective studies that evaluated intake of omega-3 fatty acids and subsequent risk of cognitive decline, dementia and Alzheimer's disease in older human subjects. Taken together, the findings of MA Beydoun et al (the *Atherosclerosis Risk in Communities Study*) and those of BM van Gelder et al (the *Zutphen Elderly Study*) indicate that a moderate intake of EPA and DHA were strongly associated with reduced risk of cognitive decline.

The ARIC Study

The *Atherosclerosis Risk in Communities Study* analyzed plasma fatty acids in cholesterol esters and phospholipids in whites residing in Minneapolis MN from 1987 through 1989. From 1990 through 1992 and from 1996 through 1998, 3 neurophysiological tests were administered. Effectively, this study studied the association between plasma fatty acids and cognitive decline in adults aged 50-65 years of age at baseline and conducted a subgroup analysis. A striking finding among the 2251 study subjects was that higher levels of omega-3 fatty acids were associated with reduced risk of decline in verbal fluency, particularly in hypertensive and dyslipedemic subjects, whose tissues are exposed to greater oxidative stress from these conditions. In contrast, the risk of global cognitive decline increased with elevated palmitic acid (a saturated fat) and in subjects with higher levels of arachidonic acid (an omega-6 fatty acid found in meat and dairy products). **(51)** It should be noted that palmitic acid is a saturated fat that is highly associated with thrombosis and the elevation of LDL-cholesterol, both of which can lead to atherosclerosis obstruction, increasing the tendency to develop dementia. **(52)**

The Zutphen Elderly Study

> In the *Zuthen Elderly Study* data on fish consumption of 210 male participants, who were aged 70-89 years of age in 1990, and data on cognitive functioning collected in 1990 and 1995 were assessed. The intake of EPA and DHA was calculated for each participant. Results showed that fish consumers had significantly less 5-year subsequent cognitive decline than did non fish consumers and a linear trend (dose-dependent trend) was observed for the relation between the intake of EPA and DHA and cognitive decline. More specifically, the results showed that elderly men who consumed an average of approximately 400 mg per day of omega-3 fatty acids from EPA and DHA had significantly less cognitive decline over the five year period than did those consuming an average of approximately 20 mg per day of omega-3 fatty acids.

At present the American Heart Association recommends the consumption of fish (preferably fatty fish) at least twice per week, a recommendation that is compatible with the results of the Zutphen Elderly Study. To achieve 400 mg per day of EPA and DHA, one would have to consume 6 servings per week of lean fish (average serving size 140 gm or about 5 ounces) or one serving per week of fatty fish, such as mackerel or herring. (10) One can also achieve this level of intake by consuming a mere 20 gm of Chinook salmon (less than one ounce) or 100 gm of cod (a little more than 3.5 ounces). As such, two to three meals of fish per week would supply approximately 380 mg of EPA/DHA per day, on average.

The *Zutphen Elderly Study* highlighted the fact that an average daily intake of 400 mg of EPA and DHA appears to be a significant threshold level at which a marked protective effect is observed. Some experts suggest that for people who are allergic to fish and/or shellfish and those who cannot or will not obtain sufficient intake of fish, that they consume 1000 mg per day of fish oil from supplementation (Connor WE, Connor SL, 2007). **(53)** A supplement containing fish oil and flaxseed oil may also be a consideration providing the total amount of EPA and DHA reaches a minimum threshold intake value of 400 mg per day. Health practitioners should bear this information and these dosage levels in mind when making recommendations about specific essential fatty acid supplement products to their patients in regards to optimizing brain function.

Summary

Although day-to-day choices around diet and nutritional supplements do not often feel like life and death decisions, the evidence suggests that nutritional components account for as much as 35% of all cancers **(37),** affect many risk factors for cardiovascular disease (e.g. cholesterol, triglycerides, homocysteine, blood pressure, eicosanoids) **(3),** and modulate inflammatory reactions associated with joint inflammation, autoimmune conditions and other inflammatory states. **(6,7)** Cancer and cardiovascular disease alone account for approximately 70% of deaths each year and many thousands of individuals suffer from arthritis and other inflammatory conditions that compromise quality of life

indices. In addition, recent evidence demonstrates that sufficient ingestion of omega-3 fatty acids is also required to inhibit the development of age-related cognitive decline. This article has drawn attention to the impact that polyunsaturated fatty acid consumption has on the eicosanoid cascade, and the effect that various eicosanoids have on these prevalent health conditions.

Based on the available evidence I recommend that health practitioners encourage their patients to limit their intake of foods rich in AA and LA, consume fish two to three times per week (more than this may increase risk of mercury toxicity), and supplement their diet daily with an essential fatty acid supplement containing 400 mg each of borage seed oil, flaxseed oil and fish oil (ensuring a 30/20% contribution of EPA/DHA, respectively). For general prevention and wellness individuals should consider two or three capsules per day. Individuals with certain health problems may require higher, more therapeutic doses.

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BREAKTHROUGH STUDY SUGGESTS ADDITIONAL WAY OMEGA-3 FATS MAY REDUCE BREAST CANCER RISK

James Meschino DC, MS, ND

The association between the intake of various fats and oils and the risk of breast cancer has been the subject of many studies. The body of evidence suggests a protective role of the omega-3 fatty

acids, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), the main components of fish oils.

As an example, in one human prospective study, researchers at the Italian National Cancer Institute followed a total of 4052 postmenopausal women for an average of 5.5 years. During this time 71 cases of invasive breast cancer were diagnosed. The cancer patients were matched with 141 controls. All study participants had blood samples drawn and red blood cell (erythrocyte) membranes were analyzed for their fatty acid content. It is well established that erythrocyte membranes are good biomarkers for not only dietary fat intake, but also for other dietary and hormonal factors.

The results of the study showed that women with DHA concentrations in the highest tertile had less than half the risk of breast cancer than did women in the lowest tertile. **(1)**

In vitro and animal studies have also shown that different fatty acids can influence breast cancer cell biology or mammary carcinogenesis. In general, cell culture and animal studies have shown a stimulatory effect of linoleic acid (LA), an (n-6) PUFA, on tumor cell growth in contrast to inhibitory effects provided by omega-3 fats such as docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA). In vitro experiments demonstrated increased proliferation of breast cancer cells as well as normal human mammary epithelial cells in the presence of LA, whereas diets enriched with omega-3 fatty acids EPA and DHA suppress both tumor growth and metastasis in nude mice (mice that have no thymus gland) bearing transplantable human breast cancers. **(2)**

Linoleic acid (LA) is found in high concentrations in common vegetable oils such as corn oil, sunflower seed oil, safflower seed oil, mixed vegetable oils and other oils. Exceptions would include olive oil and canola oil, which are high in oleic acid (a monounsaturated fat) and flaxseed oil (which is high in alpha-linolenic acid – an omega-3 fatty acid that the body can convert into EPA and DHA).

Omega-3 fats have demonstrated a number of physiological and biological effects through which they may reduce risk of breast cancer and other cancers. Some studies indicate that cell membrane-bound omega-3 fats are converted into prostaglandin hormones (prostaglandin series-3) that decrease the rate of cell division. Slowing the rate of cell division is associated with decreased cancer risk, and conversely, cells that replicate faster are more prone to generating cancerous mutations and demonstrate an inability to halt cell division to allow DNA repair enzymes to correct these mutations. As well, omega-3 fats have been shown to inhibit the conversion of arachidonic acid (an omega-6 fat) into prostaglandins (prostaglandin series-2) that increase the rate of cell division. **(3,4,5,6,7).**

Recent Studies Show That Omega-3 Fats Also Inhibit Activation of Epidermal Growth Factor Receptors

In addition to these protective effects Lisa D. Yee and fellow researchers (2005) revealed an additional way in which omega-3 fats may suppress the development of breast cancer in women who have genetic risk factors for this disease. **(2)** It is widely known that women, whose breast cells overexpress the human epidermal growth factor receptor HER-2/neu, are much more prone to breast cancer development than are women who have lower concentrations of these protein receptors on the cell membranes of their breast cells.

In healthy tissue, protein receptors transmit a signal to the cell to grow when a smaller protein, called a growth factor or ligand, binds to the receptor. (Ligands can be hormones, other growth factors, dietary agents etc). Ligand binding induces a change in the shape of the receptor, which causes receptors next to each other to connect. This change in shape results in the connection between two receptors on the outside of a cell acting like a domino falling over and starts a chain reaction throughout the cell that causes the cell to grow. Thus, all that is needed to signal a cell to grow is a connection between two receptors. Such a connection between receptors can occur spontaneously (without the growth factor binding to a receptor) if there are a lot of receptors on the surface of the cell, such that they frequently bump into each other (auto-stimulation). Thus, cells that have abnormally high levels of certain receptors can grow without receiving signals from external growth factors (such as hormones). Indeed, the most common abnormality in human cancers involves receptor overproduction. Such overproduction has been reported in breast,

prostate, ovarian, bladder and lung cancers. One particular family of receptors called the epidermal growth factor receptors (EGF/ErbB1-4) has been implicated more than any other. In approximately 30% of breast cancer patients, the ErbB2 receptor (HER-2) is overproduced, resulting in aggressive and uncontrolled growth of tumor cells. **(8,9)**

The study by LD Yee and fellow researchers is believed to be the first report to indicate a potent effect of fish oil in prolonging tumor latency and reducing tumor multiplicity in HER-2/neu overexpression-positive, estrogen receptor–negative breast cancer in a transgenic model of mammary tumorigenesis. This effect was seen by solely modulating dietary fat composition. These researchers showed that addition of omega-3 fats to the diet (25% calories from fish oil) of mice that were bred to overexpress HER-2 (and were known to have increased propensity for breast cancer) significantly reduced the development of breast cancer in these mice, compared to the mice fed a diet high in linoleic acid (25% calories from corn oil). The study revealed that by some mechanism(s) omega-3 fatty acids were able to suppress HER-2/neu signaling pathways involved in the pathogenesis of breast cancer. The researchers indicate that in this study omega-3 fats demonstrated a strong suppressive effect on HER-2/neu-positive breast cancer, suggesting a gene-nutrient interaction of critical importance for mammary carcinogenesis.

Although this study was performed on transgenic mice it provides further insight into potentially important mechanisms through which omega-3 fats may reduce risk of breast cancer, and may be especially significant to women who carry a high genetic risk for this disease. **(2)** The study by LD Yee and fellow researchers will undoubtedly prompt additional studies that will help clarify if omega-3 fats can suppress HER-2 activation in human breast cells and in human subjects who overexpress this receptor.

For more information on this or other related topics, visit Dr. Meschino's website at: http://www.renaisante.com/

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4. Reishi Mushroom Extract

REISHI MUSHROOM EXTRACT, IMMUNE SUPPORT AND CANCER

JAMES MESCHINO DC, MS, ND

Introduction

Reishi mushroom (Ganoderma lucidum) is called "the mushroom of immortality" in China and has been used in Oriental Medicine for over 2,000 years. [1,2] In recent years its active ingredients have been the subject of intensive research regarding their apparent ability to help prevent or treat certain types of cancer, aid in the treatment of liver disease, HIV infection, acute or recurrent herpetic infections, high blood pressure, chronic bronchitis, allergies and asthma, and favorably modulate immune function. [3] The reishi mushroom grows wild on decaying logs and tree stumps in the coastal provinces of China. The fruiting body of the mushroom is used medicinally. [4]

Active Constituents: Reishi mushrooms contain a number of active agents that are known to modulate function of the immune system in humans. The primary agents include:

1) Specific Polysaccharides - which occur in the form of Beta-D-glucans bound to amino acids. These agents are known to possess immune-modulating and anti-cancer properties. [3]
2) Triterpene compounds - known as ganoderic acids, which have been shown to lower blood pressure, reduce platelet stickiness and may decrease LDL-cholesterol. [5]
3) Other major active constituents – including sterols, coumarin and mannitol. [5]

Clinical Application and Mechanism of Action

1) Anti-Cancer Agent: Cancer studies in animals have shown a 50% tumor regression rate with reishi mushroom extract treatment (e.g., connective tissue cancer model in mice). [6] Reishi mushroom extract is used by some cancer surgeons in Japan to treat cancer patients and significant anti-tumor and immunostimulation effects have been noted in many of these cases. [7] Polysaccharides from reishi mushrooms and from other types of folk-medicinal funghi are patented in Japan for use as immunomodulators in the treatment of cancer. They are combined with chemo- and radiotherapy and have demonstrated an ability to reduce side effects, increase the efficacy of treatments, and are used to accelerate recovery from disease. [8,9]

   Studies from China have shown that reishi mushroom extract potentiates the tumoricidal capacity of macrophages and T-cells. [10,11] Reishi mushroom extract

is known to have other immune modulating effects and antioxidant properties as well [12,13,14,15,16]

Animal studies also show that the polysaccharide fraction of reishi mushrooms can induce apoptosis (programmed cell death of cancer cells) in leukemic cells and induce cellular differentiation in 40-45% of leukemic cells treated with reishi polysaccharides, demonstrating significant cancer treatment potential. These effects were primarily due to the increased secretion of anti-tumor cytokines (signaling agents) induced by reishi mushroom polysaccharides, namely TNF-alpha and IFN-gamma, and these two cytokines acted synergistically on the inhibition of leukemic-cell growth. [17] In a related experiment, the D-glucan polysaccharide fraction of reishi mushroom was shown to produce dramatic tumor regression in a mice sarcoma study. In many animals, complete tumor regression occurred in the group injected with beta-D glucan fractions within a 5-week period. The study by Y. Sone, et al, reported tumor inhibition rates of 90% and tumor regression in 75% of afflicted animals. [18]

A study in 2004 (Jiang) showed that reishi mushroom extract inhibited replication of human breast cancer cells by inhibiting nuclear factor-kappa beta (NF-kappa beta). NF-kappa beta is a transcription factor that is upreglated in cancer cells, prompting uncontrolled cellular proliferation. This study showed that reishi mushroom's anticancer effects may also include inhibiting the Ak/NF-kappa beta signaling system as reishi mushroom extract was shown to suppress phosphorylation of Akt on serine ^{473}, followed by down regulation of cyclin D1 and cyclin dependent kinase-4. [47]

2) Immune System Enhancement: (Bronchitis, Asthma, Allergies, Herpetic Conditions and HIV Infection) As noted above, reishi mushroom extract modulates many components of the immune system, which in part, account for its apparent anti-tumor properties. Chronic bronchitis in the elderly has been shown to respond favorably to treatment using a concentrated reishi mushroom product in a trial involving 2,000 cases in China. This study demonstrated a better than 60% success rate. After several months of treatment there was a noted rise in the levels of immunoglobulin A in the sputum. [10] Immunoglobulin A is the main immunoglobulin found in the respiratory tract. A deficiency is common in allergies,[19] systemic lupus and rheumatoid arthritis. [20] Reishi mushroom extract supplementation has been shown to help improve cases of asthma and allergies. Two constituents of reishi mushroom extract, oleic acid and cyclooctasulfur were shown to inhibit the release of histamine, which is likely how it benefits asthmatic and allergic patients. [10,21,22]

   A specific protein-bound polysaccharide component of reishi mushroom extract, known as GLhw-02, has been shown to possess potent anti-viral properties against herpes simplex virus type 1 and type-2 under experimental conditions. [23] A small human trial demonstrated that reishi mushroom extract reduced pain "dramatically" in two patients with post-herpetic neuralgia and in two other patients with severe pain due to herpes zoster infection (shingles, which is caused by a herpes virus). [24]

   Under experimental conditions, various ganoderic acids in reishi mushroom extract have been shown to be active anti-HIV agents, showing an ability to reduce viral replication by 50% at conservative doses. [25] Combined with other Oriental herbs, reishi is currently used in treatments of AIDS-related complex,

AIDS, and alone or in combination formulas to treat chronic fatigue syndrome. [26,27,28]

Finally, studies on male mice reveal that reishi mushroom extract was effective in enhancing the recovery of cellular immunocompetence after gamma-ray irradiation. Reishi mushroom supplementation significantly increased white blood cell count (leukocytes) and other parameters of immune function in these animals, [29] in a similar fashion as has been shown to occur with astragalus supplementation in patients treated with chemo- or radiation therapy.

Thus, the combination of astragalus and reishi mushroom extract represents an effective means of daily immune support and a therapeutic intervention for a large number of immune compromised states (e.g.,chronic fatigue, chronic bronchitis, herpes I and II recurrent infections, post-herpetic neuralgia, recurrent apthous ulcers or canker sores, the common cold, HIV infection, etc.) and for patients undergoing chemo-or radiation therapy.

3) Cardiovascular Health: (High Blood Pressure and Reduced Platelet Aggregation) Two human controlled studies revealed that reishi mushroom extract can reduce high blood pressure to a significant degree (systolic and diastolic), even in patients who had previously failed to respond to established anti-hypertensive medications. [30,31] Animal studies reveal that reishi mushroom extract reduces blood pressure through a central inhibition of sympathetic nerve activity, although it does not slow heart rate or induce a sedative effect in general. [32] Under experimental conditions, reishi mushroom extract has a mild to moderate effect on reducing platelet aggregation, which may further help to decrease risk of cardiovascular disease. [33] It has also been shown to increase endurance, blood flow to the brain and to improve oxygenation of cells. As such, it aids energy production on a cellular level, which may improve cardiovascular health and is used to boost memory and intellectual capacity in some cultures, [34] including success in a study of Alzheimer's patients. [35]
4) Liver Protective Effects: (Hepatoprotective Properties) Reishi is prescribed in China for the treatment of chronic and acute hepatitis. [36] Various ganoderic acids in reishi mushrooms have strong antihepatotoxic properties, [37] which under experimental conditions have been shown to protect liver cells from chemically-induced injury, including protection from the highly toxic and lethal substance, carbon tetrachloride. [38,39]

Dosage and Standardized Grade

1) Therapeutic Applications: Typically 250 mg, one to four times per day is used if taken as a single agent (standardized to 10-12.5% polysaccharide content). [40]

2) General Wellness: (immune, cardiovascular, liver support, etc.,) Consider 30-120 mg per day (standardized to 10-12.5% polysaccharide content).

Reishi mushroom extract should be taken with food or it may cause stomach upset and loose stools. [41]

Adverse Side Effects and Toxicity

Side effects are infrequent and include dizziness, dryness of the mouth, throat and nasal areas, stomach upset and loose stools. [41] Reishi mushroom contains agents that may be allergins to some patients, although allergic reactions are rare. [42] Note that there is no cross–sensitivity to the reishi mushroom if a person is allergic to the Button Mushroom or the commonly eaten white mushroom that is found in most grocery stores. [35] In general, reishi mushroom has been shown to be very non-toxic in animal toxicity studies and in humans, even when used at high therapeutic doses. [3]

Drug-Nutrient Interactions

When used at high therapeutic doses, reishi mushroom extract has the potential to potentiate (enhance) the effects of the following types of medications, and thus requires proper patient monitoring:

1) Antihypertensive medications [43,44]
2) Hypoglycemic medications [45]
3) Anticoagulant medications (e.g. warfarin, coumadin) [46]

Summary

In Asia, herbal agents such as reishi mushroom extract and astragalus have been used, alone or in combination formulas, to improve various parameters of immune function**,** treat a variety of immune-compromised states, increase the white blood cell count in patients recovering from drug toxicity, chemotherapy and radiation therapy, and provide effective complementary support for HIV and cancer patients. Recent experimental studies and intervention trials have helped to substantiate the longstanding traditional use of these natural agents and as well as to confirm their safety profile and lack of toxicity. As it is known that the human immune system becomes weaker and less efficient as we age, it seems reasonable that natural health practitioners would introduce measures to help their patients reduce or halt the age-related decline in immune system function; that appears to partially explain the rising cancer incidence that occurs as we age as well as our increased susceptibility to more virulent and life-threatening infections (e.g., Pneumonia). In this regard, antioxidant supplementation has shown a remarkable ability to forestall age-related changes to the immune system and to reverse many aspects of immune function in subjects showing some decreased immune capabilities. In concert with this intervention, the use of herbal compounds containing immune-modulating polysaccharides and other active constituents, such as reishi mushroom extract and astragalus, represent a potent means through which to further augment immune function and help patients prevent and treat a significant number of chronic degenerative diseases and reverse certain aspects of the aging process; especially in regards to immune, cardiovascular and liver function.

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5. Astragalus

Astragalus, Immune Support and Cancer

James Meschino DC, MS, ND

General Features

Astragalus has been used for at least 2000 years in China and continues to be widely used as a herb that is known to enhance function of the immune system and facilitates an increase in energy production within the heart muscle, in cases where certain forms of heart disease exist.[1,2,3,4,5] It is one of the most widely used herbs in Fu-zheng therapy − the use of herbs to augment the host defense mechanisms.[1,2,3,6] Astragalus is a herbaceous perennial with the root of the plant used for medicinal purposes.[7]

Active Constituents

These primarily include:

1. Triterpene glycosides (saponins): astragalosides, etc.
2. Polysaccharides: astragalans
3. Flavonoids[7,8,9,10,11]

Clinical Application and Mechanism of Action

1. **Immune Function** (The common cold and minimizing the effects of chemotherapy and radiation treatment)

   Astragalus is used as an immune stimulant to treat and help prevent the common cold.[4,8] It has also been used to reduce the side effects of chemotherapy and radiation treatment in human studies. A large clinical study of 572 cancer patients demonstrated that Astragalus supplementation was able to protect adrenal cortical function during radiation and chemotherapy treatment. It also helped to greatly minimize bone marrow depression and gastrointestinal side effects, such as nausea, vomiting and intestinal tract ulcerations in these patients.[6]

   In patients with very low white blood cell counts, as a side effect of drugs, radiation or chemotherapy, Astragalus supplementation has been shown to help significantly increase the number of circulating white blood cells, helping to restore normal function of the immune system in these severely immune-compromised patients.[12]

   The biological activity that can account for these outcomes is related the active constituents in Astragalus, primarily its triperpene glycosides and polysaccharide content, which have been shown to significantly increase the proliferation of lymphocytes,[3,12] enhance interferon and interleukin-2 production and activity- two powerful signalling agents that enhance the effectiveness of immune cells,[13,14,15,16] activate T cell blastogenesis,[17] increase T cell cytotoxicity,[2,17] enhance the secretion of the immune modifying chemical known as tumor necrosis factor (TNF),[9] enhance phagocytosis by immune cells,[18] increase natural killer cell cytotoxicity – the ability of these white blood cells to destroy developing cancer cells, viruses and other pathogens,[17,19] increase the activity of peritoneal macrophages,[18] and provide direct anti-viral effects.[20,21,22,23]

2. **Congestive Heart Failure and Angina Pectoris**
   The active constituents of Astragalus appear to provide an inotropic effect on the heart muscle, in a similar manner to hawthorn. An inotropic effect implies that these active ingredients in some way enhance the ability of the heart muscle to synthesize ATP energy, which is required for heart muscle contraction. In

congestive heart failure the heart muscle becomes weak, partly due to insufficient ATP production, and preliminary evidence suggests that Astragalus may help to improve these cases. Thus far, two small clinical trials have shown that patients with congestive heart failure demonstrate improvement in chest distress, dyspnea (shortness of breath), exercise tolerance and other parameters of cardiac function, when given Astragalus intravenously.[10,24]
Astragalus has also been used effectively in patients suffering from ischemic heart disease [25] and it has been shown to increase cardiac output in 20 patients with angina pectoris.[26]

3. **Anti-Cancer Effects**
   The immune- enhancing effects of Astragalus make this herb an interesting compound in terms of its potential in cancer treatment. A clinical study of 54 patients with small cell lung cancer were treated with regular medical interventions plus Traditional Chinese Medicine (including Astragalus). Increased survival was noted in comparison to the average survival statistics of conventional medicine alone.[27]

   Animal studies demonstrate quite strongly that Astragalus has the potential to prevent some cancers and has curative potential in others (e.g., renal cell carcinome model in mice).[28,29] Intensive research continues in an attempt to establish the true anti-tumor potential of Astragalus.

4. **Male Fertility**
   Astragalus has been shown to significantly increase the motility of human sperm in vitro.[30] This may be of value in the treatment of male infertility where poor sperm motility is a suspected factor. Note that L-carnitine and zinc supplementation have demonstrated similar capabilities (see details in this document under their individual headings.)

Dosage and Standardized Grade (2:1 powdered extract)

1. Common Cold – For general prevention consider 100-200 mg per day.
   During the preliminary stages of a cold consider up to 500 mg, three times daily if used as single agent.[31]
2. Radiation Treatment, Chemotherapy – Consider up to 500 mg, three times per day if taken as a single agent. (Consider combining Astragalus with reishi mushroom extract to minimize side effects of these treatments.) (Requires attending physician's approval)
3. Congestive Heart Failure, Angina Pectoris and Ischemic Heart Disease – No oral dose values have been established.
4. Decreased Sperm Motility Causing Infertilty – Dosage not established, however taking up to 500 mg, three times daily is considered to be safe.

Toxicity and Adverse Side Effects

There are no reported side effects or toxicity associated with the use of Astragalus at recommended doses.[31]

Drug-Nutrient Interactions

Immunosuppressive Medications – As Astragalus has been shown to enhance immune function, it may counter the efficacy of immnosuppressive drugs.[32,33]

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6, Vitamin E Succinate

VITAMIN E SUCCINATE: THE PREFERRED FORM OF VITAMIN E TO COMBAT BREAST, PROSTATE AND OTHER CANCERS

James Meschino, DC, MS, ND

In Brief

An abundance of research has proven that Vitamin E succinate is the most effective form of vitamin E in inducing differentiation, growth inhibition and programmed cell death (apoptosis) in cancer cells. A number of high profile, University-based researchers now suggest using this form of vitamin E (alone or in combination with other supplements) to enhance the efficacy of medical cancer treatments (in conjunction with some chemotherapy drugs, ionizing radiation, hyperthermia and biological modifiers) and as a means to decrease the adverse side effects of standard cancer treatment.

As vitamin E succinate has been shown to possess unique and markedly superior biological properties to prevent and combat cancer, when compared with other forms of vitamin E (alpha-tocopherol, alpha-tocopherol acetate and nicotinate, gamma-tocopherol, as well as tocotrienols), health practitioners should strongly consider recommending a high potency multiple vitamin supplement that contains this form of vitamin E (e.g. 400 IU vitamin E succinate) for health promotion purposes. As well, higher doses of vitamin E succinate appear to be appropriate as part of adjunctive nutritional management in select cancer patients. This article reviews the

research pertaining to the anti-cancer properties of vitamin E succinate for the purpose of educating health practitioners about this important and clinically relevant subject.

Introduction

Recent studies suggest that vitamin E succinate (VES) is the most potent antitumor analogue of vitamin E. (1) Although many individuals rely on vitamin E supplementation for its antioxidant properties, vitamin E taken in the form of vitamin E succinate provides additional anti-cancer benefits, according to numerous in vitro and in vivo studies. (2,3)

More specifically, vitamin E succinate has been shown to induce programmed cell death (apoptosis) of human prostate cancer cells, human breast cancer cells and other human cancer cells, including leukemia cells, and gastric cancer cells. Of note is the fact that VES has been shown to be exerts its antitumor influences on cancer cells, but is nontoxic to normal cells. These impressive findings have spurred on researchers to determine the mechanism(s) of action through which VES combats cancer development, inhibits growth of cancer cells and induces apoptosis of certain cancer cells (3).

Vitamin E Succinate and Prostate Cancer

A number of studies have shown that VES inhibits the growth and/or induces apoptosis of various forms of human prostate cancer cells. Researchers have identified various physiological effects through which VES induces these desirable outcomes.

The study by Y. Zhang and fellow researchers demonstrated that VES suppressed the expression of androgen receptors, meaning that fewer androgen receptors are present on the cells surface in the presence of VES. This is of great clinical significance because the androgen receptor (receptors for testosterone, dihydrotestosterone and other androgen hormones) is required for the development of both the normal prostate gland and prostate cancer**.** In the early stages of prostate cancer**,** most cancer cells are androgen-dependent and highly sensitive to anti-androgens (drugs such as hydroxyflutamide). As such, most prostate cancer cells grow and proliferate in response to testosterone and other androgens binding to their androgen receptors. Thus, if VES prevents prostate cancer cells from expressing androgen receptors, then these cells are less likely to flourish in the presence of testosterone and other androgens present in the male body.

Traditionally, the treatment of prostate cancer has included androgen ablation (e.g. castration to reduce circulating levels of testosterone from the testes, and/or anti-androgen drugs to reduce stimulation of androgen receptors by circulating testosterone and other androgen hormones). Interestingly, prostate cancer usually recurs after a few years of androgen ablative treatment, and most cancer cells become androgen-independent, rendering antiandrogen therapy ineffective as the disease becomes more advanced.

However, Zhang and fellow researchers were able to show that the combination of VES and hydroxyflutamide provided significantly greater growth inhibition of androgen dependent prostate cancer cells (LNCaP) than did hydroxyflutamide on its own, which only mildly inhibited growth of LNCaP cells when administered without VES.

The LNCaP cell line is derived from lymph node prostate cancer metastasis, and is one of the best *in vitro* models for human prostate cancer studies, as it represents a hormone-refractory prostate carcinoma, and its growth is responsive to testosterone and other androgens. In addition, LNCaP cells express a functional mutant androgen receptor, and produce PSA, which is a sensitive and specific tumor marker for prostate cancer screening and assessment

Zhang and fellow researchers also showed that VES decreases intracellular and secreted levels of prostate-specific antigen (PSA) in LNCaP cells, which was cultured either in normal serum or in androgen-stimulated conditions.

Other researchers have shown that VES can also induce programmed cell death (apoptosis) of human prostate cancer cells, in addition to its ability to down-regulate expression of androgen receptors.

For example, it has been suggested that VES could inhibit the proliferation of prostate cancer cells by arresting DNA synthesis, or by stimulating transforming growth factor beta (TGF-B), which is an intracellular anti-proliferative agent. (4)

Most recently Chung-Wai Shiau and fellow researchers showed that VES induced programmed cell death of LNCaP prostate cancer cells by blocking the action of specific cellular proteins that are required for cell proliferation by cancer cells. More specifically, these researchers showed that VES blocked the action of a cellular protein known as Bcl-xL, which is found at abnormally high levels in cancer cells. Shiau and fellow researchers determined that VES lodges in a groove in the structure of the Bcl-xL protein, disabling it, (disrupting the binding of Bak BH3 peptide to Bcl-xL and Bcl-2, which otherwise affect gene regulation in such a way as to encourage cell replication)

By disabling the Bcl-xL protein VES essentially sets into a motion a chain of events that triggers programmed cell death of prostate cancer cells (via caspase-dependent apoptosis). Of note is the fact that other forms of vitamin E are unable to perform this task. Only vitamin E succinate has been shown to appreciably induce programmed cell death of various types of cancer cells, most noteworthy, prostate, breast and gastric cancer cells. These researchers were also clear to point out that VES does not damage the function of healthy cells, but rather is able to selectively induce apoptosis of cancer cells, primarily. (5)

In another study, K Israel and fellow researchers showed that VES also induces apoptosis in certain prostate cells that are highly resistant to apoptosis. Certain cancer cells are known to resist programmed cell death triggered by stimulation of their Fas-receptors. When stimulated by a ligand (binding agent), usually from certain immune cells, the Fas-receptor stimulates the release of secondary messengers within the cell that trigger programmed cell death. As such, this in one way in which immune cells are designed to identify and kill cancer cells. However, some cancer cells are Fas-resistant in that they do not undergo apoptosis upon stimulation of their Fas-receptors. This has been shown to be the case with certain human prostate cancer cells. The study by K Israel et al, showed that vitamin E succinate could enhance the Fas-apoptosis pathway and lead to increased programmed cell death of various types of human prostate cancer cells, but did not affect normal prostate cells (6). A further explanation of the Fas-apoptosis system is provided in the following breast cancer section of this paper

Vitamin E Succinate and Breast Cancer

Vitamin E succinate has also been shown to induce programmed cell death (apoptosis) and inhibit proliferation of human breast cancer cells in numerous studies. In separate studies MP Malfa and fellow researchers and R. Schindler and fellow researchers demonstrated that VES inhibited the growth of certain breast cancer cells and induced apoptosis in other types of human breast cancer cells (MDA-MB-231 cells). (1,7)

These researchers showed that one way that VES induces apoptosis and growth inhibition is by inhibiting the release of vascular endothelial growth factor-A (VEGF). (1,7) VEGF is a protein required for angiogenesis (formation of new blood vessels) by tumor cells. The development and progression of solid tumors require rapid and persistent growth of new blood vessels to supply the growing tumor with nutrients and oxygen. Interestingly, increased vascular proliferation has been shown to correlate with a higher incidence of metastases and a worse prognosis. As mentioned, tumor neoangiogenesis is induced by angiogenesis-promoting growth factors (e.g. VEGF) produced by the tumor or nonmalignant tumor stromal cells (supporting connective tissue network cells in the vicinity of the tumor). The VEGF gene encodes differently spliced mRNAs from which pre-VEGF proteins are transcribed. These proteins are secreted directly from the cells after signal sequence cleavage in the rough endoplasmatic reticulum and glycosylation in the Golgi apparatus. Thus, VEGF is not stored in cells, and synthesis correlates with release. VEGF expression is regulated by transcription factors as well as by mechanisms that control mRNA stability.

Earlier studies showed that the soy isoflavone genistein inhibited angiogenesis, VEGF expression and release by various human breast cancer cells, which may partly account for the reduced incidence of breast cancer in populations consuming soy products as a main dietary staple. Of

note is the fact that R Schindler and fellow researchers demonstrated that certain flavonoids (naringin – from citrus fruits; rutin – a constituent of cranberries), vitamin E succinate, apigenin (found in apple skins, citrus fruits, celery roots) and genistein, were all among naturally-occurring agents shown to inhibit release of VEGF and inhibit angiogenesis of human breast cancer cells. However, it should be noted that alpha-tocopherol and gamma-tocopherol (other popular forms of vitamin E) did not show significant anti-angiogenesis effects compared to that exhibited by vitamin E succinate. Interestingly, the drug lovastatin showed an anti-angiogenesis effect that was slightly inferior to that of VES, narigin and rutin and slightly greater than apigenin and genistein. (7)

Other researchers have shown that VES induces apoptosis of human breast cancer cells by affecting cell membrane receptors, such as certain members of the epithelial growth factor family. In healthy tissue, protein receptors transmit a signal to the cell to grow when a smaller protein, called a growth factor or ligand, binds to the receptor. Ligand binding induces a change in the shape of the receptor, which causes receptors next to each other to connect. This change in shape and resulting connection between two receptors on the outside of a cell acts like a domino falling over and starts a chain reaction throughout the cell that causes the cell to grow. Thus, all that is needed to signal a cell to grow is a connection between two receptors. Such a connection between receptors can occur spontaneously (without the growth factor binding to a receptor) if there are a lot of receptors on the surface of the cell, such that they frequently bump into each other (auto-stimulation). Thus, cells that have abnormally high levels of receptors can grow without receiving signals from external growth factors (such as hormones). Indeed, the most common abnormality in human cancers involves receptor overproduction. Such overproduction has been reported in breast, prostate, ovarian, bladder and lung cancers. One particular family of receptors called the epidermal growth factor receptors (EGF/ErbB1-4) has been implicated more than any other. In approximately 30% of breast cancer patients, the ErbB2 receptor is overproduced, resulting in aggressive and uncontrolled growth of tumor cells.

It is well documented that women whose breast cells overexpress the erbB2 receptor (a member of the Epithelial Growth Factor Receptor Family) on their cell surface are at higher risk for breast cancer development. Additionally, their overexpression is associated with the estrogen receptor negative form of the disease, which is known to be the most clinically aggressive form of this disease. (8,9)

Studies by XF Wang and fellow researchers demonstrated that vitamin E succinate was able to induce programmed cell death (apoptosis) in human breast cancer cells that exhibited high levels of erbB2 expression.

A study by W Yu and fellow researchers showed that vitamin E succinate could also induce apoptosis in human breast cancer cells that Fas- resistant. (10). Fas is an immunology protein that initiates programmed cell death. Apoptosis is important in cell-mediated immune response, autoimmune tolerance, and cancer control. Fas is a surface receptor that is expressed throughout the body. The Fas ligand is expressed mostly on activated T cells, natural killer cells, and microglia, but is sometimes found on other cells of the immune system. When a cell expressing Fas encounters another cell expressing Fas ligand, the receptor is stimulated in such a way as to transmit a message to secondary protein messengers within the cytoplasm (signal transduction) of the cell. In turn this activates a series of events that lead to cell death (apoptosis). Certain breast cancer cells are resistant to the apoptosis-inducing effects of Fas, such as MCF-7, MDA-MB-231 and MDA-MB-435 human breast cancer cells. (11) The study by T. Yu et al, showed that vitamin E succinate was able to convert Fas-resistant human breast cancer cells to Fas-sensitive phenotype, allowing apoptosis to proceed. These findings are important as they suggest another mechanism through which VES may prevent breast cancer development and/or inhibit the further growth and proliferation of existing breast cancer cells.

Other Mechanisms Through Which Vitamin E Succinate May Prevent Cancer

Reports from K Wu and fellow researchers indicate that vitamin E succinate also induces apoptosis of cancer cells by first stimulating production of transforming growth factor-B, which in turn, increases kinase activity of another protein messenger known as c-Jun N-terminal kinase

(JNK), followed by phosphorylation of c-Jun, and finally activated c-Jun triggers apoptosis. K Wu et, al have shown this apoptosis pathway to exist in human gastric cancer cells, but it may also occur in many other cancer cells as well. (3) Other researchers have confirmed that vitamin E succinate induces apoptosis through stimulation of the c-Jun messenger system.(12,13) In addition, K Wu et al, report that vitamin E succinate also induces apoptosis of certain cancer cells by enhancing activity of the Fas-apoptosis system (sometimes called the Fas-death receptor, although other death receptors are also present on the cell surface), as described in the previous sections.(3).

Summary

In regards to vitamin E supplementation the current evidence suggest that health practitioners should strongly consider recommending supplements containing vitamin E succinate (alpha tocopherol succinate or alpha tocopheryl succinate), as part of a high potency multiple vitamin/mineral supplement or as a stand alone supplement, as part of a comprehensive program to reduce cancer risk. In this regard the evidence suggests that vitamin E succinate is superior to other available forms of vitamin E and related tocotrienols. Many experts suggest a daily dosage of 400 IU per day of vitamin E succinate for general health promotion and disease prevention. As an adjunct to cancer treatment some practitioners use a total daily dosage of 800-1600 IU (up to 2500 IU of vitamin E– Folkers and Lockwood, Molecular Aspects Of Medicine, 1984), in conjunction with high doses of other antioxidants and natural cancer-fighting herbs and phytonutrients. Preliminary human trials using these nutritional supplements have shown impressive results in reducing cancer progression, preventing metastasis and reducing side effects of chemotherapy and radiation. (14)

In the review presented by KN Prasad and fellow researchers (Center for Vitamins and Cancer Research, Department of Radiology, University of Colorado Health Sciences - 2003) the authors state, “Since it has been demonstrated that vitamin E succinate (alpha tocopheryl succinate) in combination with dietary micronutrients (retonoic acid, vitamin C and carotenoids) is more effective in reducing the proliferation of tumor cells in culture than the individual agents and in enhancing the effect of some chemotherapeutic agents on these cells, we have recommended the use of alpha tocopheryl succinate in combination with dietary micronutrients as an adjunct to standard therapy in the treatment of cancer”. These authors have provided an excellent review of the anti-cancer properties of vitamin E succinate for health practitioners who are interested in further reading on this subject. (15)

To sum up, abundant evidence indicates that vitamin E succinate possesses unique biological properties that enable it to inhibit or slow the replication of human cancer cells and/or induce apoptosis of many types of cancer cells, most notably prostate and breast cancer. In this regard vitamin E succinate has been shown to be superior to other forms of vitamin E. As cancer accounts for approximately 20% of all deaths in North America, this information should be of importance to health practitioners who routinely recommend multiple vitamins and/or vitamin E supplements to their patients for the purpose of reducing risk of degenerative disease, and/or who advise patients on evidence-based adjunctive nutritional therapies that may be helpful to prevent recurrences or metastasis of cancer in patients that have already been affected by this disease.

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7. Selenium

SELENIUM SUPPLEMENTATION AND CANCER WITH EMPHASIS ON THE PREVENTION OF COLON CANCER

Dr. James Meschino, D.C., M.S., N.D.

INTRODUCTION

Selenium is an essential nutrient for humans; it fulfills the physiological requirements for more than 13 human enzymes and proteins. (1) Its most well known function relates to the activation of the enzyme, glutathione peroxidase, which accounts for selenium's antioxidant role in the reduction of hydrogen peroxide to water and organic hydroxyperoxides to alcohol. The form of selenium in all selenoproteins is an amino acid, L-selenocysteine. Both inorganic (e.g., selenite and selenate) and organic (seleno amino acids) forms of selenium have shown impressive cancer – chemopreventive effects in humans and in animal models. (2)

The nutritional requirement for selenium for the synthesis of L-selenocysteine is considered to be 55 micrograms per day. However, selenium consumed as a dietary supplement, beyond levels attainable from food, at up to 200 mcgs per day is associated with reduced incidence of lung, colorectal and prostate cancer in humans.

Animal studies also strongly indicate that supraphysiological levels of selenium significantly reduces cancer incidence in various test models. [1]

Many cancer researchers now propose that the level of selenium intake required to reduce cancer risk exceeds the intake level (55 mcgs), which is required to maximize the synthesis of L-selenocysteine, upon which the RDA is based.

At supraphysiological levels of intake, selenium has been shown to exhibit a number of anti-cancer properties beyond the activation of the antioxidant enzyme, glutathione peroxidase. These general mechanisms include:

1. Apoptosis – programmed cell death of cancer cells
2. Growth-inhibitory effects of cancer cells
3. Induction of p53 tumor suppressor gene, which gives rise to proteins that arrest cell cycle division when DNA mutations occur, allowing DNA repair enzymes to correct the error.
4. Protects against DNA damage
5. Anti-promotional agent, discouraging cancer cell division.
6. The induction of apoptosis (a very important chemopreventive function) is detectable towards the upper limit of plasma selenium concentrations (approximately 5 micromoles per liter) [3,4,5]

The present view is that selenium metabolites (i.e., methylated forms of selenium), rather than simply the saturation of L-selenocysteine synthesis alone, are key factors in selenium's cancer preventive effects. [5] At physiological levels of intake, dietary selenium (inorganic and organic forms) is reduced to hydrogen selenide (H_2 Se). From H_2 Se selenium is phosphorylated and incorporated into proteins such as L-selenocysteine.

During periods of supraphysiological intake (supplementation), once L-selenocysteine levels are saturated, H_2 Se is rapidly methylated to dimethyl selenide and other methylated forms including the monomethylated form, and trimethylselenonium. Experimental studies highlight the fact that these methylated forms of selenium

(metabolites) possess direct and indirect chemopreventive effects that are not attainable by the action of glutathione peroxidase acting alone.

Thus, selenium supplementation (100-400 mcg per day) increases the levels of methylated selenium metabolites that appear to account for many of selenium's cancer preventive effects. [1,5]

Note that under toxic conditions, this methylation metabolic pathway becomes rate limiting, and dimethyl selenide is eliminated by pulmonary excretion, giving rise to garlic-breath. [1]

As reported by Combs, B., et al, selenium plasma levels of approximately 120 ng/ml (1.5 umol/ml) may be optimal for cancer prevention in general. The most recent estimates suggest that women require a minimum of 96 micrograms per day and men require at least 120 micrograms per day to support plasma levels at 120 ug/ml for Americans. These levels are 175% and 218%, respectively, of the revised RDA. [6]

In terms of specific cancers that may be prevented through selenium supplementation, colorectal cancer has received much attention in both human and animal studies and is the focus of this review. [3]

DIET AND COLON CANCER

In North America, colon cancer is the second leading cause of cancer death after lung cancer. Cancer researchers Doll and Peto suggest that up to 90% of colon cancer in this part of the world may be avoidable by altering the diet. In fact less than 5% of all colon and rectal cancers can be attributed to known causes, such as family history, colon polyps that run in families (familial polyposis) or as a sequel to inflammatory bowel diseases, such or ulcerative colitis. Thus, colon and rectal cancers are considered to be preventable in the vast majority of cases. The 90% variation rate in colon and rectal cancers that exists among countries is considered to be largely due to specific dietary factors. [7] In the U.S., 137,000 new cases and 57,000 deaths from large bowel cancer were expected in the year 2000. [3]

The body of evidence is highly suggestive that a high fat, low fiber diet increases the risk of colon cancer whereas a low fat, high fiber diet tends to decrease the risk. In

particular, wheat bran fiber has demonstrated important protective features as an anti-colon cancer agent. Studies also link higher intakes of vitamin E, vitamin C and calcium with a lower incidence of colon cancer. As well, low blood levels of beta-carotene are consistently associated with an increased risk of developing colon cancer. [8,9,10,11,12,13,14,15,16]

A lesser-appreciated fact among health professionals is the extensive research that suggests that the mineral selenium may be a highly protective micronutrient in the prevention of colon and rectal cancers.

Animal and Human Studies Using Selenium

Selenium is an essential trace mineral found in soils and crops and has been shown to prevent chemically induced colorectal cancer in animals.

In one study rats were fed a cancer-causing agent know to cause colon cancer. The rats whose diets were supplemented with selenium had a tumor incidence of only 3% whereas the rats that received no selenium supplementation had a 29% tumor incidence.

Other animal studies have shown that selenium supplementation reduces the incidence of intestinal tumors by 50% compared with rats given the cancer-causing agent without selenium supplementation. [17,18,19,20]

Human observation studies are equally as impressive. For example areas with low soil and crop selenium content have higher rates for colon and rectal cancers. Other studies demonstrate that lower blood levels of selenium are associated with an increase risk of developing colon cancer. [21,22,23,24,25]

In a study of U.S. veterans, blood levels of selenium were measured, in subjects with colorectal cancer and those free from colon cancer. The results demonstrated that subjects with blood selenium levels below 128 micrograms per liter were 4.2 times more likely to have one or more cancerous polyps. [22]

In a clinical trial using selenium to reduce risk of skin cancer 1,312 subjects were given either 200 micrograms of selenium or a placebo. Although selenium was not found to be protective against skin cancer, subjects taking the selenium experienced 58% reduction in colon and rectal cancers compared with subjects taking the placebo pill. [24]

More recently a study by Dr. Mark Russo and associates at The University of North Carolina (Chapel Hill) further supported a role for selenium in the prevention of colon and rectal cancers. In their study patients who were referred for a colonoscopy assessment also had blood tests performed. As reported by these authors, lower blood levels of selenium were associated with multiple cancerous lessons in the colon. The average blood level for patients with cancerous lessons was 107 micrograms per liter vs. 120 micrograms per liter for the cancer free subjects. [25]

The authors conclude that this data support a protective effect of selenium against colon and rectal cancers after adjustments for possible confounding factors such as smoking, alcohol intake, use of dandruff shampoo (which contains selenium), vitamin E intake, vitamin C intake, iron intake, fat intake, and fiber intake. "Our results demonstrate that individuals with high plasma (blood) selenium levels are at a decreased risk for colorectal adenomas (cancerous lesions)". An increase of 30 micrograms per liter in

blood selenium level was associated with a 50% reduction in risk of colon cancer lesions. [25]

The Protective Effect of Selenium

As noted, there are several ways that selenium is thought to reduce cancer risk. Selenium enhances antioxidant defenses by increasing activity and levels of the powerful antioxidant enzyme known as Glutathione Peroxidase.

Glutathione Peroxidase is considered a strong anticancer agent within the body.

Selenium supplementation also decreases the formation of certain cancer permissive eicosanoids, of the prostaglandin E2 series. Prostaglandin E2 has been shown to increase the rate of cell division of various cancer cells. Animal studies have demonstrated these effects quite extensively.

Selenium metabolism itself may initiate changes that lead to programmed cell death of cancer cells and pre-cancerous cells. [26,27,31,32] It is important to note, that other human studies by Dr. Willett, and Dr. Salonen, also reported, a 2 and 3 fold increased risk for colon and intestinal cancer respectively, in patients presenting with low blood selenium levels when compared to patients with higher blood levels of selenium. [28,29] In line with these observations is the emerging evidence that selenium metabolites (methylated forms) exert a broad range of anti-cancer influences on the cell, including colonic epithelial cells, and that higher levels of selenium intake is required to produce protective levels of these methylated selenium compounds [3,4,5].

Current Trends and Practical Application

The average intake of selenium from food sources is shown to be between 50-70 micrograms daily. Is it adequate? The evidence suggesting that selenium can reduce the risk of certain cancers indicates that ingesting additional selenium from a supplement may be a prudent strategy. Many health conscious physicians now recommend a supplement containing 50-200 micrograms of selenium daily as part of a preventative health strategy. [30,33]

As for safety, toxicity of selenium begins at doses starting at 1,000 micrograms per day, but doses as high as 2,000 mcgs per day have been shown to be non toxic is some individuals. [6,30,33,34] Most clinical applications have used 100-500 mcg of selenium per day [28, 34]

Ingesting a safe level of selenium from a vitamin and mineral supplement may have other health benefits as well. For instance, the National Research Council stated that "A large accumulation of evidence indicates that supplementation of the diet or drinking water with selenium protects against tumors induced by a wide variety of chemical carcinogens" in animal studies. Significant protection was demonstrated against the development of breast, colon, liver, and skin cancers. [35]

In humans, epidemiological studies have linked lower selenium intake to a higher incidence of leukemia, and cancers of the colon, rectum, pancreas, breast, ovary, prostate, bladder, skin and (in the male) lung. [36] As a result of these studies a number of clinical

trials are underway which are testing selenium as a cancer preventive agent. In the Linxian China study, the combination of selenium, vitamin E and beta-carotene was shown to reduce the incidence of stomach and esophageal cancers by 16 and 17 percent, respectively, in a very high-risk population. [37]

Studies using selenium have also suggested that it plays a role in strengthening the immune system, reducing inflammatory conditions and reducing risk of heart disease and stroke. [38,39,40]

In conclusion, the mineral selenium demonstrates very promising chemopreventive capabilities as reported in a number of animal and human studies. At recommended supplemental doses (100-500 micrograms per day) it is extremely non-toxic. Moreover, it appears that selenium supplementation may be the only viable means to provide sufficient concentrations to enable the body to generate protective levels of methylated selenium metabolites, which are now considered to be important bioactive agents that account for much of selenium's anti-cancer effects.

It may be prudent to initiate a selenium supplementation early in life. As a rule of thumb selenium supplementation at 1.5 micrograms per pound of body weight is reported to be a safe intake level that can be applied to children and young adults. [34]

The best food sources of selenium include wheat germ, oats, whole wheat bread, bran, tuna, swordfish, oysters, turnips, barley, garlic, brown rice and red Swiss chard. [30,34]

As cancer of the colon and rectum now affects one in twenty people in the population at some point in their lifetime, using pro-active nutrition and supplementation strategies to defend against this disease should be encouraged by primary health care professionals.

In addition to a low fat, high fiber diet, the body of evidence is highly suggestive that certain micronutrients, including vitamin C, vitamin E, beta-carotene, calcium, vitamin D and selenium, exert significant chemopreventive effects in regards to colon and rectal cancers. As a matter of public health policy, health practitioners should be engaged in the process of educating patients as to the much under rated relationship between nutrition and cancer. This is especially true in regards to colorectal cancers, as up to 90% of these cases are considered to be preventable through better nutrition and lifestyle practices.

For more information on this or other related topics, go to Dr. Meschino's website at: www.renaisante.com.

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8. Vitamin C

Vitamin C and Cancer

James Meschino DC, MS, ND

Vitamin C has been shown to be an important agent in the adjunct management of cancer as in impacts a number of biological targets, which include:

Inhibition of NF-kappa beta - Vitamin C has been shown to inactivate nuclear factor-κB in endothelial cells during the inflammation process, independently of its antioxidant activity. As NF-kappa beta is involved in the inflammatory process as well as the proliferation of cancer cells, the anti-inflammatory activity of vitamin C may be mediated by multi-factorial mechanisms, which are not necessarily associated only with its intrinsic antioxidant activity.

Maintains Gap Junctions - As well, cell-to-cell communication through gap junction channels is essential for maintaining homeostatic balance through modulation of cell proliferation and differentiation in multicellular organisms. Inhibition of cell-to-cell communication is strongly related to the carcinogenic process, particularly to tumor promotion. Hydrogen peroxide, a well-known tumor promoter, also inhibits GJIC. Recently, we reported that vitamin C exerts protective effects against the disruption of GJIC by hydrogen peroxide.

An analysis by Rosenkranz et al, found that the inhibition of GJIC is strongly linked to the carcinogenic process, a biological phenomenon that may involve the inflammatory process, and to developmental effects in rodents. Integration of the analysis also suggests that the inhibition of GJIC is involved in nongenotoxic cancer induction and tumor promotion. Therefore, some experts suggest that the chemopreventive effects of vitamin C in carcinogenesis may be linked to the protective effects of vitamin C against epigenetic mechanisms, such as the inflammation and inhibition of GJIC, as well as to antioxidant activities. **(1)**

Induces Apoptosis In Cancer Cells – Vitamin C has been shown to upregulate caspase 3 enzyme in certain cancer cells, increasing apoptosis. Higher concentrations of vitamin C induce apoptotic cell death in various tumor cell lines including oral squamous cell carcinoma and salivary gland tumor cell lines **(2).** Kim et al showed that in human colon cancer cells vitamin C encouraged the phosphorylation of pro-apoptotic protein, Bad In addition, they also found that the vitamin C increased expression of pro-apoptotic protein, Bax and tumor suppressor gene product, p53, which usually resulted in apoptosis, in a time dependent manner. Moreover, they reported that vitamin C treatment disrupts mitochondrial membrane potential in HCT-8, human colon cancer cell line. Taken together, vitamin C has been shown to induce redox imbalance, resulting in apoptosis in human colon cancer cells by both endoplasmic reticulum stress and Bad/p53/Bax-mediated activation of mitochondrial pathway. **(3)**

Vitamin C and Immune Function - Vitamin C concentrations in the plasma and leukocytes rapidly decline during infections and stress. Supplementation of vitamin C

was found to improve components of the human immune system such as antimicrobial and natural killer cell activities, lymphocyte proliferation, chemotaxis, and delayed-type hypersensitivity. Vitamin C contributes to maintaining the redox integrity of cells and thereby protects them against reactive oxygen species generated during the respiratory burst and in the inflammatory response. **(4)**
Supplemented dosages of vitamin C have also been shown to increase interferon levels, a factor that is important to the upregulation of death receptors on cancer cell, which may partially explain vitamin C-mediated apoptosis. **(5)**

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9. Vitamin D

Vitamin D: An Overview OF Health Benefits

James Meschino DC, MS, ND

Vitamin D acts in the body as both a vitamin and a hormone, exerting a powerful influence on maintaining bone density and preventing vital steps in the development of breast, prostate and colon cancers (and possibly others). There is also evidence to suggest that more optimal vitamin D levels can reduce the risk of multiple sclerosis by affecting immune system function.

Throughout younger adult life, supplementation of 400 IU of vitamin D per day from a high-potency multivitamin/mineral, combined with the normal amounts most people acquire from fortified dairy products and fish, is usually adequate to maintain blood levels of vitamin D in a range that is associated with healthy bone development; the prevention of breast, prostate and colon cancers; and multiple sclerosis, according to available studies. However, by age 45-50, the enzyme in the kidney (alpha-hydroxylase)

that converts vitamin D into its most active form (1,25 dihydroxy vitamin D, also known as calcitriol) becomes less active. This results in decreased synthesis of calcitriol; thus, tissues that rely on its health-promoting influence are left to feel the effects of insufficiency.

Studies strongly suggest that the age-related decline in calcitriol synthesis in the kidneys is a major contributing factor to the development of osteoporosis in women and men older than age 50, as well as the age-related increase in risk of breast, prostate and colon cancers that is common in North America and many industrialized countries. However, studies also indicate that individuals older than age 45 can compensate for the decline in calcitriol synthesis by raising their blood levels of the less potent form: 25-hydroxy vitamin D.

Abundant evidence exists suggesting that adults who maintain blood levels of this form of vitamin D in the range of 85-120 ng/mL (nanograms per milliliter) have a significantly lower risk of developing osteoporosis, breast cancer, ovarian cancer, prostate cancer, colon cancer and multiple sclerosis. Studies also show that to ensure blood levels of 25-hydroxy vitamin D are within this protective range, it is important to increase vitamin D supplementation to 800-1,000 IU per day after the age of 45. Until then, 400 IU per day from a high-potency multivitamin/mineral combination is adequate to meet vitamin D needs in most cases. However, I strongly advise the addition of a daily supplement that contains 400-600 IU of vitamin D after age 45, in a formulation that also provides an additional 500-700 mg of calcium. (It is perfectly acceptable if the product also contains some additional magnesium, zinc and other bone support nutrients.)

Vitamin D and Bone Density

Vitamin D is required at all stages in life to optimize calcium absorption from the intestinal tract, which, in turn, helps increase bone mineral density during the developmental years and helps prevent the loss of calcium from bone after age 50. The increased deposition of calcium into our bones that occurs at an optimal rate up to age 24, and the prevention of calcium being leached from bones after age 50, are both essential aspects related to the prevention of osteoporosis.

Unfortunately, vitamin D deficiency is widespread in North America, especially in the more northern regions (above 42 degrees latitude), where sunlight intensity is very limited between October and May. Older individuals have also been shown to be at very high risk for vitamin D deficiency, due to poor intake of vitamin D-containing foods, reduced sunlight exposure and reduced conversion of 25-hydroxy vitamin D into calcitriol (which is twice as potent as 25-hydroxy vitamin D with respect to influence on calcium metabolism, our bones and other tissues). Vitamin D deficiency correlates with a blood level of 25-hydroxy vitamin D below 20 ng/mL. Optimal levels are considered to be in the range of 85-120 ng/mL. It is this range of vitamin D status that is most strongly associated with a reduced risk of osteoporosis, many cancers and multiple sclerosis. Unfortunately, a great number of adults fall into the range above the deficiency threshold and below the optimal range of vitamin D blood levels, defined as a blood level of 21-79 ng/mL. This middle ground is often referred to as vitamin D insufficiency.

Your Bones Need More Vitamin D Support After Age 45

As mentioned, after the age of 45-50, the body is less able to convert 25-hydroxy vitamin D into calcitriol, due to a decline in the activity of the kidney enzyme known as alpha-hydroxylase. However, if blood levels of 25-hydroxy vitamin D can be raised into the range of 85-120 ng/mL, this amount has been shown to compensate for the drop-off in calcitriol levels. Studies show that supplementation with 800-1,000 IU of vitamin D per day after age 45 can elevate blood levels of 25-hydroxy vitamin D into the ideal range of 85-120 ng/mL, and has been shown to reduce risk of osteoporotic fractures by more than 40%. Let's look at several studies that clearly illustrate this point.

In a 1994 study involving 3,720 elderly women living in nursing homes, those supplemented with 1,200 mg of calcium and 800 IU of vitamin D daily showed a 43% reduction in hip fractures over a three-year period, compared to those not taking such supplements. A 1995 study showed that supplementation of postmenopausal women with 700 IU of vitamin D daily reduced the annual rate of hip fractures from 1.3% to 0.5%, nearly a 60% reduction. A three-year follow-up study showed that supplementation with 500 mg of calcium and 700 IU of vitamin D resulted in a significantly reduced hip fracture rate in men and women taking this combination, compared to the placebo group.

Ample evidence now suggests that after age 45, it is extremely prudent to increase your vitamin D supplementation to reach a total daily value of 800-1,000 IU, in order to significantly reduce your risk of osteoporosis. This advice is meant for both men and women, as osteoporosis now affects one in four women and one in eight men older than age 50. However, the benefits of increased vitamin D supplementation don't end with the prevention of osteoporosis; there are also important implications for the prevention of breast, ovarian, prostate and colon cancer, as we shall explore in part two of this article.

Vitamin D Prevents Cancer

Vitamin D receptors exist on intestinal cells and bone to regulate calcium absorption and bone metabolism, as discussed in part one of this article. However, these receptors are also present in a wide variety of other tissues and organs, including the brain, pancreas, skin, gonads, prostate, stomach, colon, breast, kidney, connective tissue, parathyroid gland, mononuclear cells, and activated T- and B-lymphocytes.

Recent studies indicate that tissues expressing vitamin D receptors are able to convert 25-hydroxy vitamin D, which they extract from the bloodstream, into calcitriol for their own internal use. As I have stated, calcitriol is the most potent form of vitamin D; this is not only true with respect to bone support; studies also illustrate that calcitriol exerts a number of anti-cancer effects on local tissues that convert 25-hydroxy vitamin D into calcitriol for their own purposes, such as breast cells, prostate cells and colon cells. As such, circulating levels of 25-hydroxy vitamin D serve as the raw material from which many tissues synthesize calcitriol for their own internal use. Studies reveal that higher amounts of circulating 25-hydroxy vitamin D in the blood enable local tissues, such as breast, prostate and colon cells, to synthesize greater amounts of calcitriol for their own needs.

Epidemiological (observational) studies suggest that lower blood levels of 25-hydroxy vitamin D are associated with a higher risk of developing breast, colon, ovarian, and prostate cancer. This is an important finding, as one in nine women is expected to develop breast cancer in her lifetime, one in eight men is expected to develop prostate cancer in his lifetime, and one in 16 women and one in 15 men will develop colon or rectal cancer in their lifetimes.

Low-Fat Food Sources of Vitamin D	
Foods	**Approximate I.U.of vitamin D per 3.5 oz.**
sardines (canned)	1150-1570
salmon (fresh)	154-550
salmon (canned)	220-440
herring (fresh)	315
herring (canned)	330
shrimp	150
halibut	44
chicken (raw)	50-67
oysters	5 I.U. per 3-4 medium-sized oysters
nonfat and 1% milk and yogurt (vitamin D-fortified)	100 I.U. per 8 ounces
low-fat cheese (less than 4% milk fat)	12-15

Interestingly, studies show that rates of breast, prostate and colon cancers increase further from the equator (north or south), and that populations residing further from the equator also have lower year-round average blood levels of vitamin D. This is related to the fact that our bodies make vitamin D in response to direct exposure to sunlight (but not through a window). When sunlight hits our skin, it triggers the conversion of 7-dehydrocholesterol into vitamin D (cholecalciferol) within our skin. This enters the bloodstream and travels to the liver, where it is converted to 25-hydroxy vitamin D, which is five times more potent than cholecalciferol in terms of its vitamin D activity.

Thus, populations living closer to the equator, who enjoy more year-round sunlight intensity and exposure, manufacture more cholecalciferol in their skin and demonstrate higher year-round average blood levels of 25-hydroxy vitamin D. As a general statement, populations in North America that live at or above the 42nd latitude have significantly higher rates of breast, colon and prostate cancer than populations living below it. This latitude essentially divides the U.S. into two equal halves: north and south (running through the middle of California, and the tops of Arizona, New Mexico, Texas, Tennessee and the Carolinas). Some exceptions to this observation exist, in that people

living in large cities in the South of the U.S. also have higher rates of these cancers per capita.

C.F. Garland and F.C. Garland, who first published these data in the 1980s, explain this finding by indicating that the air pollution in large cities and tall buildings block much of the sunlight, and that city dwellers tend not to wear short-sleeve shirts and shorts as often as country folk, and tend not to be outdoors during the sunniest hours of the day. As such, individuals in large cities in the South do not have blood levels of vitamin D high enough to protect them from breast, colon and prostate cancer, as do individuals living in the more rural parts of the South, according to these researchers.

Overall, studies indicate that vitamin D blood levels of 85-120 ng/mL are associated with a high degree of protection with respect to risk of breast, prostate and colon cancer, and may significantly reduce the risk of developing multiple sclerosis via immune-modifying influences.

How does calcitriol reduce cancer risk in the breast, prostate, colon and possibly other tissues? Experimental studies reveal that calcitriol exhibits a number of anti-cancer effects. Essentially, prostate, breast, colon and other cells that contain vitamin D receptors extract 25-hydroxy vitamin D from the bloodstream, and convert it into calcitriol - once inside the cell. This, in turn, slows down the rate of replication of these cells, an effect associated with decreased cancer development.

The presence of calcitriol has also been shown to slow the rate of replication of human prostate, breast and colon cancer cells, under experimental conditions. Calcitriol also promotes newly formed cells to mature to their full adult potential, which also decreases the chance of these cells being transformed into cancer cells by some external influence. Calcitriol also exerts a favorable effect on immune function, which is thought to account for some of its anti-cancer influences. It also has been shown to transform the appearance of human cancer cells (e.g., prostate cancer cells) back to healthy, nonmalignant-looking cells, and inhibit their replication, an effect that is lost once the calcitriol is no longer administered.

The problem with relying upon vitamin D from sunlight and food sources: From an evolutionary standpoint, exposure to direct sunlight is the principal way in which we are set up to derive our vitamin D stores. To maximize vitamin D synthesis within the skin, all that is required is 15-20 minutes of direct sunlight exposure to the face, arms and legs, three times per week. However, experts warn that even this amount of cumulative sun exposure increases risk of skin cancer over our lifetime, and that it is best to derive vitamin D from the consumption of fish, vitamin D-fortified dairy products and supplements. The point is that many people don't get 15-30 minutes of direct sunlight exposure each day, especially those of us living above the 42 degrees latitude within North America, where sunlight intensity between October and May is insufficient for our bodies to make vitamin D inside our skin.

Very few foods contain vitamin D in their native form. It is best to eat fatty fish, such as sardines, salmon and mackerel, 3-4 times per week, to help satisfy the body's requirement. Of course, there is supposed to be 100 IU of vitamin D in every eight

ounces of fortified milk, but studies undertaken by M. Holick and others showed that nearly two-thirds of the whole milk samples tested in one study had less than 80 percent (and several skim milk samples had between zero percent and 50 percent) of the amount of vitamin D appearing on the label. This problem will continue, as vitamin D levels in milk are affected by season; the breed of cow; the animal's diet; its exposure to sunlight; and procedures used in fortification.

Although the 1997 recommendations by the Institute of Medicine suggest that middle-aged adults (50-70 years old) should consume 400 IU of vitamin D per day, and older subjects should consume 600 IU per day, evidence is strong to indicate that in the absence of exposure to sunlight, the adequate intake for vitamin D should be at least 800-1,000 IU per day by age 45-50. M. Holick points out that intake is completely safe up to 2,000 IU per day for ages 1 year and above, and that the risk of vitamin D toxicity is greatly exaggerated by many health policy-makers.

Ensuring Optimal Vitamin D Status

The totality of evidence suggests that many North Americans are either vitamin D deficient - or more commonly, insufficient - and would benefit from additional supplementation. Most multivitamins contain 400 IU of vitamin D, an amount that is reported to raise vitamin D blood levels (25-hydroxy vitamin D) by approximately 45 ng/mL. However, even higher amounts of total supplemental vitamin D (800-1,000 IU per day) should be implemented after the age of 45 or 50, as the body's ability to convert 25-hydroxy vitamin D to calcitriol slows down to a significant degree.

Studies prove that higher blood levels of the less potent 25-hydroxy vitamin D (in the range of 85-120 ng/mL) can compensate for the reduced synthesis and availability of calcitriol in the bloodstream after ages 45-50, and can significantly reduce your risk of osteoporosis. Furthermore, this amount is also strongly associated with a reduction in risk of breast, colon, prostate and ovarian cancers, as well as multiple sclerosis.

In a study released in 2004, K.L. Munger and fellow researchers showed that participants in the Nurses' Health Study who ingested 400 IU of vitamin D from supplements daily (most notably from a multivitamin product) showed a 40 percent reduction in risk of multiple sclerosis compared to women who did not use supplements containing vitamin D. This group of 95,253 female registered nurses, residing in the United States, has been followed by researchers since 1980.

Vitamin D has been shown to exert favorable influences on immune cells that are consistent with preventing events related to multiple sclerosis, which have been confirmed in animal and human investigations. Over and above supplementation, note that sardines, mackerel, herring and salmon are excellent food sources of vitamin D, as well.

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NEW STUDY CONFIRMS THE VALUE OF VITAMIN D SUPPLEMENTATION IN THE ADJUNCTIVE TREATMENT OF PROSTATE CANCER

Dr. James Meschino D.C., M.S., N.D.

In recent years a number of epidemiological, experimental, and intervention trials have suggested that vitamin D-based compounds may be important in the prevention and treatment of prostate cancer. Epidemiological studies indicate that higher blood levels of vitamin D (25-hydroxycholecalciferol) are associated with a lower risk of developing prostate cancer. Experimental evidence has shown that prostate cells contain vitamin D receptors, which allow for the uptake of 25-hydroxycholecalciferol from the blood stream. Once inside prostate cells, an intracellular hydroxylase enzyme readily converts 25-hydroxycholecalciferol into 1,25, dihydroxycholecalciferol (calcitriol). This form of vitamin D (calcitriol) exerts a number of cancer preventive effects on prostate cells, including slowing the rate of cell division and promoting cellular differentiation.

Experimental studies also show that prostate cancer cells respond to calcitriol by slowing their rate of replication. Vitamin D metabolites have also been shown to increase differentiation and

apoptosis (programmed cell death), and decrease proliferation, invasiveness and metastasis of human prostate cancer cell lines, in laboratory studies.

Although prostate cancer cells have a more limited ability to produce their own 1,25 dihydroxycholecalciferol (calcitriol) than do non-cancerous prostate cells, studies show that many prostate cancer cells continue to express vitamin D receptors on their cell surface, extract vitamin D (25-hydroxycholecalciferol) from the blood stream and convert some 25-hydroxycholecalciferol into calcitriol.

Evidence suggests that prostate cancer cells could also be affected by calcitriol synthesized by neighboring prostate cells as well as the calcitriol made in the kidneys (the kidneys convert 25-hydroxycholecalciferol into calcitriol, which is the principle source of blood calcitriol levels).

To date, intervention studies with prostate cancer patients have shown that administration with 0.5-2.5 micrograms of calcitriol daily tends to slow the rise in PSA levels in these patients, helping to control the disease process to a very significant degree. Unfortunately, this dosage of calcitriol is well in excess of physiological levels and puts patients at risk for toxicity, including hypercalcemia with the likelihood of calcification of soft tissues and internal organs.

However, more recently, the administration of cholecalciferol, at physiological doses, was shown to decrease or slow the rise in PSA levels in men with prostate cancer without risk of vitamin D toxicity.

The indication from these recent experimental and clinical studies suggests that taking vitamin D in its native form, known as cholecalciferol (this is the over-the-counter form of vitamin D that one would purchase in a drug store or health food store), at a daily dosage of 2,000 IU (50 ug) per day, provides the body with sufficient raw material to convert much of the cholecalciferol into 25-hydroxycholecalciferol (this conversion occurs in the liver). The resulting increase in blood levels of 25-hydroxycholecalciferol, in turn, helps to drive a greater amount of this vitamin D metabolite into existing prostate cancer cells. Experimental evidence indicates that 25-hydroxycholecalciferol, itself, exerts many similar anti-cancer effects on prostate cancer cells, as those produced by calcitriol. The good news is that the ingestion of 2,000 IU of vitamin D (cholecalciferol) is nontoxic and the associated blood level rise in 25-hydroxycholecalciferol resulting from the ingestion of this dosage of vitamin D supplementation, which is usually in the range of 50 nmol/L, is well within normal limits (vitamin D toxicity occurs only when blood levels of 25-hydroxycholecalciferol levels are above 225-250 nmol/L, which does not occur with supplementation at 2,000 IU per day of cholecalciferol).

The most recent study to investigate the use of cholecalciferol in the treatment of prostate cancer was published in the journal, Nutrition and Cancer, in 2005, by TCS Woo and fellow researchers (volume 51, number 1). In this study 15 prostate cancer patients were given 2,000 IU (50 ug) of cholecalciferol daily and monitored prospectively every 2-3 months.

In 9 patients, the PSA level decreased or remained unchanged (no further rise) and these results were sustained during the 21-month course of vitamin D administration. Analysis also showed that there was a statistically significant decrease in the rate of PSA rise after administration of cholecalciferol compared with that before cholecalciferol administration. The median PSA doubling time increased from 14.3 months prior to cholecalciferol administration to 25 months after commencing cholecalciferol. In fact, 14 of the 15 patients showed a prolongation of the PSA doubling time after cholecalciferol supplementation was introduced to this group. There were no side effects reported by any patient.

The marked prolongation of PSA doubling time is an extremely important outcome to the administration of cholecalciferol in these patients, according to the recent work of Partin and fellow researchers (J Urol, 2003). Partin and fellow researchers showed that the risk of distal metastasis of prostate cancer (with respect to relapse after prostate cancer surgery) at 5 years was 65% to 75% when PSA doubling time was less than 10 months compared with 10-20% when PSA doubling time was greater than 10 months.

As such, researchers working in this field point out that agents such as cholecalciferol that lengthen PSA doubling time or decrease the rate of PSA increase, might be clinically useful in the treatment of prostate cancer cases, with respect to affecting (slowing) the clinical course of the

disease, and improving survival rates and quality of life, even if they do not reset the PSA levels into the normal range.

The key aspect of the work presented by TCS Woo and fellow researchers is that the common form of vitamin D (cholecalciferol), which is readily available from retail outlets and other sources, may be the most useful form of vitamin D to use in the prevention and treatment of prostate cancer. It is nontoxic when used at the doses required to exert the beneficial effects noted above, provides a multitude of anti-cancer effects that are similar to that of calcitriol and related vitamin D-drugs (deltanoid drugs) and is highly affordable. As cholecalciferol is a nonproprietary molecule, the cost of providing 2,000 IU of this form of vitamin D is less than $1.50 month US.

Further studies are required to confirm these findings, however, health care practitioners should be aware of the potential benefits of recommending the use of cholecalciferol supplementation as one part of the complementary management of prostate cancer.

It has the potential to slow the rate of prostate cancer cell replication, induce apoptosis (programmed cell death of existing prostate cancer cells), encourage cellular differentiation (reversing some cancerous morphological features of the cell), reduce invasiveness and metastasis, and slow the doubling time of PSA or lower the PSA concentrations in the blood.

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It Takes More Than Vitamin D Supplementation To Optimize Your Defence Against Cancer

James Meschino DC, MS, ND

As most people know a recent study in the American Journal of Clinical Nutrition showed that adults, who were given vitamin D supplements at dosage of 1000 IU per day, showed a 50-60% reduction in cancer incidence compared to individuals not taking vitamin D supplements. These findings should not be surprising as many previous studies have shown that vitamin D exhibits a number of anti-cancer properties and has been used successfully in other recent trials to slow the progression of prostate cancer. Vitamin D encourages cells to fully mature during their growth cycle and to replicate more slowly. These are biological influences are known to reduce the likelihood that a cell will become cancerous within the body. The question was how much vitamin D is required to acquire the maximum preventive benefit from vitamin D, and what is now obvious is that acquiring vitamin D from food alone is not sufficient. A number of recent studies strongly suggest that adults should ingest 800-1,000 IU of vitamin D per day (to achieve a fasting blood level of at least 85 nmol/L of 25-hydroxycholecalciferol). Individuals suffering from sarcoidosis or hyperparathryoidism should refrain from this practice, however, due to potential side effects. This is also true for pregnant and breast feeding women. Overall, there seems little doubt that vitamin D supplementation is a critical aspect of cancer (and osteoporosis) prevention for most adults

However, it takes more than a vitamin D supplement to maximize your defence against cancer, as a number of other supplement ingredients also provide important additional anti-cancer benefits as outlined below:

1. **Calcium** – the calcium your body does not absorb into the bloodstream (about 60-70% of your intake level) remains in the intestinal tract and provides important prevention against colon cancer in its travels through the large intestine. Calcium binds to bile acids in the intestinal tract, preventing their conversion into cancer-causing substances known as secondary sterols (e.g lithocholic acid and deoxycholic acid). Calcium also works with vitamin D to slow the rate of cell division of colon cancer cells. Both of these effects have been shown to reduce colon cancer. In the largest study published to date, Peters and fellow researchers showed that individuals who ingested calcium supplements at a minimum of 1200 mg per day had a 27% reduction in risk of developing colon cancer, compared to nonusers of calcium supplements.

2. **Folic acid and Vitamin B12** – folic acid and vitamin B12 are necessary to provide the body with an ingredient (a methyl group) that is necessary for DNA replication, as cells replace themselves from one generation to the next. Studies show that insufficient folic acid and/or vitamin B12 status allow DNA to become hypo-methylated, which makes them weak, fragile and susceptible to cancerous mutations. The Health Professional Follow Up Study and the Nurses' Health Study have shown that insufficient intake of folic acid increases risk of colon and breast cancer under certain conditions. Insufficient folic acid and vitamin B12 intake are also factors in cervical dysplasia and cervical cancer in women. Overall, studies support daily supplementation with 400 mcg of folic acid and 50 mcg of vitamin B12. These levels keep DNA methylated, strong and more resistant to the influence of cancer causing agents.

3. **Antioxidants Supplementation** – antioxidants such as vitamin C, selenium, lycopene, beta-carotene, vitamin A, lutein, and vitamin E help to reduce cancer risk by quenching free radicals, which are unstable compounds that damage DNA and cause cancerous mutations. Vitamin C has been shown to be especially helpful in protecting the esophagus, stomach, pancreas, cervix and colon, whereas vitamin E protects the mouth, cervix, prostate and colon. Together, vitamin C and vitamin E supplementation have been shown to reduce the concentrations of cancer-causing agents (fecal mutagens) in the colon and rectum and block the formation of cancer-causing nitrosamines within the stomach and intestinal tract. Selenium supplementation is strongly linked to prevention of prostate and colon cancer. Lycopene is acclaimed for its ability to reduce prostate cancer, but it also appears to be important in the prevention of cervical cancer and skin cancer. Vitamin C, vitamin E and selenium are also important in the prevention of skin cancer according to animal studies. Beta-carotene supplementation has been shown to reverse a precancerous condition in the mouth known as leukoplakia as well as early-to-moderate stage cercical dysplasia (a precancerous condition of the cervix). Vitamin A protects much of the interior lining of the body surfaces and

has been used to reverse a number of precancerous conditions. Although lutein is not linked to cancer prevention yet, it is vital to the prevention of macular degenerations, which is the leading cause of blindness in people over the age of 55. For all of these reasons the daily antioxidant cocktail you should consider for disease prevention purposes is as follows:

Vitamin A – 2500 - 3000 IU (anything higher can cause liver damage over time)
Beta-carotene – 10,000 – 20,000 IU
Vitamin C – 1,000 mg
Vitamin E – 400 IU (all natural)
Selenium – 100-200 mcg
Lycopene Powder – 5 mg
Lutein Powder – 5 mg

4. **Omega-3 Fats –** omega-3 fats from fish, fish oil and flaxseed oil, are converted by your body tissues into a cancer fighting hormone (prostaglandin series-3), which slows the rate of cell division within our tissues. As mentioned above when you slow the rate of cell division you reduce the chances of cancerous changes occurring in the cells of the body. This point has been illustrated many times in studies showing that women and men with higher tissue levels of omega-3 fats experience significantly lower subsequent breast and prostate cancer incidence in follow-up studies. The same appears to be true for colon cancer as well, according to some preliminary findings. Most people do not ingest the amount of EPA (eicosapentaenoic acid) and DHA (docosahexaenoic acid) that experts recommend to reduce risk of heart disease, optimize brain development, reduce dementia and Alzheimer's disease risk and help prevent cancer. Unfortunately fish supplies are running low and many fish now contain high levels of mercury, with experts warning us not to consume more than two fish servings per week. Moreover, the essential fat in borage seed oil (GLA) has been shown to work with omega-3 fats to block the production of inflammatory agents that are linked to inflammation and cancer. To optimize your intake of omega-3 fats (EPA, DHA and ALA) and GLA it is highly advisable to take three capsules per day of a supplement containing:
   Fish oil – 400 mg
   Flaxseed oil –400 mg
   Borage seed oil – 400 mg

This combination not only helps to optimize disease prevention, but also promotes the formation of skin hormones that make the texture of your skin smoother and softer and slow certain aspects of aging.

Vitamin D Reference: Lappe JM et al. Vitamin D and calcium supplementation reduces cancer risk: results of a randomized trial. Am J Clin Nutr. 2007;85:1586-91

10. Beta-Carotene

Beta-Carotene: Overview of Health Effects

James Meschino DC, MS, ND

General Features

Beta-Carotene is one of 30-50 carotenoids found in plant foods that can be converted by the body into Vitamin A. Beta-Carotene is a fat-soluble compound that is absorbed intact in the presence of bile salts from the intestine. Beta-Carotene is made up of two Vitamin A molecules (attached together). Within intestinal cells they are split to yield retinol (preformed Vitamin A). Approximately one third of all the carotene in food can be converted into Vitamin A. For Beta-Carotene specifically, about one-sixth is available to become Vitamin A, if the body requires it.

Absorption and Metabolism

The splitting of Beta-Carotene (and other carotenes) into retinol within intestinal cells is well regulated to help guard against Vitamin A toxicity. The retinol that is formed from Beta-Carotene enters the chylomicron and is metabolized from that point forward as preformed Vitamin A. Chylomicrons primarily deliver Beta-Carotene to the liver, where they are repackaged within another lipoprotein carrier system known as the very-low-density lipoprotein.

Beta-Carotene (and other carotenoids) enters the bloodstream from the liver and is transported to peripheral tissue by very-low density lipoproteins and low-density lipoproteins (VLDL and LDL, which is the remnant particle of VLDL after triglycerides are removed by fat cells, muscle fibers and other tissues). In contrast, Vitamin A is transported form the liver attached to retinal-binding protein (RBP). Beta-Carotene is stored in fat tissues, and the adrenal glands, testes, ovaries, rather than the liver and is responsible for the yellowish tinge to the skin when large amounts are stored (carotenodermia). However, carotenodermia is considered to be a nonpathological, reversible condition; not associated with any health risks. Some conversion of Beta-Carotene may take place in the liver and lungs. About 40-60% of Beta-Carotene is absorbed from food. Of interest is the fact that Beta-Carotene supplements are better absorbed than carotenes from food. Beta-Carotene comprises 20-25% of the total serum carotene level.

Functions

1. Vitamin A Precursor: because Beta-Carotene can be converted into Vitamin A, it supports Vitamin A nutritional status and all vitamin A-related functions.
2. Antioxidant: Beta-Carotene is an antioxidant and does not need to be converted into Vitamin A to perform antioxidant functions.

3. Immune System: Beta-Carotene appears to enhance thymus gland function and increases interferon's stimulatory action on the immune system.[1]
4. Other Functions: as described below, Beta-Carotene exhibits a number of immune-enhancing and anti-cancer propertied, and has therefore, been tested in patients with immune-compromised states, precancerous, and cancerous conditions, as well as in patients at high risk in developing certain cancers.

Beta-Carotene Supplementation

1. Compromised Immune Function

A number of studies reveal that older subjects can enhance various aspects of immune function through the supplementation of at least 15 mg of Beta-Carotene (25,000 I.U.) per day. The immune system tends to weaken as humans age, thus researchers have examined various nutrients that may prevent or reverse age-related decline in immune function. High doses of Beta-Carotene have been used in the treatment of immune compromised states and studies on normal human volunteers indicate that supplementation with 180 mg (300,000 I.U.) of Beta-Carotene per day, significantly increased in the number of T-helper cells by approximately 30% after seven days of supplementation, with a 30% increase in a total T-cell count after 14 days. This may be of great significance in HIV/AIDS patients, who have low T-helper cell counts and other parameters of immune function compromise.[2-5] Beta-Carotene supplementation at 50,000 I.U., twice per day administered to AIDS patients has resulted in a 66% rise in total lymphocyte count and a small rise in T-helper cell levels. With discontinuation of Beta-Carotene supplementation, lymphocyte and T-helper cell counts returned to base line levels within six weeks.[2] In a second study, 60 mg (100,000 I.U.) administered to seven AIDS patients resulted in a rise of T-helper cells over the four-week trial period. This is important as it is the T-helper cell (CD$) count that is adversely affected by the HIV virus and largely accounts for the dramatic reduction in immune function seen in HIV and AIDS patients.[27] Not all Beta-Carotene studies with AIDS patients have shown these benefits, but the lack of adverse side effects with Beta-Carotene suggests that it can be used safely as a complementary therapy in these cases.[2] Moderate dosages of Beta-Carotene supplementation may help to slow down or halt the age-related decline in immune function that increases susceptibility to infection and possibly cancer, as we age. This is true as well for other antioxidant vitamins (Vitamin C, Vitamin E, Vitamin A) and the minerals zinc and selenium.[3,4,5]

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2. Cancer Prevention

At this time it is inadvisable to give high dose Beta-Carotene supplementation (50,000 I.U. or greater) to patients who smoke one pack of cigarettes per day or more. The Alpha-Tocopherol, Beta-Carotene study and the CARET study

suggested that Beta-Carotene, in these cases, may slightly increase the risk of lung cancer, although this needs confirmation.[6,7] However, Beta-Carotene does demonstrate a number of anti-cancer properties and has been shown to reverse leukoplakia – a pre-cancerous condition of the oral cavity, as well as early-stage cervical dysplasia, a pre-cancerous condition of the uterine cervix.[8-13] In the Linxian China study, the combination of modest dosages of Beta-Carotene, Vitamin E, and selenium significantly reduced stomach and esophageal cancers, as well as total cancer incidence in high-rish individuals, compared to other vitamin and mineral combinations.[26] Beta-Carotene is an antioxidant, an immune system modulator and enahances cellular differentiation of epithelial cells. All of these effects are associates with the prevention of cancer and the reversal of some early stage cancers and states of dysplasia (pre-cancerous states).[4-13]

3. **Cervical Dysplasia**

   Beta-Carotene has been shown to influence cellular differentiation of surface lining cells (epithelial cells) and enhances immune-system function. Beta-Carotene has been shown to halt the progression of cervical dysplasia and cause a reversal in some cases involving early and moderate stages of this condition, which is known to be a pre-cancerous condition.[12,13,18]

4. **Cardiovascular Disease**

   Beta-Carotene supplementation has been shown to decrease oxidation of LDL-cholesterol, but to a lesser degree than Vitamin E. In this regard, it may help to reduce the risk of cardiovascular disease, as oxidized LDL-cholesterol appears to be more inclined to narrow arteries as part of the atherosclerotic process that leads to heart disease and ischemic stroke. However, evidence is stronger for Vitamin E. Both Vitamin E and Beta-Carotene are transported through the bloodstream within VLDL and LDL lipoproteins, where they are able to act as antioxidants in regards to reducing the oxidation of fatty acids and cholesterol within these lipoproteins (VLDL and LDL).[14,15,16] The Physicians Helath Study failed, to show a benefit in cardiovascular disease reduction with Beta-Carotene supplementation of 50 mg (83,333 I.U.), taken every other day for 12 years. However, a subgroup analysis of these 22,000 medical doctors showed that of the 333 physicians prior history of heart disease, Beta-Carotene supplementation produced a small reduction in risk of fatal and non-fatal heart attack.[30] A number of prospective studies have suggested that higher intakes of Beta-Carotene is associated with a significant reduction in heart attack and stroke, as highlighted in the Western Electric Study in Chicago and a study of Italian women by A Tavani, et al.[28,29]

Dosage

1. Compromised Immune Function: 50,000 I.U., but a dosage of up to 3,000,000I.U. has been used in short term studies[2-5]
2. HIV/AIDS: 50,000 I.U., twice daily has been used with some success[2,27]
3. Oral Leukoplakia: 50,000-1000,000 I.U. per day[9,10]
4. Cervical Dysplasia: 50,000-100,000 I.U. per day[12,13]

5. Cancer Treatment Support: 75,000-100,000 I.U. per day (lung cancer would be an exception)[11]
6. Heart Diseases and Cardiovascular Health: 10,000-75,000 I.U.[14,30]
7. General Wellness: 10,000-25,000 I.U. is commonly consumed

Adverse Side Effects and Toxicity

Overall, the experimental animal data demonstrate a high level of Beta-Carotene safety and in human trials using doses of 20-180 mg/d (up to 300,000 I.U./d) to treat patients with the genetic disease erythropoietic protoporphryria. These large doses did not produce any toxic effects. Other studies have confirmed this. Babies born to mothers with carotenemia show no untoward effects or defects and are otherwise normal.[17]

Drug-Nutrient Interactions

1. **Bile Acid Sequestrants, such as cholestyramine and colestipol may decrease absorption of Beta-Carotene (as they do other fat-soluble vitamins).[19,20]**
2. **Proton Pump Inhibitors such as omeprazole are known to decrease Beta-Carotene absorption.[21]**
3. **Other drugs that impair Beta-Carotene absorption include:**
    a. colchicines[22]
    b. mineral oil[23]
    c. neomycin[24]
    d. orlistat[25]

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11. Parthenolide (Feverfew)

Parthenolide and Cancer: Nuclear Factor kappa-beta Inhibition

James Meschino DC, MS, ND

Parthenolide (PN) is a major sesquiterpene lactone of feverfew (Tanacetum parthanium) with known anti-inflammatory activity. Feverfew has been used as a herbal medicine for the treatment of fever, arthritis and migraine in Asia and Europe for centuries. The crude extracts of this herb are known to have anti-microbial and anti-inflammatory properties. The principal active component in feverfew is the sesquiterpene lactone parthenolide (PN). There is some preliminary evidence showing the anticancer property of PN. For instance, PN is a potent inhibitor of DNA synthesis and cell proliferation in a number of cancer cell lines. Patel and co-workers have reported that PN sensitizes breast cancer cells to the chemotherapeutic agent paclitaxel via its inhibitory effect on nuclear factor kappa B (NF-κB). It has been well documented that PN is a potent inhibitor of NF-κB signaling pathway, which contributes to its anti-inflammatory activity.

More recently, PN has been reported to be a potent apoptosis inducer in human cancer cells. However, the anticancer potential of PN has not been tested on animal models.

Parthenolide and Skin Cancer

The study by Won et al (2004) was the first to provide clear evidence that PN possesses chemopreventive property against UVB-induced skin cancer in female SKH-1 hairless mice. This study showd that PN treatment delayed the onset of tumor incidence from week 13 to week 18 and that PN reduced tumor multiplicity by 30%, and that smaller papilloma sizes were found in mice treated with PN. PN treatment also reduced the hyperplastic response of the epidermis induced by UVB irradiation. The chemopreventive capability of PN was found to be as effective as the pharmaceutical COX-2 inhibitor celecoxib in preventing UVB-induced photocarcinogenesis

Nuclear transcriptional factors such as AP-1, NF-κB, NF-AT and STATs have been reported to be involved in the UVB-induced signaling pathway. The study by Won et al showed that PN subdues UVB-induced AP-1 transactivation via blockage of c-Jun and ATF-2 phosphorylations . The outcome of these series of inhibitions seems to lead to massive sensitization to UVB-induced apoptosis, suggesting an anti-apoptotic role of AP-1. Such anti-apoptotic function of AP-1 and c-Jun has been reported previously.

The UV-activated signal transduction pathway is primarily mediated by MAPKs. Mammals express at least four distinctly regulated groups of MAPK: Erks, JNK, p38 and Erk5. Once activated, MAPKs translocate to the nucleus and phosphorylate target transcription factors. In response to UV, key MAPKs such as JNK and p38 are activated, which in turn phosphorylate c-Jun and ATF-2, respectively

The study by Won et al, noted the potent inhibitory effect of PN on UVB-induced p38 activation. In turn, p38 inhibition leads to sensitization of cells to UVB-induced apoptosis, implying an anti-apoptotic role of p38 in UVB-induced apoptotic cell death.

As such, studies suggest that PN acts as a dual inhibitor of both JNK and p38 in UVB-treated cells. These findings suggest that both JNK and p38 contribute to the cell survival mechanisms in UVB-treated cells.

In summary, the study by Won et al, demonstrated for the first time the chemopreventive property of PN against UVB-induced skin cancer in female SKH-1 mice. In this study apoptosis wase mediated through inhibition on AP-1, JNK and p38 signaling pathways, which may contribute to the anticancer activity of PN. **(1)**

Parthenolide and Breast Cancer

The antitumor activity of the sesquiterpene lactone parthenolide, an active ingredient of medicinal plants, is believed to be due to the inhibition of DNA binding of transcription factors NF-kappaB and STAT-3, reduction in MAP kinase activity and the generation of reactive oxygen.

The study by Nakshatri et al (2004) showed that parthenolide activates c-Jun N-terminal kinase (JNK), which is independent of inhibition of NF-kappaB DNA binding and generation of reactive oxygen species. Parthenolide reversed resistance of breast cancer cells to **tumor necrosis factor-related apoptosis-inducing ligand (TRAIL**)-induced apoptosis. Cancer cells treated with a combination of TRAIL and parthenolide underwent massive typical apoptosis and atypical apoptosis involving the loss of plasma membrane integrity. JNK activity is necessary for the parthenolide-induced sensitization to TRAIL because a dominant-negative JNK or the JNK inhibitor SP600125 reduced TRAIL plus parthenolide-induced apoptosis.

Parthenolide through JNK increased the TRAIL-mediated degradation of the antiapoptotic **protein X-linked inhibitor of apoptosis** (XIAP). Enhanced XIAP cleavage correlated with increased and prolonged caspase 3 activity and PARP cleavage, suggesting that the sensitization to TRAIL involves activation of caspase 3. **These results identify a new antitumor activity of parthenolide, which can be exploited to reverse resistance of cancer cells to TRAIL, particularly those with elevated XIAP levels**.

In summary, PN was shown to increase TRAIL activity, which induces apoptosis via caspase-3 activation, overriding the anti-apoptotic effects of XIAP protein, common in resistant breast cancer cells. (2)

Leukemia and Parthenolide - Experimental studies demonstrate that parthenolide also destroys acute myeloid leukemia (AML) cells, leaving normal bone marrow cells relatively unscathed. Moreover, the compound may get at the root of the disease because it also kills stem cells that give rise to AML. (Blood, online February 1, 2005)

Phase I Human Trial Using Parthenolide - A Phase I trial was conducted to evaluate the pharmacokinetics and toxicity of parthenolide given as a component of "feverfew." Feverfew (Tanacet trade mark) was administered as a daily oral tablet in a 28-day cycle. A starting dose of 1 mg per day was explored with subsequent dose escalations to 2, 3, and 4 mg. The limit of detection for parthenolide in plasma was 0.5 ng/ml. Patients were evaluated for response after every two cycles. RESULTS: Feverfew given on this schedule had no significant toxicity, and the maximum tolerated dose was not reached. When parthenolide was administered at doses up to 4 mg as a daily oral capsule in the feverfew preparation, there was not detectable concentration in the plasma. CONCLUSION: Feverfew, with up to 4 mg of parthenolide, given daily as an oral tablet is well tolerated without dose-limiting toxicity, but does not provide detectable plasma concentrations. Researchers suggest that the purification of parthenolide for administration of higher doses will be needed in future, in order for follow-up testing to proceed. (Invest New Drugs. 2004 Aug;22(3):299-305)

Pancreatic Cancer and Parthenolide

A study by Yip-Schneider et al (2005) evaluated the effect of parthenolide, a sesquiterpene lactone and novel NF-κB inhibitor, isolated from the herb feverfew, in three human pancreatic tumor cell lines (BxPC-3, PANC-1, and MIA PaCa-2).

Parthenolide inhibited pancreatic cancer cell growth in a dose-dependent manner with substantial growth inhibition observed between 5 and 10 μmol/L parthenolide in **all three cell lines**. Parthenolide treatment also dose-dependently increased **the amount of the NF-κB inhibitory protein, IκB-α, and decreased NF-κB DNA binding activity**. Treatment with the combination of parthenolide and sulindac inhibited cell growth synergistically in MIA PaCa-2 and BxPC-3 cells and additively in PANC-1 cells. In addition, treatment with the parthenolide/sulindac combination lowered the threshold for apoptosis. Similarly, decreased NF-κB DNA binding and transcriptional activities were detected in cells treated with the combination compared with the single agents, demonstrating cooperative targeting of the NF-κB pathway.

"These data provide preclinical support for a combined chemotherapeutic approach with NF-κB inhibitors and NSAIDs for the treatment of pancreatic adenocarcinoma". **(3)**

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12. Flaxseeed

Flaxseed Lignans (Enterolactone and Enterdiol), Cancer and Other Health Effects

James Meschino DC, MS, ND

Flaxseed is the richest known dietary source of mammalian lignans . Thompson et al. demonstrated that secoisolariciresinol diglucoside (SDG), isolated from flaxseed, was metabolized into both enterodiol (END) and enterolactone (ENL), and showed chemopreventive properties in the DMBA-induced mammary tumor model in rats.
The inverse association between a high enterolactone (ENL) concentration in both urine and serum, and the risk of breast cancer found in epidemiological studies suggests a chemopreventive action for ENL.
Plant lignans, such as secoisolariciresinol (SECO) and matairesinol found in many edible plants, are transformed by intestinal microbiota into mammalian lignans END and ENL, respectively. ENL is the quantitatively most important lignan in human serum. An inverse association between the urinary or serum ENL contents and the risk of breast cancer has been described in several epidemiological studies. The finding has stimulated considerable interest in the possible chemopreventive action of ENL and its precursors.

In pilot human studies, dietary flaxseed given preoperatively to newly diagnosed breast cancer patients markedly reduced the Ki67 labeling index and c-erb2 of the breast cancer tissue, suggesting direct antiproliferative effects. As expected, there was a marked increase in the urinary excretion of mammalian lignans after the ingestion of flax. A recent study of short-term flaxseed supplementation and fat restriction in diet additionally showed significantly lower proliferation rates and higher rates of apoptosis in prostate cancer of men before operation.

However, the observed effects of flax do not allow for any conclusions on the possible role of lignans in the inhibition of cancer growth, because other potentially bioactive multiple compounds are present in flaxseed. ENL is assumed to mediate the anticarcinogenic action of plant lignans shown in experimental breast cancer and account for the antiproliferative effects seen in the clinical studies of flaxseed supplementation. Until recently, there was no direct evidence for the anticarcinogenic action of ENL in humans or experimental cancer models in animals. The possible anticarcinogenic effects

of ENL on breast cancer were tested for the first time by using a DMBA-induced mammary carcinoma of the rat in this study

Hutchins et al. reported increased serum prolactin concentrations, and decreased serum 17ß-estradiol and estrone concentrations in postmenopausal women after a 7-week administration of flaxseed in a daily dose of 10 g. It is possible that a long-term exposure to elevated concentrations of lignans may alter the composition and metabolic activities of intestinal microbiota, possibly resulting in altered metabolism of other dietary polyphenols. This study showed elevated serum concentrations of equol (EQ) and *O*-desmethylangolensin (*O*-DMA), metabolites of daidzein (DA), were observed. In premenopausal women, high EQ excretion has been associated with lower serum concentration of estrogens and androgens, and higher concentration of sex hormone-binding globulin, a hormonal pattern consistent with lower risk for breast cancer. Thus, the increased serum concentrations of EQ and *O*-DMA could account for chemopreventive effects observed in this study

ENL and Breast Cancer - The study by Saarinen N et al provided the first evidence that ENL (derived from flaxseed) produced growth inhibition on cancer cells. In this study rats with 7,12-dimethylbenz(*a*)anthracene-induced mammary cancers were maintained on a standard open-formula chow diet. Daily p.o. administration of ENL at a dose of 10 mg/kg of body weight for 7 weeks was shown to significantly inhibited tumor growth. The growth-inhibitory effect of ENL was more pronounced on the new tumors, which developed during the treatment period, but ENL also inhibited the growth of those tumors established before the start of the lignan administration. The rat serum concentration of ENL, which illustrated a permanent positive effect on breast cancer growth, was 0.4 µM, which is >10-fold as compared with the serum concentrations found in the general human population.
The effect of ENL was not restricted to any specific histological tumor type. ENL was demonstrated to act as a weak aromatase inhibitor *in vitro* and to reduce the relative uterine weight of the 7,12-dimethylbenz(*a*)anthracene-treated nonovariectomized rats. **(1)**

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Additional Reading On Flaxseed

General Features

Flaxseeds are a rich source of secoisolariciresinol diglycoside (SLD) and other mammalian lignan precursors. Flaxseed contains 800-times more of these mammalian lignan precursors than is found in any other food (i.e. beans, nuts). The presence of SLD and related lignan precursors as well as its rich alpha-linolenic acid (omega-3 fat) content makes Flaxseed a very unique dietary supplement providing a broad range of health-promotion effects. The consumption of Flaxseed or processed Flaxseed (i.e. muffin or

bread) has been shown to significantly raise blood levels of the mammalian lignans enterolactone and enterodiol in a dose-dependent manner. Enterolactone and enterodiol are produced in the colon by the action of bacteria on SLD.[1,4] Both enterolactone and enterodiol act as phytoestrogens and can thus, bind to estrogen receptors on breast (and other reproductive tissues) cells and inhibit certain enzymes (i.e., aromatase enzyme in adipose tissue, also known as estrogen synthase) [2]

These, and other synergistic effects have made Flaxseed a target for breast cancer research. Other Flaxseed attributes have stimulated research to examine its potential benefits in the prevention of cardiovascular disease, cancer (i.e. breast, prostate, colon), osteoporosis and the management of menopausal symptoms. [3,4]

Principle Active Constituents

a. Mammalian Lignan Precursors

In particular: Secoisolariciresinol and matairesinol. These are converted into the mammalian lignans, enterolactone and enterodiol by colonic bacteria. Enterolactone and enterodiol are phytoestrogens and exhibit other functions (i.e. antioxidants).

b. Alpha-Linolenic Acid (an Omega-3 fat)

The amount of alpha-linolenic acid in Flaxseed Powder is far less than in flaxseed oil (see flaxseed oil, alpha-linolenic acid), which is more likely to provide more significant health-promotion effects related to Omega-3 fat consumption (e.g., anti-inflammatory effects)

c. Fiber

Soluble and insoluble fiber [4,5,6]

Clinical Application and Mechanism of Action

5. Cancer Prevention and Support

a. Experimental Evidence - Enterolactone and enterodiol have been shown to exhibit the following anti-cancer functions on an experimental basis:

- Compete with estradiol for nuclear binding estrogen binding sites in rat uteri
- Inhibit growth of estrogen-sensitive breast cancer cell lines
- Inhibit angiogenesis of cancer cells
- Inhibit aromatase enzyme, reducing the synthesis of estrone hormone in adipose tissue
- Decrease aberrant crypt and foci in rat colonic epithelium
- Inhibit epithelial cell proliferation in rat mammary gland
- Decrease tumor size in promotion stage of breast cancer in animal studies [5]
- Reduced colon cancer development with concurrent intake of Flaxseed in carcinogen-induced colon cancer experiments with rats 4
- Provide antioxidant protection [7]
- Lifetime exposure and gestational exposure to Flaxseed lignans induces cancer prevention structural changes to mammary glands and endocrine changes consistent with a reduced risk of breast cancer development in animals [12]

- Induce cancer cell differentiation
- Inhibition of tyrosine kinase and DNA topoisomerase – two key enzymes that determine the rate of cell division [8]
- Anti-mitotic effect [4]
- Reduction of plasma insulin-like growth factor-1 levels (IGF-1) in rats. Higher IGF-1 blood levels is associated with an increased risk of breast cancer in some human studies [9]
- Reduction of pulmonary metastasis of melanoma cells and inhibit the growth of metastasic tumors that formed in the lungs of mice [10]

b. Human Evidence - Enterolactone and enterodiol have been shown to exhibit the following anti-cancer functions in human studies:

- Antiproliferative effect on the breast, similar to that of other selective estrogen receptor modulators, such as tamoxifen and raloxifen [11]
- Stimulate production of sex hormone-binding globulin (SHBG), decreasing levels of free unbound sex hormones [12]
- Decrease the synthesis of 16-alpha hydroxyestrone and increase the synthesis of 2 hydroxyestrone. [13] A higher 2 hydroxyestrone to 16-alpha hydroxyestrone ratio (urinary metabolites) has been shown to be related to a decreased risk of breast cancer in women. This is a suggested "chemoprotective effect". [14]
- Breast cancer patients excrete very low levels of lignans compared with non breast cancer subjects [4]
- Vegetarian women are known to have a lower incidence of breast cancer and exhibit higher blood levels of mammalian lignans [15]
- Improvement in cystic mastalgia, a potentially precancerous condition in women [16]
- Increased or longer luteal phase, increased progesterone to estradiol ratios during the luteal phase, fewer anovulatory cycles, and a decreased tendency to ovarian dysfunction; all of which are linked to a reduced risk of breast cancer in women [4]

6. Cyclical Mastalgia

In a double blind placebo-controlled study by Plu-Bureau et al, they demonstrated that patients ingesting the flaxseed muffin (25gm Flaxseed) reported a significant improvement in breast pain reduction compared to the placebo group, during the three consecutive menstrual cycle duration of the trial. Results were attributed to the antiestrogen effects of Flaxseed lignans. [16]

7. Cholesterol Lowering and Other Effects on Reducing Cardiovascular Disease

Animal studies originally suggested that Flaxseed intake had a hypocholesterolemic effect, that is due to its soluble fiber concentration, not to its alpha-linolenic acid content.[16]

In human trials including men and postmenopausal women with high blood cholesterol levels, Flaxseed Powder or Flaxseed supplementation in various forms has been shown to reduce total cholesterol (approximately 7-10 percent), reduce LDL-cholesterol (approximately 15 percent), reduce lipoprotein (a) (Lp(a)) concentrations by approximately 7 percent. Lp(a) is emerging as an important risk factor for cardiovascular disease as it increases clotting behaviour

of platelets, cholesterol deposition in the artery wall and it oxidizes LDL-cholesterol, making it very atherogenic.

Flaxseed supplementation is the only dietary factor shown to reduce Lp(a) to date.

Estrogen replacement therapy reduces Lp(a) levels, but cholesterol lowering drugs (i.e. statin drugs) do not [17,18,19]

Flaxseed lignans inhibit the cholesterol-7-alpha hydroxylase and acyl CoA cholesterol transferase enzymes, involved in cholesterol synthesis, thus reducing total cholesterol. [24]

8. Chronic Renal Failure and Renal Disease In Lupus

Animal and human studies indicate that a diet rich in soy based protein and isoflavones, and/or Flaxseed lignans retard the development and progression of chronic renal disease. The protective effect is considered to be due to effects on cell growth and proliferation, extracellular matrix synthesis, reduced oxidative stress and inflammation reduction [20]

In humans (and animals) Flaxseed supplementation has demonstrated renoprotective effects in human lupus nephritis, with significant delay in the onset of proteinuria, and preservation of the glomerular filtration rate (GFR) and renal size.

9. Bone Density and Menopausal Symptom Support

Preliminary evidence in women suggests that the phytoestrogen influence of enterolactone and enterodiol (from Flaxseed supplementation) has a positive effect on bone density and menopausal symptoms (i.e. hot flashes) [11]

10. Prostate Cancer Protective Effect

Flaxseed lignans interfere with steroid metabolism and bioavailablity in a fashion that is linked to reduced prostate cancer risk. They also inhibit enzymes such as tyrosine kinase and topoisomerase, which initiate the cellular proliferation rate. Epidemiological studies link the above endocrine and molecular events with up to an 80 percent reduction in risk of prostate cancer [22]

11. Constipation

Human studies reveal that Flaxseed supplementation improves laxation [5] It appears that Flaxseed intake increases fecal excretion of bile acids, increasing laxation and reducing the conversion of bile acids to cholesterol in the liver; thereby lowering blood cholesterol [23,5]

Dosage Ranges

1. General Health and Cholesterol Lowering: 25–50 gms of ground flaxseeds per day (unground seeds are significantly less bioavailable in regards to their lignan precursors content) [1, 13,14,17,18,19]
2. Cyclical Mastalgia: 25 gm per day of ground Flaxseed Powder[1,13,14,16]
3. Female Menopausal Support: 25-50 gm per day of ground Flaxseed Powder[13,14]

Adverse Side Effects and Toxicity

Ground Flaxseed is extremely non-toxic. It is well tolerated according to human trails with no significant side effects, other than the occasional report of mild abdominal discomfort [5,16,23,24]

Drug-Nutrient Interactions

There are no well-known drug-nutrient interactions for ground Flaxseed , Flaxseed Powder or Flaxseed -containing products [1,16,25]

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13. Soy and Soy Isoflavones

Phytoestrogens and Breast Cancer

James Meschino DC, MS, ND

The majority of breast cancers are oestrogen dependent and in postmenopausal women the supply of oestrogens in breast tissue is derived from the peripheral conversion of circulating androgens. In Eastern countries, such as Japan, the incidence of breast cancer is approximately one-third that of Western countries whilst their high dietary intake of phytoestrogens, mainly in the form of soy products, can produce circulating levels of phytoestrogens that are known experimentally to have oestrogenic effects. Their weak oestrogenicity has prompted some health practitioners to recommend soy products as a natural alternative to conventional hormone replacement therapy (HRT).

Breast cancer is the commonest form of cancer among women in the US and almost all of Europe and in the US the incidence of breast cancer amongst caucasians rose from 100 cases per 100 000 women in 1983 to over 130 in 2002. The corresponding figures for women in England were under 80 in 1983 to 120 in 2003. Apart from age (>50) and a family history of breast cancer, many of the known risk factors for this disease relate to a woman's reproductive history, such as early menarche, nulliparity or a late pregnancy, and late menopause as well as **prolonged oral contraceptive use** and **hormone replacement therapy (HRT).** More recently, dietary factors and exposure to endocrine-

disrupting chemicals have been built into the list of risk factors. High consumption of phytoestrogens are associated with a lower incidence of breast cancer.
The outcomes of the Million Women Study and the Women's Health Initiative showing an increased incidence of breast cancer as well as coronary heart disease, stroke, and venous thromboembolism raised alarms about the use of HRT to treat menopausal women. These findings were eagerly reported in the media and, together with the clinical evidence, had a negative impact on HRT prescribing. In fact, within months of these publications, there was a significant decrease in HRT prescriptions, particularly the combined conjugated equine oestrogens and progestagen products, and such declines have been reported in several countries. This could result in an increased use of 'natural' plant-derived alternatives to HRT that contain phytoestrogens.

Estrogens and Breast Cancer

Phytoestrogens are plant-derived chemicals that have oestrogenic activity, combining with oestrogen receptors and initiating oestrogen-dependent transcription. Their affinities for the oestrogen receptor, however, are at least 1000–10 000 times lower than that of oestradiol.
The majority of breast cancers (~70%) express oestrogen receptors and in these cases oestrogen is considered to promote tumor growth. Hence, treatments subsequent to surgery, radiotherapy, and/or chemotherapy are directed towards reducing oestrogenic effects within breast tissues. The incidence rate of breast cancer continues to increase with age despite the loss of ovarian hormones in postmenopausal women. This apparent paradox has been resolved by the fact that extragonadal sites, including breast, brain, muscle, skin, bone, and adipose tissue can synthesize potent androgens and oestrogens from relatively inactive circulating steroid precursors derived from the adrenal cortex and to a much lesser extent the ovaries. Indeed, after the menopause, nearly 100% oestrogens are formed in peripheral tissues and exert their effects locally in a paracrine or intracrine manner. In breast tissues, enzymes are present to convert inactive circulating precursors, such as androstenedione, dehydroepiandrostenedione (DHEA) and its sulfated form DHEA-S, and oestrone sulfate (E_1S) into biologically active androgens and oestrogens

In post-menopausal women, the concentration of 17ß-oestradiol (E_2) present in breast tumors is at least 20-fold higher than that in the circulation, but in premenopausal women with carcinomas, this ratio was only 5,

Enzyme Pathways In Estrogen Synthesis:

1. The aromatase enzyme, CYP19, is a key enzyme in the conversion of androgens to oestrogens and over 60% of breast carcinomas express this enzyme with higher levels of mRNA expression and activity compared with non-malignant tissue. This conversion has been reported in both breast cancer cells and supporting stromal tissues.

2. 17 beta-hdroxysteroid dehydrogenase (17ß-HSD) - Oxidative 17ß-HSD2 activity that converts testosterone to androstenedione and oestradiol (E_2) to oestrone (E_1) is predominant in normal breast tissue, but the reductive activity of 17ß-HSD1 that drives the reverse reaction (E_1 to E_2) is dominant in breast cancers. Estradiol is the most potent estrogen in the body, which may be a factor in promoting breast cancer cell replication.

Together, results suggest that 17ß-HSD1 may play an important role in maintaining high intratumoral oestradiol levels in postmenopausal breast cancers. Interestingly, 17ß-HSD type 12, an isoform of 17ß-HSD3 that is important in producing testosterone in the testis, has recently been characterized. 17ß-HSD12, like type 1, converts oestrone to oestradiol and recent evidence suggests that this isozyme is the major oestrogenic 17ß-HSD enzyme in the ovary.

3. **Steroid sulfatase (STS)** is an enzyme that hydrolyzes several steroids, including E_1S and DHEA-S. In breast tissue, it catalyzes the biologically inactive oestrone-3-sulfate to oestrone, which can be further metabolized to oestradiol by 17ß-HSD1. STS activity is detected in the majority of breast cancers. This is may be another reason for the high estrodiol levels seen breast cancer cells.

4. **Oestrogen sulfotransferase (EST, SULT1E1)** that converts oestrogens to inactive oestrogen sulfates is present in both normal and cancerous breast tissue, and EST immunoreactivity was detected in the carcinoma cells of 44% patients with breast cancer Despite the fact that the concentration of E_1S in breast cancer tissue is 7–11 times greater than in plasma, its significance in relation to the formation of active oestrogens in breast carcinoma cells is poorly understood

5. **Type-1 3ß-Hydroxysteroid dehydrogenase** is expressed in the placenta and other tissues, including skin and breast, where it is considered mainly to convert DHEA to androstenedione. 3ß-HSD activity and immunoreactivity have been detected in breast carcinoma cells in a minority of breast cancer tissues and thus, this enzyme may be important in increasing the local concentration of androstenedione, a substrate for oestrogen synthesis.

Phytoestrogens That Inhibit Key Enzymes In Estrogen Synthesis

1. Aromatase - Amongst the phytoestrogens, the flavones and flavonones are the most potent inhibitors of aromatase. Early studies showed that apigenin and quercetin were potent inhibitors of aromatase in human placental microsomes and subsequent studies confirmed apigenin as a relatively potent inhibitor of the enzyme, along with chrysin, 7-hydroxyflavone, and hesperetin with $IC_{50s.}$

Overall, the isoflavones have weak inhibitory activity on aromatase in both cell-free and whole-cell preparations. Similarly, lignans have weak inhibitory activity on aromatase in placental microsomes, human pre-adipocytes , human granulosa luteal cells, and MCF-7 cells. However, their inhibition of aromatase activity may still be of importance clinically, even though they are not highly potent in this regard.

2 17ß-HSD - The isoflavones have been shown to exert inhibitory effects on 17ß-HSD type 1 – the enzyme that converts oestrone to oestradiol. This appears to be especially true for genistein and diadzein. This contrasts the relatively potent effects of biochanin A on recombinant 17ß-HSD type 5 that reduces androstenedione to testosterone.

In breast cancer cells, the production of oestradiol is several-fold higher when androstenedione is used as a substrate compared with testosterone. The conversion of oestrone to oestradiol, which requires only 17ß-HSD type 1, is higher than either the conversion of androstenedione or testosterone to oestradiol. Thus, the activity of 17ß-HSDs exceeds that of aromatase and may be more important in the local generation of oestradiol from oestrone and E_1S than from androgen substrates.

3. Oestrone sulfatase (ETS) that converts E_1S to oestrone may be more important for generating oestradiol in breast tissue compared with the aromatase pathway. Early studies reported that quercetin, genistein, daidzein, and other flavonoids inhibited hepatic ETS, with quercetin being the most potent phytoestrogen.

Taken together the evidence shows that phytoestrogens have an overall inhibitory activity on enzymes involved in the interconversion of E_1S and oestrone. In addition, dietary flavonoids can be sulfated by several human sulfotransferases, including oestrogen sulfatase. This sulfation may further influence the bioavailability of endogenous oestrogens and alter the ratio of active oestrogens and inactive oestrogen sulfates

Estrogen Receptors

Two oestrogen receptors (ERs) coded on different genes have been identified – ERα and ERß – although numerous mRNA splice variants exist for these receptors in both diseased and normal tissue. Like other steroid receptors, they are transcription factors and have a similar domain structure. The N-terminal A/B domains are completely distinct between the two receptors, the DNA-binding C domain is highly conserved, whilst the C-terminal E/F domains are similar, sharing about 50% homology. That said, the ligand-binding cavity in the E domain is highly conserved in the two receptors.

There is a distinct tissue expression of ERα and ERß and of their co-regulators and thus oestrogenic ligands will have different effect in different tissues.

In addition, selective oestrogen receptor modulators (SERMs), such as tamoxifen and raloxifen can exhibit tissue-specific oestrogenic activity. Thus, they are both antagonists in breast tissue but agonists in bone, while only raloxifen is a pure antagonist in the uterus. This is because the binding of a SERM results in specific conformational change in the receptor and this determines which co-activators or co-repressors are recruited.

Ways In Which Phytoestrogens Have Been Shown To Inhibit Proliferation Via Receptor Binding

(1) Functional domains of the ERα and ERß - Activation function-1 (AF-1) can recruit co-regulators for gene transcription independent of ligand binding to the E/F domain of the receptors. The AF-1 has only weak activity in ERß receptors compared with αreceptors. The ability of AF-2 to recruit co-regulators is dependent on ligand binding and is equally active in both ERα and ERß.

(2) Homodimers of ERß have greater transcriptional activity at response elements of the DNA (e.g. AP-1) other than the specific oestrogen-response element (ERE). This

contrasts the binding of ERα homodimers to the ERE. **Dimerization of ERß with ERα is thought to silence ERα.**

(3) Many phytoestrogens bind preferentially to ERß, dimers of which may bind to consensus sites, such as AP-1 and activate/inhibit genes that regulate tumor growth. **Binding to ERß may also induce heterodimerization with ERα and hence silence its activation of genes stimulating cell proliferation**.

Both steroids and growth factors stimulate proliferation of steroid-dependent tumor cells and interaction of these pathways can occur at several levels. This is important in relation to the possible effects of phytoestrogens because although these compounds are all weakly oestrogenic, some are also inhibitors of cell signaling pathways.
Whilst activation of ERα is known to promote growth in breast tumors, the specific functions of ERß are less well understood, although in cell lines activation of ERß inhibits cell proliferation. The ratio of ERα:ERß is increased in carcinogenesis and expression of ERß was associated with low breast tumor aggressiveness and improved disease-free survival rates compared with those with ERß-negative tumors. **One theory for the effects of ERß on the growth of breast tumors is that it may dimerize with ERα and silence its activation functions and that loss of ERß in cancer cells could signal the stage of oestrogen-dependent tumor progression.**
Some phytoestrogens may increase the expression of ERß isoforms and this could be important in limiting the oestrogenic stimulation of growth. This is under study at this time.

Activation and interaction of oestrogen receptors with cell-signaling pathways and their inhibition by phytoestrogens.

Growth factors activate cell-signaling pathways, such as Externally Regulated Kinase (ERKs), and phosphatidylinositol kinase (PI3K)-AKT pathway. They also activate membrane-associated estrogen receptors. In turn activated kinases phosphorylate and activate oestrogen receptors.
Activated cytosolic ERs may also modulate the activity of cell-signaling pathways.
Genistein is a potent tyrosine kinase inhibitor and inhibits signal transduction of growth factors.
Apigenin (a flavone isolated from parsley and celery inhibits PI3K and further inhibits cell signaling.

Overall, studies have shown that low doses (≤10 µM) of genistein, biochanin A, and daidzein stimulate in vitro growth of MCF-7 and T-47D cells, but not the ER-negative MDA-MB 231/435 breast cancer cell lines. Howerver, at higher doses growth and survival of both ER-positive and ER-negative breast cancer cell lines is inhibited.

Some phytoestrogens are inhibitors of cell-signaling pathways. For example, **genistein** is an inhibitor of **protein tyrosine kinase** whilst **apigenin** and **quercetin**

are inhibitors of the **phosphatidylinositol kinase (PI3K)** pathway. Resveratrol has been reported to inhibit Src tyrosine kinase and blocks Stat 3 activation in malignant cells. Both MEK/ERK 1/2 and PI3K/AKT pathways can be activated by growth factors and thus phytoestrogens may modulate such control of breast tumor growth.

Growth inhibitory effects of high doses of phytoestrogens may involve inhibition of cell-cycle progression, inhibition of growth factor cell signaling pathways (e.g. tyrosine kinase and/or PI3K) or antagonism of endogenous oestrogens.
Some experts suggest that high concentrations of certain phytoestrogens are simply cytotoxic to cancer cells, since concentrations of certain phytoestrogens, such as genistein and prenylnaringenin show little or no dose inhibitory effects on cell growth at low concentrations with a steep reduction being observed at 10 μM and above.

Overall, studies have shown that the average daily intake of phytoestrogens in some Eastern populations is around 30 mg/day compared with Western societies in which the dietary intake is <10 mg/day or much lower. Asians consume 20-50 times more soy per capita than their Western counterparts and a high dietary intake of soy products has been linked to a lowered incidence of breast cancer. Reported circulating concentrations of various phytoestrogens range from nanomolar to low micromolar concentrations and yet, with the exception of in vitro studies on the growth of ER-positive breast cancer cell lines, most experiments have shown that relatively high concentrations of phytoestrogens, in the micromolar range, are required to exert any pharmacological effects, i.e. at concentrations higher than those likely to be achieved as a result of dietary exposure, which implies that supplementation with phytoestrogens is required to reach concentrations associated with combating growth of existing cancer cells. **(1)**

An important study by Nobert et al, explored the potential differential responses of the ER+ MCF7 primary breast carcinoma cell line (estrogen receptor positive human breast cancer cells) cultured as microscopic solid tumors in vitro to the growth inhibitory effects of genistein as opposed to that of tamoxifen.

The results showed that 5 μM genistein increased the inhibitory effects to almost 100 % 48 hours post treatment. In the absence of supplemental β-estradiol, treatment with 10 μM tamoxifen as the sole agent inhibited cell growth by only 20 %. However, combining this dose of tamoxifen with 50 μM genistein produced an 85 % reduction in viability 48 hours post treatment.
These pre-clinical findings suggest potential therapeutic applications. First, they suggest that combined **tamoxifen/genistein** treatment protocols might produce **significantly greater therapeutic** effects than either of these drugs administered individually. Secondly, the observed reduction in therapeutic effect of tamoxifen under conditions of low estrogen suggest a potential explanation for the decreased clinical effectiveness and increased resistance to tamoxifen observed in post-menopausal women whose levels of circulating estrogen are generally low. The molecular basis of the estrogen-dependence of tamoxifen's growth inhibitory effect on MCF-solid tumors is not clear.

Additional studies reported here suggest that combined treatment of MCF7 cells with **tamoxifen/genistein** produced enhanced **cytotoxic effects.** The effects of genistein on **AKT** pathway activation may affect downstream targets such as the transcriptional regulator **NFκB**. **Moreover, recent studies have suggested that NFκB inhibition markedly enhances the sensitivity of resistant breast cancer tumor cells to tamoxifen**. The studies reported here show that this effect is estrogen-dependent, whereas combined therapy using genistein and the **NFκB inhibitor parthenolide** produced enhanced cytotoxic responses in MCF7 solid tumors independent of estrogen levels

Parthenolide, the active component of the medicinal plant Feverfew, is known to exert inhibitory effects on the nuclear factor kappa B (NFκB) survival pathway by blocking the phosphorylation and degradation of the NFκB inhibitor IκB. Since activation of the NFκB survival pathway has been shown to limit the effectiveness of conventional chemotherapy interventions, successful inhibition of this pathway may result in the restoration of cell sensitivity to chemotherapeutic agents, thereby thwarting mechanisms of acquired drug resistance.

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MCF7 MTS treated with parthenolide at a dose range of 1-5 μM for 48 hours showed that a complete cytotoxic response was achieved at 3 μM in the presence or absence of supplemental 10 nM β–estradiol. Moreover, combination therapy **using genistein and parthenolide** produced a synergistic effect independent of added estrogen. Combined treatment using **50 μM genistein and 2.5 μM parthenolide produced almost complete cytotoxicity under conditions that produced only 60 % growth inhibition with parthenolide alone as compared to 80 % growth inhibition using genistein alone.** Similar results were obtained in the absence of added estrogen.

Combined treatment of MCF7 spheroids for 48 hours with 10 μM tamoxifen and 2.5 μM parthenolide produced approximately 98 % growth inhibition in the presence of estrogen. In contrast, tamoxifen alone produced only 30 % loss of viability and parthenolide showed 40 % cell viability. When the assays were repeated in the absence of supplemental estrogen, combined treatment produced only 70 % inhibition

In conclusion, these pre-clinical research findings suggest possible clinical applications relevant to the use of tamoxifen in women with low levels of circulating estrogen and suggest that genistein might be a useful clinical alternative, particularly in post-menopausal women in whom breast cancer occurs more frequently. Moreover, this research suggests that combined treatment approaches involving the use of tamoxifen in conjunction with agents that inhibit NFκB pathway signaling, such as parthenolide and genistein, warrant further study. **(2)**

Genistein, Heat Shock Proteins, Angiogenesis and Apoptosis

Soy products contain high levels of genistein, a phytoestrogen that is a potent inhibitor of cell proliferation and angiogenesis. Genistein has been found to inhibit the growth of carcinogen-induced cancers in rats and human leukemia cells transplanted into mice. The induction of stress proteins (e.g., glucose- related proteins and heat shock proteins) in tumor cells has been shown to protect them against programmed cell death; this stress response is inhibited by genistein.

Heat shock proteins (HSPs) and Glucose-related proteins (GRPs) are synthesized and secreted by cells in response to adverse physiologic or metabolic conditions. High levels of HSPs and GRPs occur in cancer and virally transformed cells. GRPs have been shown to **upregulate tyrosine kinase** activity in cancer cells and the presence of HSPs and GRPs make cancer cells more **resistant to chemotherapy and radiation and apoptosis**. Recent studies indicate that genistein inhibits the induction of HSPs and GRPs by interference of a key common transcription factor (CBF/NF-Y) binding to the most proximal CCAAT site of the HSP70 and GRP78 promoters. It has been shown that CCAAT binding factor is required for full stress inducibility of both the HSP70 and GRP78 promoters in mammalian cells. Thus, genistein is natural compound shown to inhibit the release of HSPs and GRPs in cancer cells under experimental conditions, and may be one additional way that genistein is linked to lower cancer risk, apoptosis and anti-angiogenesis. **Genistein** has also been shown to be a potent **inhibitor of tyrosine kinase** activity in mammalian cells.

The mechanism(s) by which genistein affects certain stress response genes was explored in the study by Zhou et al. In this study **genistein** was shown **to antagonized the binding of a specific transcription factor**, known as nuclear factor-Y/CCAAT binding factor (NF-Y/CBF), to the CCAAT sequence in the hsp70 and grp78 promoters; this CCAAT element was previously shown to be necessary for full-stress inducibility of both genes. Treatment of cells with genistein converted NF-Y/CBF into a nonbinding, transcriptionally inactive form. These findings support previous work suggesting that one of the anticancer effects of genistein may be related to its ability to reduce the expression of stress response-related genes (HSPs and GRPs). **(3)**

Other Anticancer Effects Of Genistein

Genistein has been found to have a number of antioxidant activities. It is a scavenger of reactive oxygen species and inhibits lipid peroxidation. It also inhibits superoxide anion generation by the enzyme xanthine oxidase. In addition, genistein, in animal experiments, has been found to increase the activities of the antioxidant enzymes superoxide dismutase, glutathione peroxidase, catalase and glutathione reductase.

In summary, several mechanisms have been proposed for genistein's putative anticarcinogenic activity. These include upregulation of apoptosis, inhibition of angiogenesis, inhibition of DNA topoisomerase II and inhibition of protein tyrosine kinases. Genistein's weak estrogenic activity has been suggested as another mechanism for genistein's putative anti-prostate cancer activity. In addition to the above mechanisms, other mechanisms of genistein's putative anti-prostate cancer activity include inhibition of nuclear factor (NF)-Kappa B in prostate cancer cells, downregulation of TGF

(transforming growth factor)-beta and inhibition of EGF (epidermal growth factor)-stimulated growth. Genistein's anti-estrogenic action may be another possible mechanism to explain its putative anti-breast cancer activity. **(4,5).** Other studies suggest that inhibition of heat shock proteins and glucose-related proteins (stress proteins) may also factor into anticancer effects of genistein. **(3)** Various isoflavones and flavonoids may also retard estradiol synthesis (by acting as enzyme inhibitors) within breast cancer and stromal cells (as well as in other tissues), which may help to retard cancer growth. **(1)**

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Two Recent Studies Suggest That Soy Isoflavone Supplementation May Benefit Breast And Prostate Cancer Patients

James Meschino D.C., M.S., N.D.

Breast cancer is the most common cancer diagnosed in women in the United States. Epidemiological, migration and experimental studies suggest that environmental factors, including nutritional practices, play a key role in the etiology of breast cancer, in addition to age, ethnicity, hormonal factors, growth factors, obesity and physical activity. The age-adjusted death rates from breast cancer are twofold to eightfold less in Asian countries than in the United States and Western Europe. The rate in Asian American women born in China or Japan and in their U.S. –born counterparts is about 50-75% lower than in U.S.-born white women. A number of researchers have suggested that the frequent consumption of soy products is largely responsible for the significant reduction in breast cancer incidence among Asian women. However, controversy about the use of soy was sparked when some recent animal studies suggested that soy may stimulate breast cancer growth and interfere with antihormonal treatments. Conversely, other animal studies have suggested that soy induces apoptosis (programmed cell death) of breast cancer cells, and is beneficial in the prevention of breast cancer as well as preventing recurrence of breast cancer.

It is particularly crucial to determine the answer to the question of whether soy is beneficial in humans because hundreds of thousands of patients with breast cancer consume soy supplementation either as chemoprevention of recurrent cancer and/or as treatment of their postmenopausal symptoms (soy isoflavones are known to reduce hot flashes and other menopausal symptoms). In addition, many women would like to know if the regular consumption of soy products and/or supplements can reduce their risk of developing breast cancer. In some Asian countries, where breast cancer rates are as much as 75% lower than in U.S. white women, the daily consumption of soy is on average 20-50-times higher per capita, compared to consumption rates by U.S. white women. Asian women, following a traditional Asian diet, have been shown to consume 30-50 mg of soy isoflavones per day. Soy isoflavones have been shown to produce many of the anti-cancer effects attributed to soy consumption.

The most definitive study performed to date, addressing the effects of soy on breast cancer, was published recently by M. Sartippour, JY Rao, S Apple et al, in Nutrition and Cancer in 2004. These researchers performed the first known pilot study, which examined the association between administration of soy isoflavone tablets in human breast cancer patients and their effects on human breast cancer tissue, as well as the proliferative activity of human blood form breast cancer patients.

The study was performed on 17 human breast cancer patients. After the diagnosis of breast cancer was made on the core-needle biopsy procedure, patients were enrolled, at which time blood and urine were collected in conjunction with routine preoperative tests. Breast cancer subjects received four tablets of soy isoflavones daily until their scheduled surgery date. Each tablet contained 50 mg of soy isoflavone as the active ingredient (200 mg of soy isoflavones ingested daily). Twenty-six age-matched control subjects who did not have breast cancer, but had similar demographic, lifestyle and other characteristics to the breast cancer patients, were selected for comparison. The control subjects did not ingest the soy isoflavone tablets during the course of the study.

In this study the researchers observed that the 17 breast cancer patients receiving the soy isoflavone tablets did not show an increased rate of proliferation of breast cancer cells or endothelial cells during the course of the study (as determined by regular needle biopsy throughout the study period) compared to the breast cancer-free control subjects who were not ingesting soy isoflavone tablets. Furthermore, the researchers noted a statistically non-significant trend towards cancer growth inhibition in the isoflavone treated group, as manifested by higher apoptosis/mitosis ratios compared with those from the breast cancer-free, untreated control group. The apoptosis/mitosis ratio is a comparison of the rate of programmed cell death (growth inhibition) measured against the rate of cell division (proliferation). A faster rate of cell division (proliferation) is associated with breast cancer growth and spread of the disease. In this study researchers were able to show that the administration of soy isoflavones produced a favorable outcome on the apoptosis/mitosis ratios in breast cancer patients, which is consistent with the containment of breast cancer.

Overall, these findings suggest that supplementation with 200 mg of soy isoflavones daily by breast cancer patients may help to suppress the growth of breast cancer cells. As such, soy isoflavone supplementation deserves consideration as an intervention during the period leading up to breast cancer surgery in breast cancer patients and as a daily supplement to potentially help reduce the recurrence of breast cancer in breast cancer survivors.

Previous investigations have built a compelling case for regular soy consumption as a means to prevent the development of breast and ovarian cancers in women and prostate cancer in men. Soy isoflavones have been shown to lower endogenous estrogen levels, stimulate the production of sex hormone-binding globulin by the liver (which in turn leads to more bound and less free estradiol, reducing the amount of estrogens available for binding with estrogen receptors), inhibit the enzymes that promote cell proliferation (protein tyrosine kinase, DNA topoisomerase and ornithine decarboxylase), inhibit angiogenesis (which prevents the building of life-supporting blood vessels in and around malignant tumors), provide antioxidant defense and induce cell differentiation. Further, the weak estrogenic potential (more than 1,000 times weaker than estradiol) of soy isoflavones do not elicit a strong estrogenic response and thus have an anti-estrogenic effect that tends to inhibit the growth and proliferation of estrogen-dependent cancer cells, as demonstrated by the research of A. Molteni et al.

The recent study by M. Sartippour et al, provides further evidence that a high intake of soy isoflavones may not only prevent reproductive cancers, but may be beneficial in controlling the progression and recurrence of breast cancer.

In a similar intervention study, involving men with existing prostate cancer, supplementation with 100 mg per day of soy isoflavones showed a favorable outcome in stabilizing PSA levels (M Hussain et al, Nutrition and Cancer, 2003). In this study soy isoflavone supplementation was shown to decrease the rate of rise in serum PSA (prostate specific antigen) levels in patients with androgen dependent and androgen-independent prostate cancer. These researchers concluded that their data suggests that soy isoflavones may benefit some patients with prostate cancer by slowing the progression of the disease and therefore, potentially delaying the development of symptoms, improving quality of life, and perhaps even prolonging survival.

The positive findings reported in these recent studies has prompted a great deal of interest in developing larger intervention trials using soy isoflavone supplementation in the treatment of breast, ovarian and prostate cancers. Studies involving a larger number of patients are certainly required before definitive statements can be made regarding the use of soy isoflavones in the treatment of these conditions, however, health practitioners should be aware of the encouraging results seen in these recent intervention trials, which they may wish to share with selected patients within their practice.

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14. Lycopene

Lycopene and Cancer

James Meschino DC, MS, ND

Lycopene is a non-provitamin A carotenoid that gives tomatoes their red color. Humans and animals do not synthesize lycopene, and thus rely on food sources. Other sources of lycopene include tomato products (tomato sauce, tomato juice), watermelon, pink grapefruit, apricots, pink guava and papaya. Lycopene is a potent antioxidant and quenches singlet oxygen and may modulate mutagenesis, carcinogenesis, cell differentiation and proliferation, according to experimental evidence. More specifically, experimental evidence suggests that lycopene may be beneficial in adjunctive cancer management in that it has been shown to:

1. Inhibit replication of cancer cells via its influence on cell cycle regulatory proteins.
2. Induce cellular differentiation of cancer cells, helping to control their malignant behavior
3. Modulate the IGF-1/IGFBP-3 system, which induces apoptosis via its effects on transforming growth factor-beta-1 and p53 tumor suppressor gene activity. In the human body 97-99% of IGF-1 is bound to one of six IGF binding proteins. . IGFBP-3 is the most abundant of these binding proteins, accounting for approximately 80% of IGF-1 binding. Induction of IGF-BP3 gene expression by lycopene is associated with enhanced secretion of an active form of IGF-BP3 capable of inhibiting mitogenic signaling by the insulin-like growth factor IGF-1. Other results indicate that IGF-BP3 up-regulates the p53 tumor suppressor gene thereby, inducing apoptosis of cancer cells and other damaged or infected cells
4. Induce up-regulation of gap junction communication between adjacent cells (via gap-junctional gene connexin 43 (Cx43).
5. Affect modulation of redox signaling (antioxidant function, as free radical levels augment synthesis of 5 and 12 lipoxygenase enzymes, as well as cyclo-oxygenase enzyme, which affect the cellular proliferation rate)
6. Inhibit inteukin-6 and androgens
7. Inhibit the 5-lipoxygenase enzyme
8. Modulate carcinogen metabolizing enzymes (increased detoxification at the cellular level)
9. Modulate immune function

Human Prostate Clinical Trial Using Lycopene

Dietary intake of lycopene and soy have been associated with a lower risk of prostate cancer. In vitro studies with lycopene and genistein, a soy isoflavone, have shown induction of apoptosis and inhibition of cell growth in androgen-sensitive (LNCaP) and androgen–independent (PC3 and VeCaP) prostate cancer cell lines.

In a Phase II clinical trial (Vaishampayan U et al, 2007) researchers investigated the efficacy of lycopene alone or in combination with soy isoflavones on serum PSA levels in men with prostate cancer. To be eligible for the study, men with prostate cancer had to have rising serum PSA following local therapy or while on hormone therapy. The study population included 71 patients who had 3 successive rising PSA levels or a minimum PSA of 10 ng/ml at 2 successive evaluations prior to starting therapy.

Subjects were randomly assigned to receive a tomato extract capsule containing 15 mg of lycopene alone (n = 38) or together with a capsule containing 40 mg of a soy isoflavone mixture (n = 33) twice daily orally for a maximum of 6 mo. One patient on the lycopene arm did not receive therapy due to his inability to ingest the study pill.

There was no decline in serum PSA in either group. However, 35 of 37 (95%) patients in the lycopene group and 22 of 33 (67%) patients in the lycopene plus soy isoflavone group achieved stable disease, meaning that they achieved stabilization in serum PSA level. The data suggest that lycopene and soy isoflavones have important adjunctive effects in prostate cancer patients with PSA relapse disease and may delay progression of both hormone-refractory and hormone-sensitive prostate cancer.

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Additional Information Regarding Lycopene And Cancer

Lycopene represents as much as 50% of the carotenoids found in human serum. Lycopene demonstrates a high level of bioavailability, which is enhanced when tomatoes and other lycopene-containing vegetables and fruits are heated, chopped and processed. This explains why tomato sauce is such a good and bioavailable source of lycopene. Lycopene has been shown to concentrate in the prostate gland, adrenals, testes, skin, liver and kidneys. The body cannot convert lycopene into Vitamin A, as it can with some other carotenoids (e.g. beta-carotene), but lycopene demonstrates powerful antioxidant activity and is the most effective quencher of oxygen free radicals among the many carotenoids found in nature, being twice as potent as beta-carotene in this regard.

Under experimental conditions lycopene is able to reduce lung adenomas and carcinomas, colon cancer, prostate cancer, breast (mammary) tumor, and to inhibit endometrial cancer as well as HL-60 leukemic cell growth.

Epidemiological evidence indicates that the ingestion of tomato products is associated with a reduction in risk of overall cancer mortality or incidence. The evidence appear to be strongest for reduced risk of pancreatic, rectal, colon, esophageal, oral, cervical, breast and prostate cancer. The Mediterranean-type diet in particular, which has a higher intake of tomato products, is associated with lower incidence of digestive tract cancers.

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Prostate Cancer

Lycopene intake of 6.5 mg per day has been linked to a 21% decreased risk of prostate cancer compared with those consuming the least. This study also reported that those who ate more than ten servings per week of tomato-based foods had a 35% decreased risk of prostate cancer compared with those eating less than 1.5 weekly servings.[1]

Lycopene is the most abundant carotenoid in the prostate[2], and high blood levels of Lycopene have been linked to prostate cancer prevention,[3] including prospective or longitudinal studies.[4]

Women's Health

Higher intakes of Lycopene have also been associated with a lower risk of cervical intraepithelial neoplasia – precancerous changes of the cervix and cervical dysplasia.[5-8] Some preliminary evidence also suggests that Lycopene may help reduce the risk of breast cancer.[9]

Immune System

Lycopene supplementation has also boosted immune function in the elderly (15 mg of Lycopene per day increased natural killer cell activity by 28% in twelve weeks) [9]

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15. Induribin

Induribin and Cancer

James Meschino DC, MS, ND

It has been the custom to treat chronic myelocytic leukemia (CML) with a traditional herbal mixture, **Danggui Longhui Wan**. In 1966, the Institute of Haematology of the Chinese Academy of Medical Sciences identified the active factor in Danggui Longhui Wan, a complex mixture of 11 herbs. **Antitumor activity** was traced to one ingredient, **Qing Dai (Indigo naturalis).** This dark blue powder could be isolated from the leaves of Baphicacanthus cusia, Polygonium tinctorium, Isatis indigotica, Indigofera suffrutticosa, and Indigofera tinctoria. The powder contained a high level of the **blue dye, indigo**.

Indirubin works to stop the uncontrolled growth of tumor cells by inactivating enzymes called **cyclin-dependent kinases (CDKs).** These enzymes regulate cell division. In non-cancerous cells these enzymes normally turn off at an appropriate time. In cancerous cells they stay activated and the cells keep dividing uncontrollably. Indirubin can bind to CDKs, block their activity, and halt the wild cell division.

Researchers at the University of Kaiserslautern in Germany demonstrated that indirubin had significant anti-cancer properties against chronic myeloid leukemia (CML). In addition to CML, indirubin has been approved for clinical trials against chronic granulocytic leukemia (CGL). In one study, **26 percent** of **314 CGL patients** showed **complete remission** and **33 percent showed partial remission** when given **indirubin** treatment. Toxicity was low and side effects included mild stomach pain, diarrhea, nausea, and vomiting. When indirubin was compared to busulfan (a drug for treating cancer) in CGL patients, there was similar efficiency in producing remission, although the drug had a higher rate of complete remissions. However, use of busulfan increases susceptibility to infection and can cause blood problems, both of which present various problems of serious nature for patients with leukemia. **(1,2)**

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2. Jean Barilla, M.S., is one of the nation's foremost medical and nutrition experts. The exciting details of andrographis, its history, use, and future promise can be found in her book the Good Health Guide: Andrographis paniculata: Fighting Cancer and AIDS (Keats Publishing, May 1999).

16. Cruciferous Vegetbles and Indole-3-Carbinol

HOW CRUCIFEROUS VEGETABLES AND THEIR INDOLE-3-CARBINOL CONTENT HELP REDUCE CANCER RISK

James Meschino, D.C., M.S., N.D.

Introduction

There is a growing awareness among health practitioners and the general public about the importance of nutrition and supplementation in the prevention of cancer and other degenerative diseases. A great deal of this attention has focused on various protective nutrients such as antioxidants, various flavonoids, soy isoflavones and dietary fiber. One of the more under-appreciated bioactive agents that is strongly associated with reducing risk of many common cancers is the indole-3-carbinol, found exclusively in cruciferous vegetables and in various indole-3-carbinol-containing supplements. This important biological agent is known to speed up the detoxification of many potentially harmful chemicals (including carcinogens), provide antioxidant support, block the overproduction of certain hormones that are linked to increased risk of breast and prostate cancer, and act as a phytoestrogen (plant-based estrogen), which can bind to estrogen receptors on reproductive tissue and exert anti-cancer influences.

As such, indole-3-carbinol has been shown to be one of the major anti-cancer substances found in cruciferous vegetables. Frequent consumption of these vegetables (broccoli, cauliflower, cabbage, Brussels sprouts, kale and bok choy) is associated with reduced risk of cancer in many human epidemiological studies and in animal experiments. (1- 8) Indole-3-carbinol is a member of the class of sulfur-containing chemicals called glucosinolates (previously called thioglucosides). (9) It is formed from parent compounds whenever cruciferous vegetables are crushed or cooked. (10, 11) Indole-3-carbinol and other glucosinolates (e.g., other indoles and isothiocyanates such as

sulforphane) are antioxidants and potent stimulators of Phase I and Phase II detoxification enzymes in the liver and intestinal epithelial cells. (12, 13, 14)

Detoxification Support

The liver and epithelial cells of the intestinal tract contain the major detoxification centers in of the body, referred to as Phase I and Phase II detoxification. Almost 2 quarts of blood pass through the liver every minute of our lives. Among other functions, liver cells are capable of detoxifying a large number of end products of metabolism, drugs, xenbiotics, hormones and other compounds, including certain carcinogens. The Phase I and Phase II detoxification processes are known to be vital aspects of preventing the accumulation of toxins in the body and neutralizing and eliminating various cancer-causing agents and procarcinogens. The Phase I detoxification enzymes can directly neutralize some dangerous chemicals, but primarily convert most compounds into intermediate end products that must be further acted upon by the Phase II enzyme system. In fact, many of the intermediates formed by Phase I detoxification are more dangerous to the body than were the original compound. Many of these intermediates are free radicals that are known to cause DNA mutations and other damage, and can deplete the liver of its glutathione stores, if sufficient nutritional support for glutathione synthesis is not available. The Phase II detoxification enzymes primarily act to conjugate intermediate end products (formed in Phase I detoxification) with various amino acids and other chemicals that neutralize these intermediates and make them easier for the body to eliminate (e.g., attaching sulfur via sulfation, makes many compounds more water- soluble and easier to eliminate in the urine). Indole-3-carbinol has been shown to be one of the very few exogenous agents that can speed up both Phase I and Phase II detoxification centers in the liver and the intestinal epithelial cells. It has even been shown to improve the function of Phase II glutathione-S-transferase detoxification

activity, which is an extremely important pathway in the elimination of many dangerous chemicals. Many researchers indicate that the ability of cruciferous vegetables to stimulate Phase I and Phase II detoxification, especially their indole-3-carbinol content, is a primary factor in which these nutrients are related to reduced cancer risk in humans. Animal studies have repeatedly shown that when animals are exposed to or injected with carcinogens, the animals receiving the cruciferous vegetables or the indole-3-carbinol in their food supply have a significantly lower tumor yield and incidence than the animals fed the same diet, but without cruciferous vegetables or indole-3-carbinol fortification. (15, 16, 17)

Phytoestrogen Support

Indole-3-carbinol also acts as a phytoestrogen (plant-based estrogens) and in this capacity can bind to estrogen receptors in the body, reducing the ability of stronger estrogens from over stimulating reproductive tissues such as the breast, cervix, uterus, and in males, the prostate gland. Researchers have recently discovered that breast cells, for instance, contain alpha and beta estrogen receptors. The body's estrogens (estradiol, estrone and estriol), estrogen replacement therapy and the estrogen in oral contraceptives primarily stimulate the alpha-receptors, which encourage breast cells (and estrogen-dependent breast cancer cells), to rapidly divide and proliferate. As breast cells divide more rapidly, they are more inclined to make genetic mistakes and allow cancerous mutations to express themselves. This is how high exposure to estrogen, hormone replacement therapy and oral contraceptives are linked to the increased risk of breast cancer. Conversely, phytoestrogens are known to primarily stimulate the beta- receptors on breast cells, which in turn encourage a slower, more controlled cell division rate, which is associated with reducing the risk of breast cancer. Further, phytoestrogens also bind to alpha–receptors, but have only 1/1000 to 1/10,000 the estrogen effect as estradiol and thus compete for binding on these receptors with other more powerful estrogens. In this way phytoestrogens are also capable of toning down the estrogenic influence of more powerful estrogens on various reproductive tissues. This effect also helps to prevent hyperproliferation of breast cells. Epidemiological studies consistently show that a higher ingestion of indole-3-carbinol foods is highly associated with the prevention of reproductive organ cancers in women and men. (3, 4, 8)

Indole-3-carbinol also promotes the metabolism of certain endogenous estrogens (estrone) into a safer, less cancer-promoting form (2-OH-estrone), further helping to reduce risk of reproductive organ cancers, according to modern wisdom. Some women naturally convert more of their estrone hormone to 16-hydroxyestrone, which has been shown to be a biomarker for increased risk of breast cancer, by some researchers. Supplementation with indole-3-carbinol has been shown to alter genetic expression in such a way as to encourage greater activity of the enzyme that converts estrone into 2-hydroxyestrone, which is considered to be protective against breast cancer. Thus, all women may benefit in this regard as the intake of indole-3-carbinol helps to improve the 2-hydroxy to 16-hydroxyestrone ratio. This may be important in men as well from the standpoint of preventing prostate cancer. (18, 19, 20) Thus far, human studies have used

a dose of 300-400 mg per day to demonstrate a significant change in the 2-hydroxy to 16-hydroxyestrone ratio, but a lower dosage may still be effective. (33)

1. **Prevention of Female Reproductive Cancers**

   In experimental animal testing with mice and rats, indole-3-carbinol and brussels sprouts, respectively, have demonstrated an ability to reduce mammary cancer incidence in animals exposed to carcinogens that are known to promote mammary cancer in these species. (21, 22) As mentioned above in human studies, the ingestion of indole-3-carbinol has been shown to increase the metabolism of estrone hormone to 2-hydroxyestrone rather than the 16-alpha-hydroxyestrone metabolite. Studies indicate that 16-alpha-hydroxyestrone is associated with an increased risk of breast cancer in humans and conversely, 2-hydroxyestrone is associated with a reduction in breast cancer risk. Thus, indole-3-carbinol influences the body's enzyme systems in a fashion that favorably influences the 2-hydroxyestrone to 16-alpha-hydroxyestrone ratio, helping to reduce risk of breast cancer. (20, 21, 23, 24,) A large prospective study involving 5,000 Italian women and a second study of patients with either benign or malignant breast lesions highlighted the ability of a higher 2/16 hydroxyestrone ratio to predict, which women were less prone to breast cancer development. (33)

   a. Breast Cancer

      Epidemiological studies and experimental evidence strongly suggests that indole-3-carbinol may reduce breast cancer risk through the above-cited mechanisms. (25, 26, 27, 28) To date there are no human

intervention trials that have tested indole-3-carbinol as a preventive or therapeutic agent against breast cancer.

b. Cervical Cancer
In a 12-week double-blind study, 8 of 17 patients with early-stage cervical cancer given 200 or 400 mg of indole-3-carbinol per day experienced a complete reversal of their condition (29). Animal studies have also shown that indole-3-carbinols can help prevent cervical cancer in the presence of various carcinogens. (30, 31)

2. Respiratory Tract Papillomas

Indole-3-carbinol supplementation reduced or halted the formation of papillomas (precancerous lesions) in 12 out of 18 patients with recurrent respiratory tract papillomas in a small trial. (32)

3. Prostate Cancer

In animal studies, the ingestion of indole-3-carbinol has been shown to inhibit the growth of PC-3-type human prostate cancer cells by arresting their cell division cycle and by promoting apoptosis (programmed cell death). (8) A Seattle study of men living in that city indicated that men consuming three or more servings per week of cruciferous vegetables had a risk of prostate cancer that was 50% lower than men consuming fewer servings of these vegetables, after controlling for other confounding variables. (33) To date, no human intervention trials have tested indole-3-carbinol as a preventive or therapeutic agent against prostate cancer.

Adverse Side Effects And Toxicity

At doses of 800 mg per day indole-3-carbinol has caused dizziness and unsteady gait (signs of nervous system toxicity) in humans and in animal studies. As well, indole-3-carbinol is a powerful stimulator of Phase I detoxification enzymes, and as such it may speed up the detoxification of certain medications, changing their required dosage. However, one challenge study of this kind revealed that indole-3-carbinol intake did not interfere with oral contraceptive medications. (33) Nevertheless, health practitioners and patients should monitor their response to indole-3-carbinol supplementation, if taken at therapeutic doses concurrently with other drugs. According to animal studies, this appears to be especially true in regards to the following medications: (33)

- Testosterone replacement therapy
- Oral contraceptives
- Hormone replacement therapy
- Anti-seizure medications
- Immune-suppressant and anti-viral drugs
- Digoxin

Drug-Nutrient Interactions

1. Antacids and Heartburn Medications (H-2 antagonist drugs)

By reducing stomach acidity these drugs reduce the absorption of indole-3-carbinol. Therefore, they should not be taken at the same time of day or at the same meal. (33)

2. More Rapid Detoxification Of Other Drugs
   As stated above, indole-3-carbinol may speed up the detoxification of any number of drugs due to its stimulation affect on Phase I detoxification centers. Thus, patient monitoring is required with indole-3-carbinol supplementation at the therapeutic doses mentioned previously (300-400 mg per day). (33)

Summary and Conclusion

Despite the lack of extensive human intervention trials, the overall body of evidence strongly suggests that indole-3-carbinol, and possibly other nutrients in cruciferous vegetables, act through various biological means to help defend against certain cancers, particularly reproductive cancers in women and men. Given our high exposure to environmental toxins, additives, pollutants and contaminants that find their way into our body from food, water and air, it is of great importance to realize that indole-3-carbinol ingestion can help to optimize the body's detoxification processes, reducing the potential damage and carcinogenic effects of many of these exogenous agents. The phytoestrogen effects of indole-3-carbinol have been well studied and appear to account for much of its ability to prevent reproductive cancers in animal. Based upon numerous animal experiments, human epidemiological studies, treatment of cervical cancer patients with indole-3-carbinol supplementation, and studies in humans evaluating the influence of indole-3-carbinol on estrone metabolism, it appears likely that indole-3-carbinol may be one of the most important cancer protective nutrients discovered to date. In my view, health practitioners should encourage patients to consume at least three servings per week of cruciferous vegetables and consider ingesting 30-60 mg of indole-3-carbinol as part of a cancer prevention and detoxification-booster supplement cocktail. A growing number of such supplements are now available in the marketplace due to the growing scientific understanding of the important biological activities exhibited by indole-3-carbinol.

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Natural Health Products That Inhibit Angiogenesis

James Meschino DC, MS, ND

The following discussion primarily pertains to the ability of various natural health products to inhibit angiogenesis – the process by which cancer cells sprout new blood

vessels to feed themselves the oxygen and nutrients they require to thrive and metastasize. However, it should be noted that many natural health products that inhibit angiogenesis also influence other molecular pathways associated with cancer, providing effects on multiple targets that help reduce the development and growth of cancer. A potential advantage of phytochemicals and other compounds derived from natural health products (when compared with drugs) is that they may act through multiple cell-signalling pathways and reduce the development of resistance by cancer cells. The primary molecular pathways affected by natural health products include:

- angiogenesis
- epidermal growth factor receptor
- HER2/neu gene
- cyclo-oxygenase-2 enzyme
- nuclear factor kappa-B transcription factor
- protein kinases
- Bcl-2 protein
- coagulation pathways

The Importance of Angiogenesis to Cancer Cells

Human tumours can remain dormant for years because of a balance between cell proliferation and apoptosis. To progress, cancers require a source of nutrition and oxygen. Tumours that outgrow their oxygen supply cannot form masses greater than 1–2 mm without developing central necrosis. A critical part of this process is the induction of local small blood vessels, termed "angiogenesis. Tumours do not grow progressively unless they induce a blood supply from the surrounding stroma. Cancers that lack angiogenesis remain dormant. The induction of new blood vessels provides tumours with a survival advantage. The survival and growth of cells depends on an adequate supply of oxygen and nutrients and on the removal of toxic products. Oxygen can diffuse radially from capillaries for only 150–200 μm. When distances exceed this maximum, cell death follows. Thus, the expansion of tumour masses beyond 1 mm in diameter depends on the development of a new blood supply—angiogenesis.

An increasing density of tumour vasculature raises the probability that the tumour will metastasize. Generally, increased microvascular density ("angiogenesis index") is a significant indicator of poorer prognosis. Angiogenesis plays a central role in the progression of most solid tumours, including those of bladder, brain, breast, cervix, colon, lung, and prostate. Increased vascular density has also been found in the bone marrow of patients with acute myeloid leukemia and myeloma

The balance between factors that stimulate new blood vessel growth and those that inhibit it determines the vascular density. The inhibitory influence predominates in normal tissues; in tumours, many neoplastic cells switch from an angiogenesis-inhibiting to an angiogenesis-stimulating phenotype. That switch coincides with the loss of the wild-type allele of the *TP53* tumour suppressor gene and is associated with reduced production of thrombospondin-1 (TSP-1), a controller of angiogenesis in fibroblasts.

The production of VEGF is considered essential for most cancer cell migration and for angiogenesis. Expression of VEGF messenger RNA is upregulated by many oncogenes (including H-*ras* and K-*ras*, *src*, *TP53*, and c-*jun*) and growth factors including epidermal growth factor, transforming growth factors alpha and beta, insulin-like growth factor–1, and platelet-derived growth factor. The list below outlines the number of endogenous angiogenic polypeptides that have angiogenesis-stimulating effects:

Endogenous Angiogenic Polypeptides

Activator protein 1 (AP-1)

Angiogenin (AG) and angiotropin (AT)

Angiopoietin (APN)

Basic fibroblast growth factor (bFGF)

Cyclooxygenase (COX) and lipoxygenase (LOX)

Granulocyte-colony stimulating factor (G-CSF)

Hepatocyte growth factor (HGF)

Insulin-like growth factors 1 and 2 (IGF-1 and -2)

Interleukin-8 (IL-8)

Nuclear factor kappa B (NF-κB)

Placental growth factor (PGF)

Platelet-derived endothelial cell growth factor (PD-ECGF)

Pleiotrophin (PTN)

Proliferin Thrombospondin-1 (TSP-1)

Transforming growth factor alpha (TGFα)

Transforming growth factor beta (TGFβ)

Tumour necrosis factor alpha (TNFα)

Vascular endothelial growth factor (VEGF)

Vascular permeability factor (VPF)

Neoplasms are able to synthesize or induce some of these polypeptides, an activity that is partly achieved by the secretion of vascular endothelial growth factor (VEGF) and angiopoietins (APNS). Hypoxia stimulates these peptides; the result is a sprouting of endothelial cords. This sprouting creates profuse but immature networks of thin endothelial-lined channels, essential for tumour oxygenation. Although these networks permit progressive tumour growth, they are less efficient than the vascular supply to normal tissues. Tumours often secrete a relative excess of VEGF that results in disorganized and leaky vessels that cause local bleeding and edema.

The growth of many cancers is associated with an absence of the endogenous inhibitors of angiogenesis—for example, interferon beta (INFβ). A potent inhibitor of angiogenesis,

INFβ works by blocking interleukin-8 (IL-8), basic fibroblast growth factor (bFGF), and collagenase type V, which are all potent angiogenic factors that aid tumour development and invasiveness. Thus, the importance of a strong immune system is highlighted in that immune cells secrete various anti-angiogenic interleukins and interferon.

Monoclonal Antibody Drugs That Inhibit Angiogenesis

Recently, targeted therapies using monoclonal antibodies that antagonize the formation of new blood vessels have been developed. One example is bevacizumab (Avastin: Genentech, San Francisco, CA, U.S.A.). Bevacizumab is a genetically engineered humanized monoclonal immunoglobulin G antibody that blocks the VEGF receptor in endothelial cells, thereby shutting off the tumour blood supply. Although bevacizumab increases survival for some patients, it increases the risk of adverse effects, including leucopenia, diarrhea, and hypertension.

Use of bevacizumab is also associated with major risks of thrombosis (resulting in stroke and myocardial infarction), fatal hemorrhage (such as gastrointestinal bleeding or hemoptysis), and visceral perforation. During a course of radiotherapy, some tumours increase their angiogenic activity.

Some anti-angiogenic agents also have anticoagulation activity that may also be associated with a reduction in metastasis. Heparin is a well-known example of a therapy with both anticoagulation and anti-angiogenic activities.

Natural Health Products That Inhibit Angiogenesis In Cancer Cells

Rather than develop multiple monoclonal antibodies to target the multiple peptides and their receptors, an alternative approach might be to evaluate phytochemicals and certain animal-derived chemicals that influence multiple pathways. The science of pharmacognosy evaluates natural drugs derived from herbal remedies or phytomedicines. Natural health products contain a cocktail of biologic chemicals that act on multiple pathways that initiate and maintain tumour angiogenesis. (A great deal of experimental and preclinical evidence suggests that some of these agents represent appropriate interventions in the complimentary management of cancer as well possibly serving a role in cancer prevention):

Curcumin - Curcumin inhibits the transcription of two major angiogenesis factors, VEGF and bFGF. It interacts with VEGF- and nitric oxide–mediated angiogenesis in tumours. Elevated levels of nitric oxide correlate with tumour growth. Curcumin reduces nitric oxide generation in endothelial cells. The membrane-bound enzyme CD13 (aminopeptidase N) is found in blood vessels undergoing active angiogenesis. Curcumin binds to CD13 and blocks its activity, thereby inhibiting angiogenesis and invasion by tumour cell. Derivatives of curcumin are in development by the pharmaceutical industry for this purpose.

Curcumin downregulates the expression of the *VEGF* and *MMP9* genes that are associated with angiogenesis. Curcumin can interfere with the activity of both *MMP2* and *MMP9,* the basis of the angiogenic switch, thereby reducing degradation of the extracellula matrix (ECM) . It also interferes with the release of angiogenic factors that are stored in the ECM. It inhibits growth factor receptors such as EGFR and VEGF receptor and the intracellular signalling tyrosine kinases. This cell signalling system can promote further angiogenesis through gene activation that increases levels of cyclooxygenase-2 (COX-2), VEGF, IL-8, and the MMPs,

Chinese Scullcap and the Baicalein Flavonoid - Baicalin and baicalein are the main derivatives of the Chinese skullcap herb. They are potent anti-angiogenic compounds that reduce VEGF, bFGF, 12-lipoxygenase activity, and MMP. Scutellaria baicalensis is one of the herbs found in PC-SPES, a complex of Chinese herbs that is clinically active against advanced prostate cancer.

Milk Thistle and the Silymarin flavonoids - Silibinin and silymarin are polyphenolic flavonoids isolated from the fruits or seeds of *Silybum marianum (milk thistle).* In the laboratory, silymarin demonstrates strong activity against a variety of tumours by downregulation of VEGF and EGFR. Silymarin suppresses VEGF when used as a single agent against human ovarian cancer.

Quercetin – Quercetin is a flavone found in apples, onions, raspberries, red grapes, citrus fruit, cherries, broccoli, and leafy greens. It inhibits angiogenesis through multiple mechanisms, including interaction with the COX-2 and lipoxygenase-5 enzymes, EGFR, the *HER2* intracellular signalling pathway, and the NF-κB nuclear transcription protein. Quercetin may enhance the anticancer effects of tamoxifen through anti-angiogenesis **(1)**

Quercetin has also been shown to induce apoptosis of cancer cells by upregulating certain death receptors. Cytokines such as tumor necrosis factor–related apoptosis-inducing ligand (TRAIL) can induce apoptosis in colon cancer cells through engagement of death receptors. Nevertheless, evading apoptosis induced by anticancer drugs is common feature of many types of cancers. This results in the need for combination therapy. A study published in 2007 reported that quercetin, through its ability to redistribute death receptors at the cell surface, facilitates death-inducing signaling complex formation and activation of caspases in response to death receptor stimulation. **(2)**

Green Tea and its Polyphenols – Green tea is a rich source of polyphenols and catechins, mainly epigallocatechin-3 gallate (EGCG). These constituents inhibited proliferation of MDA-MB231 breast cancer cells and HUVECS and, in rodent studies, also suppressed breast cancer xenograft growth and reduced the density of tumour vessels. This activity was associated with a decrease in VEGF, regulated at the level of

transcription. In addition, EGCG suppresses protein kinase C (PKC), another VEGF transcription modulator. **(1)**

Sartippour M and fellow researchers showed that ECGC decreased c-fos and c-jun RNA transcripts, suggesting that activator protein (AP)-1–responsive regions present in the human VEGF promoter may be involved in the inhibitory effect of EGCG. Furthermore, EGCG suppressed the expression of protein kinase C, another VEGF transcription modulator, in breast cancer cells. Inhibition of VEGF transcription appeared to be one of the molecular mechanism(s) involved in the antiangiogenic effects of green tea, which may contribute to its potential use for breast cancer treatment and/or prevention.

The inhibition of breast cancer angiogenesis by green tea likely involves multiple pathways other than VEGF transcription. It has been reported in other cell types that green tea inhibits other angiogenic molecules, i.e., urokinase , matrix metalloproteinases (MMP-2 and MMP-9), and platelet-derived growth factor. Tumor necrosis factor-α gene expression has also been shown to be inhibited by EGCG. Other studies suggest that another mechanism involves the suppression of interleukin-8 production by endothelial cells **(3)**

A dose of 1.0 g/m^2 three times daily [equivalent to 7–8 Japanese cups (120 mL)] has been recommended. In practice, lower total daily doses of 2–4 g standardized green tea extract (95% polyphenols and 60% catechins) are usually prescribed. Each gram of this extract provides 400–500 mg of EGCG. The dose-limiting adverse effects are the gastrointestinal and neurologic effects of caffeine. However, the caffeine may potentiate the anti-angiogenic effect of EGCG

Artemisinin (Chinese Worm Wood) - is the active constituent extracted from the plant *Artemisia annua.* Artemisinin has been used clinically as an anti-malaria drug. More recently, it was shown to be cytotoxic to cancer cells through induction of apoptosis. also lowered VEGF expression by tumour cells and *KDR* expression by endothelial cells. Artemisinin also has anticancer activity through other pathways. It inhibits the activation of nuclear factor kappa-B (NF-κB), an important activator protein in cancer development and progression

Viscum album (European Mistletoe) - is also known as Iscador (Weleda, Palisades, NY, U.S.A.). It is often used as an anticancer agent in anthroposophic and homeopathic medicine. Laboratory studies show that it is anti-angiogenic by downregulation of VEGF; it also induces apoptosis of cancer cells.

Shark Cartilage - The resistance of cartilage to tumour formation has been correlated with its capacity to inhibit the formation of new blood vessels. A number of *in vitro* and *in vivo* studies have suggested the existence of anti-angiogenic compounds in shark and

bovine cartilage. The clinical effectiveness of whole cartilage for the treatment of cancer was not confirmed in a recent phase III randomized controlled trial. The main problem is lack of data correlating bioavailability with pharmacologic effects in the oral use of shark cartilage. Unsatisfactory outcomes in clinical trials may be secondary to inadequate bioavailability of the active constituents

Dogfish Shark and Squalamine - Squalamine is a steroid isolated from the liver of the dogfish shark. Squalamine significantly blocks VEGF-induced activation of mitogen-activated protein kinase and cell proliferation in human vascular endothelial cells. Squalamine is anti-angiogenic for ovarian cancer xenografts, and it appears to enhance the cytotoxic effects of cisplatin chemotherapy, independent of *HER2* status. Overexpression of *HER2* is normally associated with resistance to cisplatin and promotion of tumour angiogenesis. In a phase II trial of patients with advanced small-cell lung cancer, squalamine was administered at a dose of 300 mg/m^2 by continuous infusion for 5 days, with paclitaxel and carboplatin given on day 1. Patient survival data and a satisfactory safety profile indicated that the combination should be explored further. **(1)**

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Part III Nutritional Supplements During Chemotherapy: The Evidence To Support The Enhanced Anti-Neoplastic Effects Of Concurrent Therapy

Can Nutritional Supplements Be Used With Traditional Medical Treatments For Cancer?

James Meschino DC, MS, ND

In Brief

A number of studies have shown that various supplements that enhance immune function, stimulate apoptosis of cancer cells and/or acta as anti-angiogenesis agents of

cancer cells, appear to be excellent concurrent therapies to use in conjunction with chemotherapy and radiation treatment during cancer therapy. The primary controversy in this area pertains to the concurrent use of supplements that possess antioxidant effects**.** Several studies suggest that dietary supplementation with antioxidants can influence the response to chemotherapy as well as the development of adverse side effects that results from treatment with antineoplastic agents. Administration of antineoplastic agents results in oxidative stress, i.e., the production of free radicals and other reactive oxygen species (ROS). Oxidative stress reduces the rate of cell proliferation, and that occurring during chemotherapy may interfere with the cytotoxic effects of antineoplastic drugs, which depend on rapid proliferation of cancer cells for optimal activity. Antioxidants detoxify ROS and may enhance the anticancer effects of chemotherapy. For some supplements, activities beyond their antioxidant properties, such as inhibition of topoisomerase II or protein tyrosine kinases, may also contribute to enhancing the antineoplastic action of chemotherapy drugs. ROS cause or contribute to certain side effects that are common to many anticancer drugs, such as gastrointestinal toxicity and mutagenesis. ROS also contribute to side effects that occur only with individual agents, such as doxorubicin-induced cardiotoxicity, cisplatin-induced nephrotoxicity, and bleomycin-induced pulmonary fibrosis. Antioxidants can reduce or prevent many of these side effects, and for some supplements the protective effect results from activities other than their antioxidant properties (e.g. glutamine provides nutrients to intestinal epithelium and coenzyme Q10 preserves function of cardiac mitochondria, preventing cardiotoxiciy associated with anthracyline drugs). Certain side effects, however, such as alopecia and myelosuppression, are not prevented by antioxidants, and agents that interfere with these side effects may also interfere with the anticancer effects of chemotherapy, as interference on this level suggests overriding the anti-proliferative effects of chemotherapy (blood cell proliferation and hair follicle proliferation).

A recent report indicates that approximately 60% of cancer patients use dietary supplements without the knowledge of their oncologist while undergoing cancer treatment. (Prasad 2004)

Cancer Cells Decrease Mitotic Activity In The Presence Of Excess Free Radicals: Making Them Less Susceptible To Chemotherapy Drugs

Cancer cells have "highly evolved" mechanisms to prevent lipid peroxidation. But excess reactive oxygen species (ROS) (primarily induced by chemotherapy drugs) causes excess oxidative stress which results lipid peroxidation in cancer cells (as well as normal cells). This prolongs the G1 phase and may result in cells entering the dormant G0 phase. Thus, excess ROS ("free radicals") interfere with the cytotoxic effects of antineoplastic agents on cancer cells by interfering with a cancer cell's progression through its cell cycle. Tumor cells in G0 are “unaffected” by chemotherapy and can usually reenter the division cycle after chemotherapy is completed. They can also repair damage done by drugs that do not require ongoing DNA synthesis (such as cisplatin).

Since anticancer drugs are effectively ONLY when cells are proliferating rapidly, it stands to reason that oxidative stress, which slows or arrests cell growth, interferes with the effectiveness of chemotherapy. It also explains why slow-growing tumors (such as lung or colon) are relatively unresponsive to chemotherapy.

Oxidative stress, possibly through aldehyde-mediated enzyme inhibition of cyclin-dependent kinases, inhibits transition of cells from the G_0 phase (quiescent phase) of the cell cycle to the G_1 phase, blocks progression through the restriction point, and causes arrest of the cell cycle at the G_1, S, G_2, and M phase checkpoints. For antineoplastic agents that exhibit cell cycle phase-specific activity, e.g., those that interfere with DNA synthesis or block the mitotic process, interference with cell cycle progression may diminish their cytotoxicity. Even platinum coordination complexes and alkylating agents, which are not considered to be phase-specific agents, require cells to progress through the S phase and G_2 phase for apoptosis to occur.

As well, checkpoint arrest also may allow for DNA repair of damage caused by platinum coordination complexes and alkylating agents, and checkpoint abrogation (the opposite of what happens during oxidative stress) has been shown to enhance the cytotoxicity of several types of antineoplastic agents. By reducing aldehyde generation, antioxidants may counteract the effects of chemotherapy-induced oxidative stress on cell cycle progression and enhance the cytotoxicity of antineoplastic agents.

Concurrent Administration Of Antioxidants With Chemotherapy Drugs Enhances Their Antineoplastic Efficacy

As note above, administration of anticancer agents results in a much greater degree of ROS than cancer itself. This high level of ROS during chemotherapy may overcome the antioxidant defenses of cancer cells, which may result in that lipid peroxides reduce or *halt cancer cell proliferation* and thereby interfere with chemotherapeutic agents. Therefore, individuals with a relatively impaired antioxidant status are relatively unresponsive to chemotherapy. A number of researchers suggest giving the patient high doses of vitamin E, vitamin C, selenium, beta-carotene, coenzyme Q10 and other selective antioxidants during chemotherapy. This will prevent the cancer cell from becoming dormant and thus expose it to the cytotoxic effects of the drugs, which enhances the cancer killing effects of chemotherapy drugs

According to Kenneth Conklin, “nutritional therapy with antioxidants during chemotherapy may overcome the growth-inhibiting effects of oxidative stress and maintain responsiveness to anti-neoplastic agents" (Conklin 2000).

Can dietary supplements with antioxidants interfere with the mechanism whereby antineoplastic agents are cytotoxic to cancer cells? According to Conklin, "This is unlikely, since ROS are not involved in the mechanism of action of most anticancer drugs

in current use." **Bleomycin** is an exception to the rule. Thus, it may unwise to recommend antioxidants when bleomycin is the chemotherapeutic agent of choice.

Conklin's article provides not just an excellent review of the data, but a cogent reason why antioxidants are in fact beneficial when used with conventional treatments. While we await additional evidence from well-designed randomized controlled trials on this topic we can reassure patients that the overwhelming mass of data accumulated so far supports the concurrent use of chemotherapy with dietary antioxidants.

Free Radical Assault Is Not The Means By Which Most Cancer Drugs Kill Cancer Cells, But ROS Does Cause Chemo Side Effects

The drugs of many classes of antineoplastic agents are known to generate a high level of oxidative stress in biological systems. These classes of drugs include the anthracyclines, most alkylating agents, platinum-coordination complexes, epipodophyllotoxins, and camptothecins. For these drugs, the hepatic microsomal monooxygenase system is a primary site where ROS are generated, although other enzymatic (e.g., xanthine oxidase) and nonenzymatic (Fenton and Haber-Weiss reactions) mechanisms also play a role. The electron transport system of cardiac mitochondria is another site where significant levels of ROS are generated by anthracyclines .

Although some classes of antineoplastic agents generate high levels of oxidative stress, others, including the taxanes, vinca alkaloids, antifolates, and nucleoside and nucleotide analogues, generate only low levels. Nevertheless, all drugs generate some free radicals.

The mechanism of action of only a few antineoplastic agents has been definitively linked to a free radical intermediate of the parent drug. Examples of antineoplastic agents that have been so linked include mitomycin-C, which creates DNA inter-strand crosslinks following reduction of its aziridine ring, and bleomycin, which cleaves DNA by hydrogen abstraction by its iron-binding arm, which functions as a ferrous oxidase.

Most of the other major classes of antineoplastic agents have well-established mechanisms of action that are independent of free radical intermediates or free radical generation. These include the antifolates and nucleoside and nucleotide analogues that impact DNA synthesis, the vinca alkaloids and taxanes that interfere with microtubule function, the epipodophyllotoxins (etoposide, teniposide) that interfere with topoisomerase II activity, and the camptothecins (topotecan, irinotecan) that interfere with topoisomerase I activity. The platinum coordination complexes (cisplatin, carboplatin, oxaliplatin) and most alkylating agents form strong electrophilic intermediates that act via nucleophilic substitution reactions to form inter- and intrastrand DNA crosslinks. Although toxicity among these agents varies, most side effects also are attributed to nucleophilic substitution reactions, e.g., cisplatin toxicity (nephrotoxicity, neurotoxicity, ototoxicity) is attributable to protein sulfhydryl binding and inactivation of thiol-containing enzymes. Antioxidants that act as reducing agents do not appear to

interfere with the antineoplastic activity of these agents nor do they prevent the development of side effects, which suggests that free radical generation does not play a role in the antineoplastic activity or the toxicity of these agents.

Glutathione, NAC and Thiosulfate May Not Be The Best Choice Of Antioxidants During Chemotherapy

In contrast to antioxidants that act as reducing agents, nucleophiles, such as glutathione (GSH), N-acetylcholine (NAC), and thiosulfate, can bond covalently to electrophilic compounds such as cisplatin, and mixing cisplatin with thiosulfate prior to administration blocks the antineoplastic agent's activity .

However, several animal and clinical studies have shown that intravenous administration of GSH shortly before administration of cisplatin reduces the drug's toxicity without reducing its anticancer activity. This may be explained by the high renal and neural intracellular levels of gamma-glutamyl transpeptidase (GGT), an enzyme that hydrolyzes circulating GSH (GluCysGly) to **Glu**tamine and **Cys**teine-**Gly**cine and transports these products into the cell. The high enzyme levels in normal tissues allow for rapid clearing of circulating GSH, thus preventing cisplatin binding to GSH in the bloodstream, and also results in high GSH levels in renal and neural tissue, thus protecting them from cisplatin toxicity. Renal cells also have unique mechanisms for concentrating selenium and for formation of methylselenol and glutathionylselenol, compounds that also protect the kidneys from cisplatin toxicity.

Most cancer cells have low levels of GGT. Thus, intravenous administration of GSH does not increase the GSH content of most cancer cells that could reduce the antineoplastic activity of cisplatin. However, GTT may be expressed in higher amounts or be inducible in some neoplastic cells. The potential selective protection of normal tissues from cisplatin toxicity warrants further investigation.

How Anthracyclines Work (Doxorubicin) – Intercalation Agents

Anthracyclines are a very commonly used cancer drug in this day and age. Overall, there is evidence to suggest that concurrent antioxidant administration with these drugs enhances their anti-neoplastic effects, by preventing cancer cells from entering into the G_0 quiescent stage. However, administration of these anticancer agents results in a much greater degree of ROS than cancer itself. This high level of ROS during chemotherapy may overcome the antioxidant defenses of cancer cells, which may result in that lipid peroxides reduce or halt cancer cell proliferation and thereby interfere with chemotherapeutic agents. Therefore, individuals with a relatively impaired antioxidant status are relatively unresponsive to chemotherapy. Several mechanisms have been proposed for the anticancer activity of anthracyclines. Although the most studied anthracycline, doxorubicin, alters membrane function, signal transduction such as pathways involving protein kinase C, and many other cellular functions, the most compelling evidence for its primary mechanism of action is via **intercalation with double-stranded DNA and inhibition of topoisomerase II activity**. This effect is

evident at clinically relevant concentrations, with the drug being localized primarily in the nucleus of neoplastic cells and **acting in the S-phase of the cell cycle**. **This supports topoisomerase II inhibition as the drug's primary cytotoxic mechanism.** However, doxorubicin readily undergoes a one-electron reduction to its semiquinone, which can donate an electron to molecular oxygen resulting in superoxide generation. **Although generation of hydroxyl radicals from superoxide is an attractive explanation for the cytotoxicity of doxorubicin, several lines of evidence suggest that this mechanism does not contribute significantly to the drug's anticancer activity**

Cardiotoxicity From Anthracyclines and Its Prevention With CoQ10

Although free radical generation may not play a significant role in the antineoplastic activity of doxorubicin, there is compelling evidence that disruption of the electron transport system and hydroxyl radical generation in mitochondria of cardiac cells accounts for the drug's acute and chronic cardiotoxicity. The selective toxicity of doxorubicin to cardiac cells is accounted for by the unique structure of the cardiac mitochondrial inner membrane, which possesses a cytosolic (outer surface, or intermembranous) NADH dehydrogenase in addition to the matrix (inner surface) NADH dehydrogenase that is present in the mitochondria of all cells. Doxorubicin (a tetracycline ring with a sugar moiety) is hydrophilic and cannot penetrate the inner membrane and be reduced by the matrix enzyme. However, in cardiac mitochondria, doxorubicin, which can penetrate the outer membrane and enter the mitochondrial cytosol, is reduced by the cytosolic NADH dehydrogenase to its semiquinone. Intramolecular rearrangement results in formation of the lipophilic deoxyaglycone of doxorubicin that penetrates the inner membrane where it then inhibits coenzyme Q_{10}-dependent enzymes, accounting for disruption of mitochondrial energetics and resulting in the development of acute cardiotoxicity (arrhythmias and reduced ejection fraction).

The deoxyaglycone also competes with coenzyme Q_{10} (both structurally are quinones) as an electron acceptor, diverting electrons to molecular oxygen with the formation of superoxide radicals, and displaces coenzyme Q_{10} from the electron transport chain, resulting in elevated plasma levels of coenzyme Q_{10}. These effects explain the generation of elevated levels of ROS in cardiac cells that lasts for several weeks following administration of doxorubicin and the high levels of mitochondrial DNA adducts that form in heart mitochondria and result in suppression of mitochondrial gene expression for critical components of the electron transport system, such as coenzyme Q_{10}. These long-lasting effects most likely explain mitochondrial disruption, the first cytological evidence of chronic cardiotoxicity, which leads to myocyte degeneration and **cardiac failure.** Preclinical and a limited number of clinical studies suggest that, whereas antioxidants in general do not prevent doxorubicin cardiotoxicity, **administration of coenzyme Q_{10} does prevent the development of both acute and chronic cardiotoxicity without interfering with the drug's anticancer efficacy**.

Antioxidants Do Not Suppress Apoptosis As Apoptosis Results From Cytochrome c Release, Which Induces Free Radicals As A Result Of Apoptosis: Free Radicals Do Not Induce Apoptosis

One of the primary pathways of drug-induced apoptosis is the pathway that involves release of cytochrome c from mitochondria. When cytochrome c is displaced from the electron transport chain, instead of electrons being transferred to oxygen via cytochrome c oxidase with the formation of water, electrons are diverted from NADH dehydrogenase (Complex I) and reduced coenzyme Q_{10} to oxygen with the concomitant formation of superoxide radicals. Although superoxide is not highly toxic, mitochondrial superoxide dismutase generates hydrogen peroxide from superoxide and, in the presence of reduced iron that is abundant in mitochondria, and highly toxic hydroxyl radicals are formed via Fenton and Haber-Weiss reactions. Thus, all drugs that induce apoptosis by this mechanism generate some degree of oxidative stress, although this does not imply that free radical generation is necessary for a drug to exert its cytotoxic effect on neoplastic cells, because the apoptotic process is initiated by cytochrome c release and superoxide generation occurs secondarily.

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and Cytotoxic Cancer Treatments: A Speech to the 7th International Symposium for Biologically Closed Electric Circuits in Biomedicine. July 19-22, 2001. Helsingør, Denmark.

Specific Antioxidants Of Choice:

Vitamin C - studies support vitamin C daily dosages in the range of 2,000-5,000 mg (Pauling suggests up to 10,000 mg per day – as dictated by bowel tolerance (Prasad 1999)

Vitamin E Succinate – studies support daily dosages in the range of 2,000-3,000 IU

Beta-carotene – studies support daily dosages in the range of 50,000-100,000 IU

Selenium – studies support daily dosages in the range of 600-800 mcg

Coenzyme Q10 – studies support daily dosages in the range of 150-300 mg

Preliminary human studies have shown that the above antioxidants can improve patient outcomes when used in combination with chemotherapy drugs in cases of various types of cancer (small-cell lung cancer; breast cancer stage 0-III, ovarian cancer) – (Prasad

2004). As a single agent Beta-carotene has been shown to reverse oral leukoplakia, likely via its effects on cell differentiation. (Prasad 1999)

Studies suggest that the above antioxidants can reduce recurrence of breast cancer in patients who were previously treated with conventional therapies. (Lockwood 1994; Fleischauer 2003)

Glutamine – glutamine has been shown to enhance the antitumor effectiveness of methotrexate in laboratory animals and reduce the bacteremia and mucosal injury associated with entercolitis of rats given methotrexate. Human studies suggest that glutamine protects the intestinal tract from damage from a variety of chemotherapy drugs. A typical schedule is 6 gm, three times per day (total of 18 grams), beginning three days prior to onset of chemotherapy administration.

Genistein – Genistein inhibits DNA topoisomerase I and II in a similar fashion as the chemodrugs doxorubicin and etoposide – via enhanced topoisomerase IImediated DNA cleavage, resulting in cell cycle arrest. Genistein also inhibits the binding of ATP to its binding site on topoisomerase, which also inactivates the enzyme – These effects lead to inhibition of DNA replication and subsequent antiproliferative effects. (Conklin 2000)

Quercetin – Like genistein quercetin is able to inhibit DNA topoisomerase II, thereby shutting down cancer cell proliferation in experimental studies

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Cancer Diet

According to Prasad et al, a low fat diet (10% calories from fat) and high fiber (25-30 gm) from fruits, vegetables and cereals should be continued during and after cancer treatment. The fat content can be increased to 15% of total calories during complete remission. A low fat diet decreases cancer growth factors such as prostaglandins, leukotrienes and estrogens. A high fiber diet increase bowel production of butyric acid from gut bacteria, which is a powerful anti-cancer agent. Patients must also avoid tobacco smoking or its oral consumption, reduce caffeine intake and drink no more than two glasses of wine weekly. They should also include moderate daily exercise and use stress-reduction techniques. (Prasad 1999)

Part IIII Adjunctive Nutritional Management Considerations In Cancer: Protocols To Consider

James Meschino DC, MS, ND

A significant number of experimental and clinical research studies suggest that certain dietary practices and the use specific nutritional supplements may be of value in helping to prevent cancer recurrence or its progression. These measures are not to be used in place of recommended medical protocols, but rather, may be considered as adjunctive (additional) interventions that may further help in these cases. In all instances, the following considerations should not be employed without the attending physician's knowledge and consent.

Dietary Considerations:

1. Low Animal Fat Diet – evidence suggests that the consumption of high fat animal foods is linked to increased risk of cancer development and recurrence. As an example, breast cancer patients have been reported to be at increased risk of suffering a recurrence if they eat higher levels of fatty foods, such as butter, margarine, red meat, and bacon (Herbert JR, Hurley TG, Ma Yunsheng. The effect of dietary exposures on recurrence and mortality in early stage breast cancer. Breast Cancer Res Treat 1998;51:17–28.). This may be do to the fact that the high levels of saturated fat in these foods can increase secretion rates of various hormones that foster cancer growth or by increasing the secretion of bile acids into the intestinal tract, which are subsequently metabolized by gut bacteria into cancer-causing secondary sterols. High animal fat foods are also a rich source of a polyunsaturated fat known as arachidonic acid, which provides the raw material from which body tissues make a hormone-like substance known as prostaglandin series-2. Prostaglandin series-2 encourages cells to divide at a faster rate, which poses a threat because faster rates of cell division tend to increase risk of cancer development and cancer progression. Thus, consuming a low animal fat diet is a very important consideration for cancer prevention and as a means to reduce cancer recurrence. For

further details on this subject consult the book, The Meschino Optimal Living Program: 7 Steps To A Healthy, Fit, Age-Resistant Body (Wiley Publishing – 2004).

2. Alcohol Avoidance – alcohol may promote cancer development and recurrence by increasing free radical damage, speeding up the delivery of cancer-causing agents into the cells of the body (co-carcinogen role), weakening the immune system, over stimulating the release of certain hormones associated with cancer, and by speeding up rates of cell division. Thus, avoiding alcohol consumption is a strong consideration in these cases. For further details on this subject consult the book, The Meschino Optimal Living Program: 7 Steps To A Healthy, Fit, Age-Resistant Body (Wiley Publishing – 2004).

3. More Cruciferous Vegetables – cruciferous vegetables including broccoli, cauliflower, Brussels sprouts, cabbage, bok choy, and turnips, contain indole-3-carbinol, a substance that helps the body detoxify cancer-causing agents (and other foreign compounds), and has been shown to act in other ways to help lower cancer risk for several types of cancer. One should consider consuming any combination of these vegetables on a daily basis. For further details on this subject consult the book, The Meschino Optimal Living Program: 7 Steps To A Healthy, Fit, Age-Resistant Body (Wiley Publishing – 2004).

Supplement Considerations:

1. High Potency Multi-Vitamin and Mineral – a number of vitamins, minerals and antioxidants have been shown to help suppress the growth of various forms of cancer under experimental conditions and reduce the rate of cancer recurrence in some clinical trials, as outlined below. A high potency multi-vitamin and mineral provides your body with a strong head start in regards to the overall vitamin and mineral supplementation program that may be considered in these cases.

2. Additional Vitamin E Succinate (2,000-3,000 IU per day) – experimental studies demonstrate that vitamin E succinate can help suppress the growth of various types of cancer by inhibiting cancer cell division and by enhancing programmed cell death of cancer cells. (Prasad KN, et al. Effects of tocopherol (Vitamin E) acid succinate on morphological alterations and growth inhibition in melanoma cells in culture; Sigounas G, et al. DL-alpha tocopherol induces apoptosis in erythroleukemia, prostate and breast cancer cells. Nutr and Cancer 1997;28(1):30-5). Taken in conjunction with other antioxidant supplements (vitamin C, selenium, beta-carotene, co-enzyme Q10), several human studies have shown that it may be helpful in reducing the risk of recurrence of some forms of cancer (Lockwood K. et al. Apparent partial remission of breast cancer in "high risk" patients supplemented with nutritional antioxidants, essential fatty acids and coenzyme Q10. Mol Aspects Med 1994;15(Suppl):231S-40S).

3. Additional Vitamin C (2,000-10,000 mg per day) – experimental studies demonstrate that vitamin C can help reduce the risk of cancer recurrence for certain types of cancer by inhibiting the production of cancer-causing nitrosamines, boosting immune function and through other mechanisms of action (Tannenbaum SR, et al. Inhibition of nitrosamine

formation by ascorbic acid. Am J Clin Nutr 1991;53(1):247-50; Anderson R, et al. The effects of increasing weekly doses of ascorbate on certain cellular and humoral immune functions in normal volunteers. Am J Clin Nutr 1980;33:71). Taken in conjunction with other antioxidant supplements (vitamin E, selenium, beta-carotene, co-enzyme Q10), several human studies have shown that it may be helpful in reducing the risk of recurrence of some forms of cancer (Lockwood K, et al. Apparent partial remission of breast cancer in "high risk" patients supplemented with nutritional antioxidants, essential fatty acids and coenzyme Q10. Mol Aspects Med 1994;15(Suppl):231S-40S).

4. Additional Selenium (700-800 mcg per day) – experimental studies indicate that selenium can help suppress the growth of certain cancers by inhibiting the cellular replication of cancer cells and by boosting immune function (Kiremidjian-Schumacher L, Roy M, Wishe HI, Cohen MW, Stotzky G. Supplementation with selenium and human immune cell functions; II, effect on cytotoxic lymphocytes and natural killer cells. Biol Trace Elem Res 1994;41:115-27; Combs, Jr., Gerald F. Impact of Selenium and Cancer-Prevention Findings on the Nutrition-Health Paradigm. Nutrition and Cancer: 40 (1); 6-11). Taken in conjunction with other antioxidant supplements (vitamin E, vitamin C, beta-carotene, co-enzyme Q10), several human studies have shown that it may be helpful in reducing the risk of recurrence of some forms of cancer (Lockwood K, et al. Apparent partial remission of breast cancer in "high risk" patients supplemented with nutritional antioxidants, essential fatty acids and coenzyme Q10. Mol Aspects Med 1994;15(Suppl):231S-40S).

5. Beta-carotene (50,000-90,000 IU per day) – experimental studies indicate that beta-carotene can help reduce the recurrence of certain cancers because it acts as an antioxidant, an immune system modulator and enhances cellular differentiation (increases the maturation) of many human cells. All of these effects are associated with the prevention of cancer and the reversal of some early stage cancers and states of dysplasia (pre-cancerous states) (Santos MS, Meydani SN, Leka Lwu D, Fotouhi N, Meydani M, et al. Natural killer cell activity in elderly men is enhanced by B-Carotene supplementation. Am J Clin Nutr 1996;64:772-7; Stich HF, Rosin MP, Vallejera MO. Reduction with Vitamin A and Beta-Carotene administration of proportion of micronucleated buccal mucosal cells in Asian betel nut and tobacco chewers. Lancet 1984;1:1204-6; Stich HF, Rosin MP, Vallejera MO. Reduction with Vitamin A and Beta-Carotene administration of proportion of micronucleated buccal mucosal cells in Asian betel nut and tobacco chewers. Lancet 1984;1:1204-6). Taken in conjunction with other antioxidant supplements (vitamin E, vitamin C, selenium, co-enzyme Q10), several human studies have shown that it may be helpful in reducing the risk of recurrence of some forms of cancer (Lockwood K, et al. Apparent partial remission of breast cancer in "high risk" patients supplemented with nutritional antioxidants, essential fatty acids and coenzyme Q10. Mol Aspects Med 1994;15(Suppl):231S-40S).
It should be noted that high dose beta-carotene supplementation should not be used by smokers or in the adjunctive management of lung cancer, based upon findings from two clinical trials.

6. Coenzyme Q10 (150-300 mg per day) – experimental studies indicate that coenzyme Q10 may help reduce the recurrence of certain cancers because it acts as an antioxidant, immune system modulator and exhibits tumor suppressive effects. Taken in conjunction with other antioxidant supplements (vitamin E, vitamin C, selenium, beta-carotene), several human studies have shown that daily supplementation with 390 mg of coenzyme Q10 may be helpful in reducing the risk of recurrence and progression of some forms of cancer (Folkers K, Shizukuishi S, Takemura K, et al. Increase in levels of IgG in serum of patients treated with coenzyme Q10. Res Commun Pathol Pharmacol 1982;38:335-8; Lockwood K, Moesgaard S, Yamamoto T, Folkers K. Progress on therapy of breast cancer with vitamin Q10 and the regression of metastases. Biochem Biophys Res Commun 1995;212:172-7).

7. Reishi Mushroom Extract - (typically 250 mg, four times per day, standardized to 10-12.5% polysaccharide content) - animal cancer studies have shown a 50% tumor regression outcome with Reishi Mushroom Extract treatment (e.g., connective tissue cancer model in mice). Reishi Mushroom Extract is used by some cancer surgeons in Japan to treat cancer patients and significant anti-tumor and immunostimulation effects have been noted in many of these cases. Polysaccharides from reishi mushrooms and from other types of folk-medicinal fungi are patented in Japan for use as immunomodulators in the treatment of cancer. They are combined with chemo- and radiotherapy and have demonstrated an ability to reduce side effects, increase the efficacy of treatments, and to accelerate recovery from disease.
Studies from China have shown that Reishi Mushroom Extract potentiates the tumor-killing action certain immune cells. Reishi Mushroom Extract is known to have other immune modulating effects and antioxidant properties.

Animal studies also show that the polysaccharide fraction of reishi mushrooms can induce apoptosis (programmed cell death of cancer cells) in leukemic cells and induce cellular differentiation in 40-45% of leukemic cells treated with reishi polysaccharides, demonstrating significant cancer treatment potential. These effects were primarily due to the increased secretion of anti-tumor cytokines (signaling agents) induced by Reishi Mushroom polysaccharides, namely TNF-alpha and IFN-gamma, and these two cytokines acted synergistically on the inhibition of leukemic-cell growth. In a related experiment, the D-glucan polysaccharide fraction of Reishi Mushroom was shown to produce dramatic tumor regression in a mice sarcoma study. In many animals, complete tumor regression occurred in the group injected with beta-D glucan fractions within a 5-week period. The study by Y. Sone et al, reported tumor inhibition rates of 90% and tumor regression in 75% of afflicted animals.

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8. Astragalus (500 mg, three times per day; 2:1 extract) - experimental studies have shown that the active constituents in astragalus, primarily its triperpene glycosides and polysaccharide content, can modify immune function and increase the ability of immune cells to recognize and destroy cancer cells. A more detailed explanation of these activities would include the following: the active ingredients in astragalus have been shown to significantly increase the proliferation of lymphocytes, enhance interferon and interleukin-2 production and activity- two powerful signalling agents that enhance the effectiveness of immune cells, activate T cell blastogenesis, increase T cell cytotoxicity, enhance the secretion of the immune modifying chemical known as tumor necrosis factor (TNF), enhance phagocytosis by immune cells, increase natural killer cell cytotoxicity – the ability of these white blood cells to destroy developing cancer cells, viruses and other pathogens, increase the activity of peritoneal macrophages, and provide direct anti-viral effects.

In human studies astragalus has also been used to reduce the side effects of chemotherapy and radiation treatment. A large clinical study of 572 cancer patients

demonstrated that astragalus supplementation was able to protect adrenal cortical function during radiation and chemotherapy treatment. It also helped to greatly minimize bone marrow depression and gastrointestinal side effects, such as nausea, vomiting and intestinal tract ulcerations in these patients.

In patients with very low white blood cell counts, as a side effect of drugs, radiation or chemotherapy, astragalus supplementation has been shown to help significantly increase the number of circulating white blood cells, helping to restore normal function of the immune system in these severely immune-compromised patients

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reinforcing the Qi (vital energy.) Journal of Traditional Chinese Medicine. 1980;3:p67

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9. Chinese Scullcap (150 to 200 mg per day of pure Baicalein Flavonoid derived from Chinese scullcap) - many experimental studies indicate that the baicalein flavonoid found exclusively in Chinese scullcap prevents and inhibits cancer growth via a number of direct and indirect physiological actions:

Baicalein has been shown to inhibit the 12-lipoxygenase enzyme, which converts arachidonic acid into a hormone-like substance that is required for cancer cells to replicate. Studies demonstrate that, by inhibiting the 12-lipoxygenase enzyme, baicalein has been shown to inhibit cancer cell proliferation and induces apoptosis (programmed cell death) of many different human cancer cells. Experimental evidence suggests that, in the presence of baicalein, various cancer cells can be prevented from multiplying and metastasizing to other tissues and that a primary mechanism through which this occurs is via the inhibition of 12-lipoxygenase enzyme activity.

Baicalein has also demonstrated an ability to slow or inhibit the replication rate of many different cancers under experimental conditions, which appears to be due to its ability to suppress the release of enzymes required for cancer cell division (protein tyrosine kinase activity and protein kinase C activity).

Baicalein is also a strong antioxidant, and has been shown to protect DNA from undergoing cancerous mutations in challenge studies using potent carcinogens such as benzo{a}pyrene, and toxins such as aflatoxin (AF) B.

In vitro evidence indicates that baicalein stimulates recombination and repair of damaged DNA, supporting its traditional inclusion in cancer formulas, and suggesting possible use after sunburn and radiation damage.

Baicalein has also been shown to inhibit the 5alpha-reductase enzyme, which converts testosterone to dihydrotestosterone (DHT). DHT is strongly associated with the development of prostate enlargement (benign prostatic hyperplasia) and prostate cancer. As such, baicalein is reported to be potentially useful for the prevention and/or treatment of androgen-dependent (testosterone-driven) disorders, including prostate enlargement and prostate cancer.

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10. Curcumin – (480 mg, three time per day) – curcumin has been shown to inhibit epidermal growth factor receptors and the 12 lipoxygenase enzyme system, which are commonly upregulated in cancer cells. This upregulation leads to uncontrolled cellular proliferation, and indirectly inhibits apoptosis and increases angiogenesis. Animal and human studies indicate that curcumin can decrease proliferation rate of certain types of cancer.

11. Essential Fatty Acids (combination of borage seed, flaxseed and fish oil) – this combination of oils provides the body with essential fatty acids that are used by the body to make local hormones (prostaglandin series 1 and 3), which are known to slow rates of cell division and reduce inflammatory reactions. Both of these activities are known to play a role in cancer prevention. The daily therapeutic dosage to be considered would be 4-6 capsules of a 1200 mg essential oil capsule, with each capsule containing 400 mg each, of borage seed, flaxseed and fish oil.

12. Genestra's Water Soluble Vitamin A– 100,000 IU for three months, followed by an on-going maintenance dosage of 50,000 IU per day.

Volumes of research show that vitamin A can suppress the development and progression of many different cancers. Vitamin A encourages cells to develop and mature, and can encourage cancer cells to look and act more normal and with less malignant behavior.

A more detailed accounting of the research shows that vitamin A-like substances are known to possess antiproliferative, differentiative, immunomodulatory and apoptosis-inducing properties. A growing body of evidence supports the hypotheses that the vitamin A receptor B_2 gene is a tumor suppressor gene, and that the cancer prevention effects of vitamin A compounds are due to induction of this receptor. A unique Vitamin A compound is presently being used in cancer prevention and treatment. This form of Vitamin A, known as 9-cis-retinoic acid, has been used to suppress premalignant oral lesions and prevent the development of secondary primary cancers among patients with head and neck and lung cancers. This form of Vitamin A is now being considered in the treatment of breast cancer, which often displays under-expression of the retinoic acid (vitamin A) receptor B2. Note that vitamin A in high doses can be very toxic and thus, if one considers the use of high dose vitamin A supplementation then it should be with a water-soluble form of vitamin A. The vitamin company known as Genestra makes a water soluble vitamin A product. For more details on this product they can be contacted by phone: 1-800-263-5861((A-Mulsion Liquid, code 101 in catalogue)

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13. Vitamin D (2,000 IU per day) – studies indicate that many human breast cancer cells posses vitamin D receptors on their outer membrane (outer skin of the cell). Stimulation of these receptors by vitamin D has been shown to increase the maturation of these cells (makes them look more normal), reduces their malignant behavior and slows down their rate of cell division. In experimental studies, Vitamin D has been shown to suppress cancer cell proliferation, induce cancer cell apoptosis (programmed cell death), and differentiation, demonstrating a strong potential role in the prevention and management of colon, breast and prostate cancer.

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Breast Cancer Add:

1. Ground Flaxseed – 50 gm per day (2 heaping tablespoons) – enterolactone and enterodiol formed from lignan precursors found in flaxseeds, inhibit key enzymes that convert androgens and estrogens into estradiol – the most potent estrogen driving cellular proliferation. Enterolactone and enterodiol also compete with other estrogens for binding to estrogen receptors, toning down that estrogenic influence on breast cells, which in turn encourages decreased proliferation. This is similar to the effects of tamoxifen.
2. Soy Isoflavones – 200 mg per day – soy isoflavones inhibit key enzymes that convert androgens into estrogen, compete with other estrogens for binding to

estrogen receptors, and bind with higher affinity to beta estrogen receptors than alpha- estrogen receptors, all of which tend to decrease cellular proliferation. An important study demonstrated that soy isoflavone supplementation decreased the mitosis to apoptosis ratio in the tumors of women with breast cancer. However, there is still some controversy about the use of soy isoflavones for with women with a history of reproductive organ cancers, and thus, physician approval should be sought before making this recommendation.

3. Melatonin – 10-20 mg per day (requires physician monitoring) – preliminary studies in Italy suggest that melatonin can suppress cancer growth of breast cancer. Preliminary laboratory and clinical evidence also suggests that melatonin may enhance the effects of some chemotherapy drugs used to treat breast cancer. In a study that included a small number of women with breast cancer, melatonin (administered 7 days before beginning chemotherapy) prevented the lowering of platelets in the blood. This is a common complication of chemotherapy, known as thrombocytopenia that can lead to bleeding.

In another study of a small group of women whose breast cancer was not improving with tamoxifen (a commonly used chemotherapy medication), adding melatonin caused tumors to modestly shrink in over 28% of the women. However, this is a high dosage and physician monitoring is required with this intervention.

Melatonin References:

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2. Lissoni P, Barni S, Meregalli S, Fossati V, Cazzaniga M, Esposti D, Tancini G. Modulation of cancer endocrine therapy to melatonin: a phase II study of tamoxifen plus melatonin in metastatic breast cancer patients progressing under tamoxifen alone. *Br J Cancer* . 1995;71(4):854-856.
3. Lissoni P, Paolorossi F, Tancini G, et al. A phase II study of tamoxifen plus melatonin in metastic solid tumour patients. *Br J Cancer* . 1996;74(9):1466-1468.
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Prostate Cancer Add:

1. Prostate Support Nutrients – it is known that prostate cancer cells divide and spread through the body under the influence of dihydrotestosterone hormone. Certain natural herbal agents have been shown to block the build up of dihydrotestosterone, some of which have demonstrated an ability to slow or retard the progression of prostate

cancer and/or lower or slow the rate of rise in blood levels of the prostate specific antigen (PSA), which is an indicator of prostate cancer progression.

For this reasons prostate cancer patients (including those who have had their prostate gland removed due to the presence of prostate cancer) should consider taking a combination product containing the following natural prostate support nutrients: Saw Palmetto, Pygeum Africanum, Beta – sitosterol, Stinging Nettle, and Lycopene. The daily dosage to be considered would include:

A. Saw Palmetto – this herb should be a standardized grade of 45% fatty acids and sterols, taken at a dosage of 720 mg, twice daily (or an 85-90% standardized grade of fatty acids and sterols, taken at a dosage of 160 mg, twice daily).

B. Pygeum Africanum – this herb should be a standardized grade of 12-14% triterpenes, taken at a dosage of 200 mg, twice daily.

C. Beta –Sitosterol – this plant-based substance should be taken at a dosage of 120 mg, twice daily

D. Soy Isoflavones – this special class of flavonoids derived from soybeans should be taken at a minimum dosage of 50 mg per day (up to 200 mg per day)

E. Stinging Nettle – this herb should used concurrently with other prostate support nutrients at a minimum dosage of 60 mg, twice daily, using 5:1 extract.

F. Lycopene – lycopene is a carotenoid found in tomatoes and other red and pink fruits and vegetables. It accumulates in the prostate gland once ingested orally from food and supplements. There is some evidence that lycopene may be helpful in the treatment of prostate cancer. In one study, 26 men with prostate cancer were randomly assigned to receive lycopene (15 mg twice a day) or no lycopene for three weeks before undergoing prostate surgery. Prostate tissue was then obtained during surgery and examined. Compared with the unsupplemented men, those receiving lycopene were found to have significantly less aggressive growth of cancer cells. In addition, a case report has been published of a 62-year-old man with advanced prostate cancer who experienced a regression of his tumor after starting 10 mg of lycopene per day and 300 mg of saw palmetto, three times per day.

Prostate Support References:

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4. Hussain M, Banerjee M, Sarkar FH et al. Soy isoflavones in the treatment of prostate cancer. 2003. Nutr and Cancer, 42;2: 111-117

5. Boccafoschi and Annosica, S. Comparison of Serenoa repens extract (saw palmetto) with placebo by controlled clinical trial in patients with prostatic adenomatosis. Urologia 1983;50:1257-1268

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7. Can men avoid prostate cancer? A brief review of diet and the prostate. Nutrition Health Review: The Consumer's Medical Journal 1995;72:3

8. Dufour B, Choquenet C. Trial controlling the effects of Pygeum africanum extract on the functional symptoms of prostatic adenoma. Ann Urol 1984;18:193-195

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10. McCaleb R. Synergistic action of pygeum and nettle root extracts in prostate disease. Herbalgram 1996;40:18

11. Mitchell J, et al. Effects of phytoestrogens on growth and DNA integrity in human prostate tumor cell lines: PC-3 and LNCaP. Nutr and Cancer 2000;38(2): 223-228

12. Naik HR, et al. An in vitro and in vivo study of anti-tumor effects of genistein on hormone refractory prostate cancer. Anticancer Res. 1994;14:2617-20

2. Ground Flaxseed – 50 gm per day (2 heaping tablespoons) - enterolactone and enterodiol formed from lignan precursors found in flaxseeds, inhibit key enzymes that convert androgens into estrone hormone and estrone into estradiol. It has been shown that these forms of estrogen encourage prostate cancer development and progression by decreasing the break down of dihydrotestorsterone. Enterolactone and enterodiol may also compete with other estrogens for binding to estrogen receptors on prostate cells, toning down that estrogenic influence on prostate cancer cells, which in turn, slows proliferation rate of these cells

3. Soy Isoflavones – 100- 200 mg per day – a study showed that men with prostate cancer who ingested 100 mg per day of soy isoflavones showed a slower rise in their blood PSA levels. In this study men with existing prostate cancer, who were supplemented with 100 mg per day of soy isoflavones showed a favorable outcome in stabilizing PSA levels. Soy isoflavone supplementation was shown to decrease the rate of rise in serum PSA (prostate specific antigen) levels in patients with androgen dependent and androgen-independent prostate cancer. The

researchers concluded that their data suggests that soy isoflavones may benefit some patients with prostate cancer by slowing the progression of the disease and therefore, potentially delaying the development of symptoms, improving quality of life, and perhaps even prolonging survival.(Hussain M, Banerjee M, Sarkar FH et al. Soy isoflavones in the treatment of prostate cancer. 2003. Nutr and Cancer, 42;2: 111-117)

4. Modified Citrus Pectin - Modified Citrus Pectin is a dietary supplement that has demonstrated an ability to prevent the spread of cancer (metastasis), with strong evidence to support its use in the prevention and/or management of prostate cancer metastasis. Modified Citrus Pectin is a special form of pectin that has been altered in the laboratory by a proprietary process that shortens the length of pectin's polysaccharide chain. One of the dominant carbohydrates contained within Modified Citrus Pectin is galactose. Galactose has a strong affinity for binding to the surface of metastatic cancer cells, which express a particular cell surface receptor known as galectin-3 (a galactoside-binding lectin). In turn, the binding of Modified Citrus Pectin to the galectin-3 receptor on metastatic cancer cells creates a type of galectin-3 blockade. With the galectin-3 receptor blocked in this fashion, cancer cells are less able to adhere to other healthy tissues and cells, essentially inhibiting cancer cells from invading and spreading to new areas and tissues in the body (anti-metastatic effect). As well, the blockade of the galectin-3 receptor prevents cancer cells from adhering to each other, discouraging their ability to form colonies (tumor mass). If cancer cells are deprived of their own adhesive ability, they fail to thrive and can be more easily destroyed by the body's immune system. Thus, Modified Citrus Pectin has been shown to attach to galectin-3 receptors on metastatic cancer cells, preventing their clustering and colonization into a larger tumor mass and blocking their ability to spread to other tissues.
A pilot study involving prostate cancer patients who failed first-line androgen-deprivation therapy, were in relapse after radical prostatectomyexternal beam radiation therapy or cryosurgery, and were either untreated or off intermittent hormone blockade, demonstrated that supplementation with 15 gms per day of Modified Citrus Pectin (5 gms, three times per day) increased the length of the PSA doubling time by 30% in 4 of 7 patients, one patient had a partial response, one patient had stable disease and one patient did not respond. The researchers conclude that Modified Citrus Pectin appears to slow the PSA doubling time in prostate cancer patients with low levels of PSA, and that all patients were still alive three years after the official end of the study. The therapeutic dosage is typically 5 gms, three times per day. Dilute Modified Citrus Pectin powder in a favorite beverage. Up to 30 gms per day may be taken safely. It is also available in capsules and tablets.

1. Eliaz I. The role of modified citrus pectin in the prevention of cancer metastasis. Townsend Letter for Doctors & Patients, Jul99(192):p64

2. Kidd PM. A new approach to metastatic cancer prevention: modified citrus pectin (MCP), a unique pectin that blocks cell surface lectins. Alternative medicine Review Jan 1, 1997;1(1):4-10

3. Strum S, Scholz M, McDermed J. Modified citrus pectin slows PSA doubling time: A pilot clinical trial. Presentation: International Conference On Diet and Prevention of Cancer. Tampere, Finland. May 1999.

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5. Melatonin -10-20 mg per day (requires physician monitoring) - preliminary studies in Italy suggest that melatonin can suppress cancer growth of breast prostate cancer in human subjects. (Lissoni P, Cazzaniga M, Tancini G, Scardino E, Musci R, Barni S, Maffezzini M, Meroni T, Rocco F, Conti A, Maestroni G. Reversal of clinical resistance to LHRH analogue in metastatic prostate cancer by the pineal hormone melatonin: efficacy of LHRH analogue plus melatonin in patients progressing on LHRH analogue alone. *Eur Urol* . 1997;31(2):178-181.)

Colon Cancer Add:

1. Calcium (1200-2,000 mg per day) - A high fat diet, particularly saturated fat, is associated with a higher incidence of colorectal cancer in most countries studied. A higher fat intake results in greater secretion of bile acids into the small intestine (to emulsify the fat in the gut), and in turn results in greater concentrations of bile acids reaching the large intestine. These bile acids are metabolized by large bowel bacterial and converted into the cancer-causing secondary sterols, namely lithocholic and deoxycholic acids. Observational studies and experimental animal studies suggest that higher calcium intake can reduce risk of colo-rectal cancer by binding to bile acids in the gut, forming an insoluble calcium soap that is unavailable for conversion to secondary sterols by gut bacteria. Additionally, calcium and Vitamin D have been shown to slow the rate of cell division of colonic epithelial cells, which is another biomarker suggestive of a cancer protective effect. A number of small intervention trials involving high-risk human colon cancer and polyp-prone subjects have shown that supplementation with calcium (calcium carbonate), Vitamin D and/or wheat bran fiber, can improve the appearance of colon cells and reduce the tendency toward malignant behavior in a manner which is consistent with a reduction in risk of colo-rectal cancer development and recurrence.

Approximately, twenty recent human studies, including 8 cohort studies of colorectal cancer and 2 case-control studies of almost 2000 colon cancer cases reported an inverse association between calcium intake and overall risk of colorectal tumors. In addition, three recent randomized clinical trials showed that calcium supplementation reduced recurrence of colon cancer. Baron et al found a significant 15% reduction in adenoma recurrence after 4 years of calcium supplementation at 1200 per day. Bonithon-Kopp et al found a 34% reduction in adenoma recurrence after 3 years of supplementation at 2000 mg per day. Hofstad et al found a significant reduction in new adenoma formation in

polyp-bearing patients after 3 years of supplementation with a combination of antioxidants and calcium at 1600 mg per day.
In the largest study published to date, Peters et al showed that individuals who ingested calcium supplements at more than 1200 mg per day had a 27% reduction in risk of developing colon ademoma, compared to nonusers of calcium supplements. After adjusting for known risk factors, they also showed that adenoma risk was 12% lower in individuals consuming more than 1767 mg per day of calcium (from food and supplementation), compared to individuals with a total daily consumption of calcium below 731 mg. The National Health and Nutrition Examination Survey III conducted in 1988-1994 suggests that median intake for calcium from food among the American adult population is only 631 mg per day.

Calcium References:

1. Bonithon-Kopp C, Kronborg O, Giacosa A, et al. Calcium and fibre supplementation in prevention of colorectal adenoma recurrence: a randomized intervention trial. European Cancer Preventiionon Organisation Study Group. Lancet 2000;356:1300-6

2. Hofstad B, Almendingen K, Vatn M, et al. Growth and recurrence of colorectal polyps: a double-blind 3-year intervention with calcium and antioxidants. Digestion 1998;59:148-56

3. Peters U, Chatterjee N, McGlynn A, et al. Calcium intake and colorectal adenoma in a US colorectal early detection program. Am J Clin Nutr. 2004;80:1358-65

4. Baron JA, Beach M, Mandel JS, et al. Calcium supplements for the prevention of colorectal adenomas. Calcium Polyp Prevention Study Group. N Engl J Med 1999;340:101-7

2. Wheat Bran Supplementation (11-15 grams per day) – in conjunction with vitamin D and calcium supplementation, the addition of 11-15 grams per day of wheat bran to the diet of individuals with previous colon cancer, or with genetically inherited susceptibility to rectal polyps, has been shown to improve the appearance of colon cells and reduce the tendency toward malignant behavior in a manner which is consistent with a reduction in risk of colo-rectal cancer development and recurrence. For more information on this subject consult the book, The Meschino Optimal Living Program: 7 Steps To A Healthy, Fit, Age-Resistant Body (Wiley Publishing, 2004)

Wheat Bran References:

1. De Cosse JJ, Miller HH, Lesser ML. Effect of wheat fiber and vitamins C and E on rectal polyps in patients with familial adenomatous polyposis. J Natl Cancer Instit 1989 Sep 6;81 (17):1290-7

2. Alberts DS, Einspahr J, Rees-McGee S, Ramanujam P et al. Effects of dietary wheat bran fiber on rectal epithelial cell proliferation in patients with resection of colorectal cancer. J Natl Cancer Instit 1990 Aug 1;82 (15):1280-5

*Curcumin (480 mg, three time daily) – as noted in the general cancer report, curcumin possesses important anti-cancer properties and has been shown to inhibit the recurrence of familial adenomatous polyposis – an aggressive form of colon cancer. Curcumin is a strong consideration for all cancers and should not be overlooked especially in regards to the adjunctive management of colo-rectal cancers.

Oral Leukoplakia

You indicated that you have a history of oral leukoplakia, which is a precancerous condition found in the mouth. Leukoplakia appears as white spots on the mucous membranes of the tongue and inside of the mouth. Individuals with leukoplakia should refrain from alcohol intake, smoking, the use chewing tobacco and the chewing of betel nut. In addition a number of clinical trials have been undertaken, which have shown that supplementation with certain nutrients can help to reverse leukoplakia.

Supplement Considerations:

1. Beta –carotene – a number of clinical trials have shown that a daily dosage of 100,000 to 150,000 IU of beta-carotene supplementation per day can reverse oral leukoplakia in a significant number of cases. Beta-carotene encourages cellular differentiation, meaning that it encourages these precancerous cells to mature in a more normal way, reversing their precancerous appearance and behavior and transforms them back to more normal-looking cells, with a lower tendency toward malignant degeneration.

References:

1. Stich HF, Rosin MP, Hornby AP, et al. Remission of oral leukoplakias and micronuclei in tobacco/betel quid chewers treated with beta-carotene and with beta-carotene plus vitamin A. Int J Cancer 1988;42:195–9.

2. Garewal HS, Katz RV, Meyskens F, et al. β-Carotene produces sustained remission in patients with oral leukoplakia. Arch Otolaryngol Head Neck Surg 1999;125:1305–10.

3. Liede K, Hietanen J, Saxen L, et al. Long-term supplementation with alpha-tocopherol and beta-carotene and prevalence of oral mucosal lesions in smokers. Oral Dis 1998;4:78–83.

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6. Van Eenwyk J, Davis FG, Bowne PE. Dietary and serum carotenoids and cervical intraepithelial neoplasia. Int J Cancer 1991;48:34-8

7. Liu T, Soong SJ, Wilson NP, Craig CB, Cole P, Macaluso M, et al. A case control study of nutritional factors and cervical dysplasia. Cancer Epidemiol Biomarkers Prev 2. 1993;2(6):525-30.

8. Stich HF, Rosin MP, Vallejera MO. Reduction with Vitamin A and Beta-Carotene administration of proportion of micronucleated buccal mucosal cells in Asian betel nut and tobacco chewers. Lancet 1984;1:1204-6.

2. Combining Beta-carotene with other antioxidants - the combination of beta-carotene and vitamin E has led to complete or partial remissions in six of eight trials studying people with leukoplakia. In one trial, administration of 50,000 IU of beta-carotene, 1,000 mg of vitamin C, and 800 IU of vitamin E per day for nine months led to improvement in 56% of people with leukoplakia, with stronger effects in those who also stopped using tobacco and alcohol (Garewal H. Antioxidants in oral cancer prevention. Am J Clin Nutr 1995;62(suppl):1410S–6S [review]; Kaugars GE, Silverman S Jr, Lovas JG, et al. A clinical trial of antioxidant supplements in the treatment of oral leukoplakia. Oral Surg Oral Med Oral Pathol 1994 Oct;78:462–8).
It should be noted that high dose beta-carotene supplementation should not be used by smokers or in the adjunctive management of lung cancer, based upon findings from two clinical trials.

3. Vitamin E - One trial used 400 IU of vitamin E two times per day on its own, as a treatment for oral leukoplakia. After 24 weeks, 46% showed some improvement in signs or symptoms of leukoplakia or related conditions and 21% showed microscopic evidence of improvement.
(Benner SE, Winn RJ, Lippman SM, et al. Regression of oral leukoplakia with alpha-tocopherol: a community clinical oncology program chemoprevention study. J Natl Cancer Inst 1993;85:44–7.)

Cervical Dysplasia

Cervical dysplasia describes abnormal cervical cells discovered upon pap smear analysis. Cervical dysplasia is usually graded according to its severity, which can range from mild inflammation to precancerous changes to localized cancer. Cervical cancer is a common, sometimes fatal disease. It is now known that human papilloma virus is the major cause of cervical dysplasia, which is transmitted via sexual intercourse. More than 90% of cervical cancers contain DNA of the higher risk HPV viruses and that DNA has been found to be present in the early stage lesions of cervical cancer.

There are no symptoms of cervical dysplasia until the disease has progressed into advanced cancer. Therefore, it is crucial that sexually active women, or women over age 20, have yearly Pap smears until the age of 65. Women who experience bleeding between menstrual periods, bleeding after intercourse, abnormal vaginal discharge, abdominal pain or swelling, urinary symptoms, or pelvic pain should be evaluated by a healthcare provider, even if it is not the regular time for a Pap test. The presence of cervica dysplasia requires medical attention in all cases.

From a lifestyle standpoint, cigarette smoking increases the risk of cervical dysplasia, and increases the likelihood that mild forms of dysplasia will progress to more severe forms. Women who become sexually active at an early age and have multiple sexual partners are at increased risk for developing cervical dysplasia. For those who are sexually active, using barrier methods of contraception, such as a condom or diaphragm, is associated with reduced risk of cervical dysplasia.

A superior level of nutritional status of vitamin A, carotenoids such as lycopene, vitamin C, vitamin E, and folic acid have been reported to be associated with a reduced risk of cervical dysplasia.

Supplement Considerations: In the early stages of cervical dysplasia doctors sometimes take a "wait and see what happens" approach, known also as "watchful waiting". During this period it may be beneficial to consider an aggressive supplementation program involving nutrients that have shown promise in either preventing or reversing this condition. Supplementation considerations in this case include:

1. High Potency Multi Vitamin and Mineral – this will provide a foundation of nutrients that are important to the normal development of cervical cells and provides a boost in levels of antioxidants and B-vitamins that are associated with the prevention of cervical dysplasia and the promotion of healthy cervical cells.

2. Additional Beta-carotene----------50,000-90,000 IU per day (Palan PR, Mikhail MS, Basu J, Romney SL. Plasma levels of antioxidant beta-carotene and alpha-tocopherol in uterine cervix dysplasias and cancer. *Nutr Cancer* l991;15:13–20.; Ho GY, Palan PR, Basu J, et al. Viral characteristics of human papillomavirus infection and antioxidant levels as risk factors for cervical dysplasia. *Int J Cancer* 1998;78:594–9; Wylie-Rosett

JA, et al. Influence of vitamin A on cervical dysplasia and carcinoma in situ. Nutr Cancer. 1984;6(1):49-57).

Note that individuals who are smokers and individuals diagnosed with lung cancer should refrain from using high doses of beta-carotene (no higher than 10,000-15,000 IU), according to the implications of two clinical studies.

3. Folic acid--------5-10 mg per day (note that this is a very high intake of folic acid and requires physician consent and monitoring) (Butterworth CE Jr, Hatch KD, Gore H, et al. Improvement in cervical dysplasia associated with folic acid therapy in users of oral contraceptives. *Am J Clin Nutr* 1982;35:73–82; Zarcone R, Bellini P, Carfora E, et al. Folic acid and cervix dysplasia. *Minerva Ginecol* 1996;48:397–400; Butterworth CE, Hatch KD, Soong S-J, et al. Oral folic acid supplementation for cervical dysplasia: A clinical intervention trial. *Am J Obstet Gynecol* 1992;166:803–9; Butterworth CE Jr, Hatch KD, Macaluso M, et al. Folate deficiency and cervical dysplasia. *JAMA* 1992;267:528–33).

4. Additional Vitamin E----------400 IU per day (Palan PR, et al. Plasma levels of antioxidant beta-carotene and alpha-tocopherol in uterine cervix dysplasias and cancer. Nutr Cancer. 1991;15(1):13-20).

5. Additional Lycopene-25 mg per day (Kantesky PA, Gammon MD, Mandelblatt J, et al. Dietary intake and blood levels of lycopene: association with cervical dysplasia among non-Hispanic, black women. *Nutr Cancer* 1998;31:31–40; VanEenwyk J, Davis FG, Bowen PE. Dietary and serum carotenoids and cervical intraepithelial neoplasia. *Int J Cancer* 1991;48:34–8).