Curcuminoids

antioxidant Phytonutrients

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Curcuminoids-Pharmacological Actions Including Preclinical and Clinical Evaluations

The use of *Curcuma longa* to treat a variety of inflammatory, biliary and respiratory disorders, has generated scientific interest in the curative properties of the turmeric rhizome.
volatile oil and the active principles, the curcuminoids.

Most researchers used alcoholic extracts of the rhizome.
a diversity of biological effects and initiated further investigation into the mechanism of these processes. Of prime importance was the discovery of the antioxidant property of curcumin, which is largely responsible for its wide range of pharmacological activities.

1. Antioxidant properties of turmeric and curcumin

During the physiological process of respiration, inhaled and tissue incorporated oxygen oxidized cellular components and biomolecules. This
toward excessive activity by external factors, such as nutrition, or internal factors such as disease, results in the generation of oxidative products known as free
These free radicals have highly reactive chemical molecules, which react with biological compounds causing tissue damage by a process called "free radical

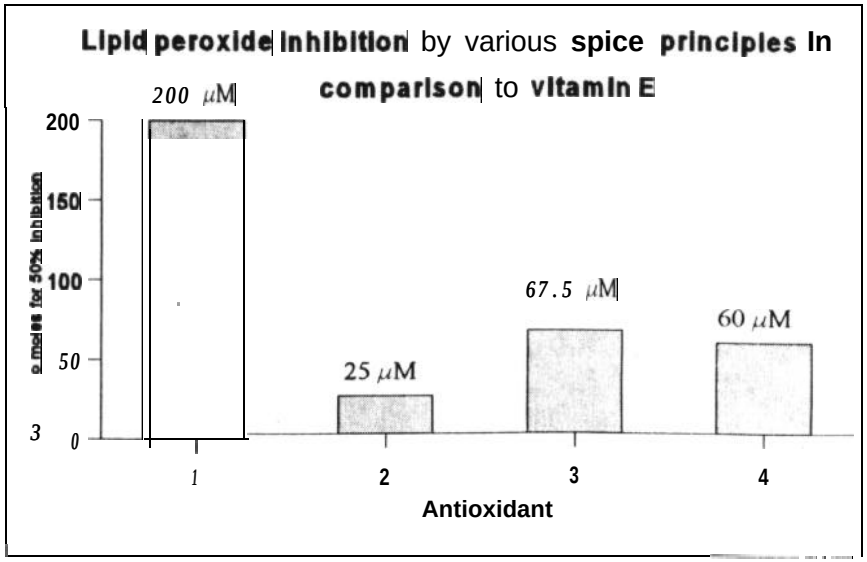
The sequence of changes resulting from an injury (due to burns, thermal shock, radiation, etc.) are as follows?

1. The sensation of pain
2. Phagocyte activation and the production of damaging free radicals.
3. Arachidonic acid release and enzymatic peroxide formation.
4. Metal ion release from storage sites.
5. Release of haem protein which may react with peroxides to promote free radical damage.
6. Depletion of antioxidant defense mechanisms, such as GSH (glutathione) from the cells.
7. Oxidative stress.

The aging process exemplified the cumulative result of deterioration of individual cells, tissues and organs, caused and promoted by free radicals. The human body has built-in mechanisms to counteract free radicals. These mechanisms are collectively known as the body's antioxidant defense reaction. Unfortunately, in most instances, the antioxidant defense is gradually overwhelmed by the **aging** process, or a disease, or both. The inflammatory processes associated with microbial or viral infections and the progression of cancer are just a few disease conditions which contribute to depletion of the antioxidant defense system of the body. Therefore, it is important to preserve the body's defenses against damages by free radicals. Some vitamins, minerals, and natural compounds such as phenolics, flavonoids and carotenoids, have the ability to counteract free radical damage by scavenging or neutralizing the free radicals. These diversified groups of nutrients, micronutrients and food supplements belong to a category of biologically important substances known as "antioxidants."

Curcuminoids are natural phenolic compounds, with potent antioxidant properties. Several research groups have recently provided convincing evidence for the antioxidant properties of cumuminoids. Both turmeric and curcuminoids inhibited generated of potent free radicals like superoxide and hydroxyl radicals.* The antioxidant properties of curcumin in prevention of lipid peroxidation, another process that generates free radicals, is well recognized.[27,71]

Studies on spice principles as antioxidants in the inhibition of lipid peroxidation of rat liver microsomes revealed that curcumin was a potential antioxidant.*



1. Vitamin E 2. Curcumin 3. Eugenol 4. Capsaicin (Ref 8)

Among the spice principles tested, curcumin showed the highest ability to prevent lipid peroxidation. Curcumin, in this study, was found to be eight times more powerful than vitamin E.

The biological effects of curcuminoids to counteract free radicals, have been assessed in animal models with chemically induced inflammation and swelling. Inflammation is known to be associated with increased levels of lipid peroxides and free radicals, which are generated by the liver as well as by the inflamed tissued in the body. Lipid peroxidation is essentially a "free radical chain reaction" involving the following stages?

1. Initiation

Hydrogen is abstracted from a polyunsaturated fatty acid side chain by a free radical species:

- CH+R* → C*+RH
- $C^{*}+O_2$ →

2. Propagation

The fatty acid side chain peroxyl radical -CO,* attacks adjacent fatty acid side chains.

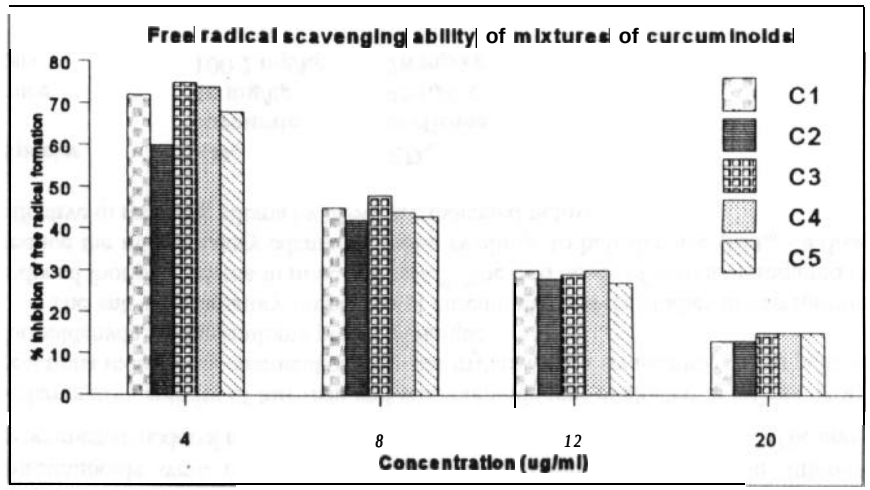
- CO,* + -CH → $-CO_2H$ + -C*

The propagation continues, leading to the accumulation of lipid peroxides in the membranes, destabilizing them and permitting the entry of damaging ions. Peroxyl radicals attack ions as well as membrane proteins. An antioxidant terminates the propagation of free radicals either by accepting and quenching them or by retarding the "initiation" step by reducing the generation of free radicals.

Examples of Free Radicals

Name	**Formula**	**Formation / effect *in vivo***
Trichloromethyl	CCl_3*	During metabolism of CCI, in the liver and contributing to the toxic effects of this solvent.
Hydroxyl	OH*	Attacks all molecules in the human body
Peroxyl, alkoxyl	RO,', RO*	Formed by several routes especially during the breakdown of organic peroxides.
Oxides of nitrogen	NO*	Formed in ***vitro*** from the amino acid L-arginine.
	NO,*	From cigarette smoke

In another set of studies by Sabinsa Corporation, it was shown that the composition of curcuminoid mixtures may not affect their free radical quenching ability, as evidenced in the figure below:



(Ref **17)**

The curcuminoid formulations contained curcumin (C), bisdemethoxycurcumin (BDMC) and demethoxycurcumin @MC) in the following proportions:

C-l 78.6% C + 2.2% BDMC + 16.7% DMC
C-2 86.6% C + 1.9% BDMC + 8.3% DMC
C-3 80.2% C + 2.5% BDMC + 15.5% DMC
C-4 70.8% C + 4.5% BDMC + 8.5% DMC
C-5 67.2% C + 2.8% BDMC + 14.8% DMC

The results of the various experiments validate the existence of two distinct modes of antioxidant action of curcuminoids:

1. **The preventive mode,** preventing free radical formation, (measured by the Rancimat method).
2. **The intervenient mode,** whereby preformed free radicals are quenched by the curcuminoids, (measured by the DPPH-radical scavenging method).

It can therefore be concluded that carefully balanced compositions of curcuminoids would provide optimal antioxidant activity and the subsequent bioprotective effect.

Another research project conducted by Sabinsa Corporation indicated that the addition of micromolar concentration of piperine can potentiate the antioxidant activity of curcuminoids.

Further analysis of turmeric has shown that besides the phenolics, turmeric is also a source of a water soluble peptide with antioxidant properties." The water soluble compound from turmeric was isolated and identified as turmeric, a heat stable noncyclic peptide, having 40 amino acid residues. It is resistant to the proteolytic action of trypsin and pepsin, Turmeric is present in turmeric at a concentration of 0.1%. Turmeric is reported to be a more potent antioxidant than curcuminoids or butylated hydroxyanisole (BHA). This compound is active even at nanomolar concentrations, while curcumin and BHA are active at micromolar concentrations. Since turmeric is rich in the sulfur-containing amino acid, methionine, a known antioxidant, this may, in part, explain the strong antioxidant properties of the compound.

The antioxidant mechanism of curcuminoids may include one or more of the following interactions:

1) scavenging or neutralizing of free radicals;
2) interacting with oxidative cascade, and preventing its outcome;
3) oxygen quenching, and making it less available for oxidative reactions;
4) inhibition of oxidative enzymes like cytochrome P-450; and,
5) chelating or disarming oxidative properties of metal ions such as iron, (Fe).

In conclusion turmeric and its active constituent curcumins or curcuminoids, and the water-soluble peptide, turmeric, have antioxidant properties and effectively inhibit the free radical damage to biomolecules both in *vitro* and *in vivo* conditions. The fact that curcuminoids act as antioxidants by prevention and intervention processes, makes them very unique natural antioxidants.

2. Anti-inflammatory activity or turmeric extract and curcuminoids

Inflammation results from the complex series of actions and/or reactions triggered by the body's immunological response to tissue damage. Many diseases as well as physical trauma, including surgery, induce inflammatory reactions. These reactions, although necessary to start the healing process, too often create an unbearably painful condition, which may even perpetuate the disease.

Arachidonic acid is a compound metabolized in the body to yield important hormone-like substances which play major roles in the process of inflammation. Arachidonic acid can be converted by the action of the enzyme cyclooxygenase to prostaglandins (PG) and thromboxanes (TX), and by action of another enzyme lipoxygenase to hydroxyeicosatetraenoic acids (HETE) and leukotrienes (LT). Some of the prostaglandins like PGE, and PGI_2 dilate the blood vessels, while certain leucotrienes LTR,, LTC_4, and LTD_4 increase vessel permeability resulting in tissue swelling, which characterized inflammation. Increased levels of some prostagalandins like PGE produces redness, swelling and pain of the inflamed part of the body (cardinal signs of inflammation), Other factors like TXA_2, LTC, and LTD., contribute to inflammation by cutting-off the blood and nutrient supply to the tissue.[135]

Steroidal drugs like cortisone, and non-steroidal anti-inflammatory drugs (NSAID) like phenylbutazone and indomethacin, are used in clinical practice to subdue the inflammation. Some of the anti-inflammatory drugs inhibit the lipoxygenase pathway, some the cyclooxygenase pathway, resulting in different potency and clinical application for the anti-inflammatory drugs.[73] Unfortunately, most of the anti-inflammatory drugs, particularly steroids, besides being effective, can also produce dangerous side effects such as changes in blood pressure levels.

Curcuminoids and other constituents of turmeric are well known for their anti-inflammatory activity. Turmeric is one of the oldest anti-inflammatory drugs used in Ayurvedic medicine.- In fact, it was in India that the research on the anti-inflammatory properties of turmeric was initiated. Turmeric extract, volatile oil from turmeric and curcuminoids were reported to possess anti-inflammatory activity in different experimental model of inflammation in mice, rats, rabbits and pigeons.[12,14,76] The anti-inflammatory activity of turmeric and curcuminoids was evaluated in inflammatory reactions induced by chemical or physical irritants like carrageenin, cotton pellets, formaldehyde, and granuloma pouch technique.

The anti-inflammatory properties of curcuminoids were studies in carrageenin induced foot paw edema in mice and rats.[20] The oral doses of curcumin required to reduce the inflammatory edema, or tissue swelling, to half the size ($ED_{,}$ - a dose effective in reducing edema by 50%) are indicated below:

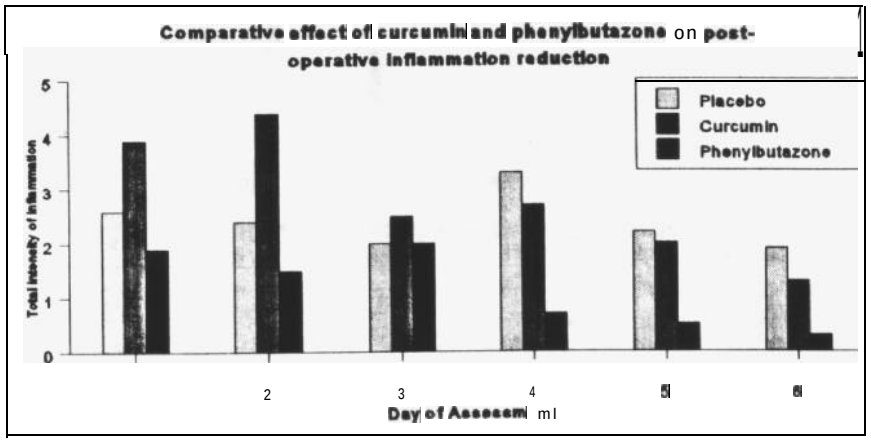
Species	ED_{50} curcumin	ED_{50} cortisone
mice	48 mg/kg	45mg/kg
rats	100.2 mg/kg	78 mg/kg.

Utilizing cotton pellet and granuloma pouch tests in rats, both curcumin and non-steroidal drug, phenylbutazone, were effective at $ED_{,}$ dose of 48 mg/kg.[20]

The anti-inflammatory activity of curcumin was evaluated in a group of patients who underwent surgery or suffered from trauma.[75] A double-blind controlled trial in

which three groups of patients received curcumin (400 mg), a placebo (250 mg of lactose powder) or phenylbutazone (100 mg) respectively, three times a day for five consecutive days after surgery (far hernia or hydrocele). The treatment with curcumin resulted in reduced inflammation and was as equally effective as the treatment with phenylbutazone."

Turmeric has also been evaluated in the treatment of inflammation associated with various forms of arthritis. Oral administration of curcumin at a dose of 3 mg/kg and sodium curcumin at a dose of 0.1 mg/kg inhibited formalin-induced arthritis in rats. Curcumin was comparable effective to phenylbutazone in arresting formalin-induced arthritis in rats.[21] Oral administration of 0.1 mg/kg of volatile oil isolated from *Curcuma longa* decreased arthritis induced in rats by Freud's adjuvant.[76]



The antirheumatic properties of curcuminoids were tested in a double-blind clinical trial in 49 patients with diagnosed rheumatoid arthritis.[22] Curcumin administered at a dose of 1200 mg/per day for five to six weeks, produced a significant improvement in all patients. All patients showed overall improvement in morning stiffness, and physical endurance. The therapeutic effects were comparable to those obtained with phenylbutazone. Both compounds, curcumin and phenylbutazone, failed to reduce some inflammatory indices such as erythrocyte sedimentation rate (ESR).

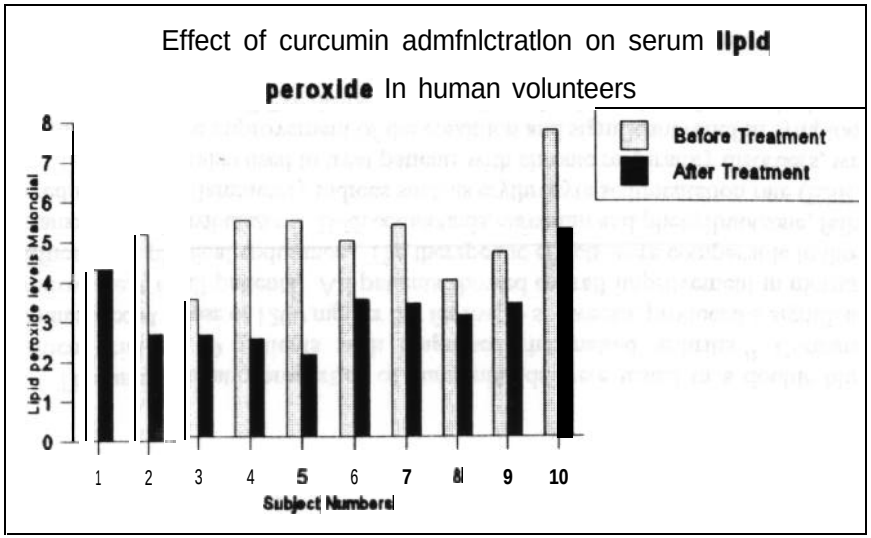
Turmeric was also used to treat patients with chronic respiratory disorders, with resulting subjective improvement of the condition and significant relief in symptoms like cough and dyspnea.[78] Eye drops prepared from the decoction of turmeric, known as "Haridra Eye Drops," were used in 25 cases of an inflammatory condition of the eye, bacterial conjunctivitis.[79] Clinical symptoms such as eye redness or burning sensation, started subsiding from the third day of treatment. The cure rate evaluated

during the six-day treatment was 23 out of 25 patients completely relieved from the condition.

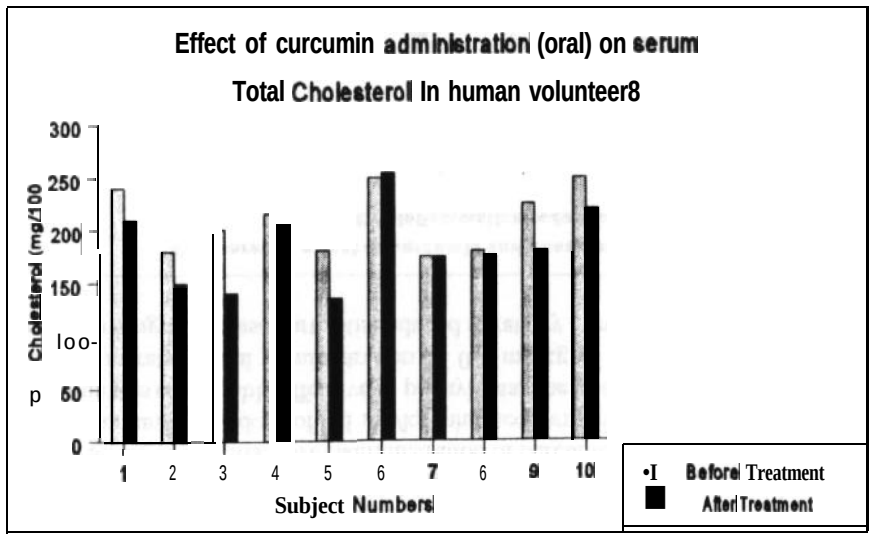
Curcumin and its four synthetic analogs were examined for anti-inflammatory potential in carrageenin induced foot paw edema and cotton pellet granuloma models of inflammation in rats.[77,117] The anti-inflammatory potency of tested curcumin, curcumin analogs and phenylbutazone were established in the following order: sodium curcumin > tetrahydrocurcumin > curcumin > phenylbutazone and triethylcurcumin. Sodium salt of curcumin was effective at half the dose of the parent compound, curcumin. Comparison of curcumin and its analogs in acute and subacute models of inflammation revealed that curcumin analogs are more active in alleviating acute inflammation.

Animals fed with curcumin showed decreased levels of lipid peroxides and subsequent reduction in the chemically-induced inflammation.[26,72] Thus, it is obvious from these studies that curcumin prevents the production of tissue-damaging free radicals. Also, under in *vitro* conditions, in tissue culture, rat and mouse liver cells incubated in the presence of micromolar concentrations of curcumin reduced the generation of lipid peroxides.[26] Curcumin also prevented the oxidative damage and alteration of the DNA genetic material in cultured fibroblasts.[71]

A potential role of curcumin in preventing oxidative damage to the arterial wall has been studied. Cardiovascular disease is caused by the progressive narrowing of the arterial walls. This is essentially due to the deposition of cholesterol plaque inside the arterial walls resulting from high levels of oxidized cholesterol in the blood. Oxidation of blood cholesterol is evaluated in clinical studies by measuring blood levels of lipid peroxides. Administration of 500 mg of curcuminoids daily to healthy humans for seven days lowered the levels of blood lipid peroxides by 33%, as well as the levels of blood cholesterol by 29%. The authors of this study indicate a possible use of curcuminoids in the prevention of cardiovascular disease. The key data from this study are presented below:



Significant: p<.005 (Ref 18)



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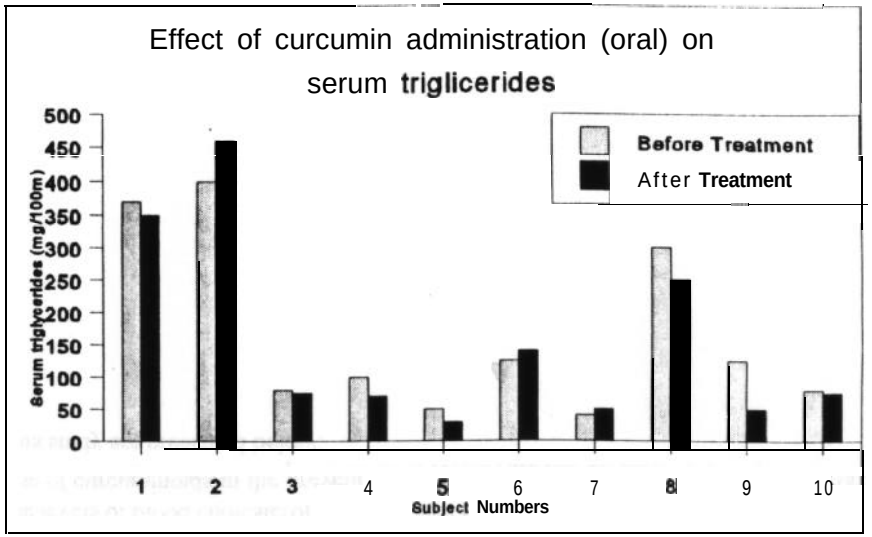

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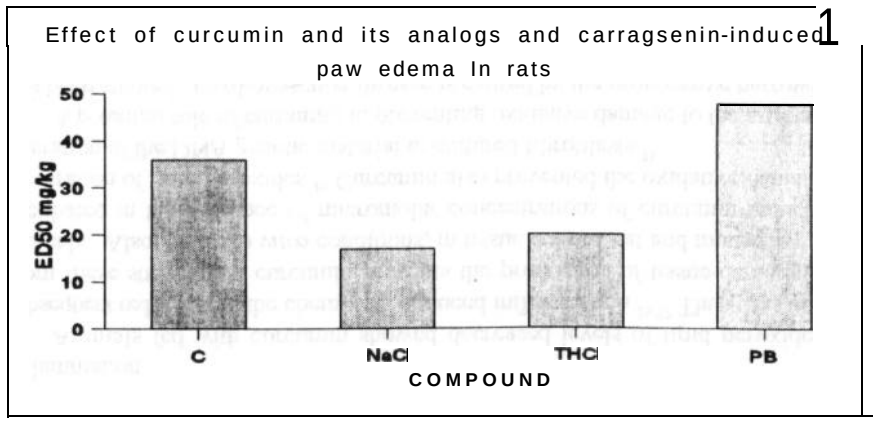


(Ref. 77)

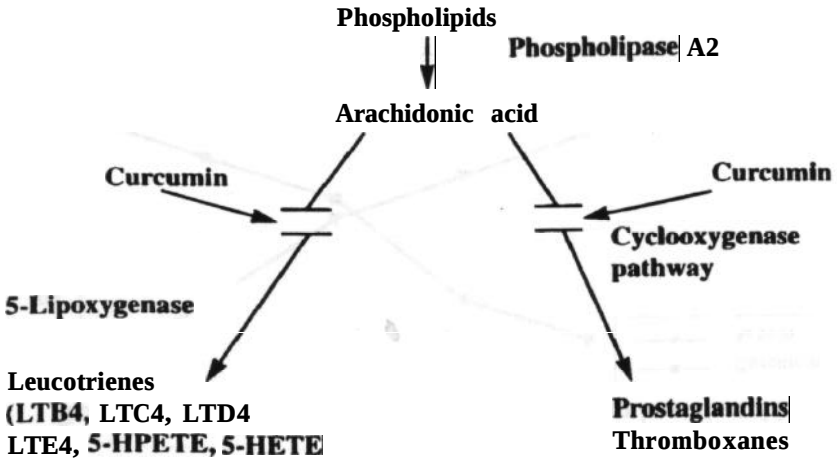
C: Curcumin
NaC: Sodium Curcuminate
THC: Tetrahydrocurcumin
PB: Phtnylbutazone

The improved anti-inflammatory activity of sodium curcuminate, as well as other curcumin salts like potassium and calcium curcumin, may be related to improved water solubility of curcumin salts in comparison to curcumin alone."

Studies correlating chemical structure of curcumin with its biological activity showed that the presence of olefenic double bonds at C 3,4 and C 3', 4' positions, and hydroxy groups at C8, 8' positions in the benzene rings are important for the anti-inflammatory activity of curcumin.[77] On the other hand, blocking the para hydroxy groups in curcumin molecule still did not diminish its anti-inflammatory property in preventing tissue release of histamine-the important inflammatory substance. The critical structure for anti-histamine activity has been identified as ketone groups present in side chain of the curcumin molecule." These have been indicated in an earlier figure.

One of the better understood mechanisms of the anti-inflammatory action of curcumin is its inhibition of cyclooxygenase and lipoxygenase enzymes - a group of enzymes which coordinate metabolism or arachidonic acid in the body.[135]

Arachidonic acid cascade. Inhibition by Curcumin.

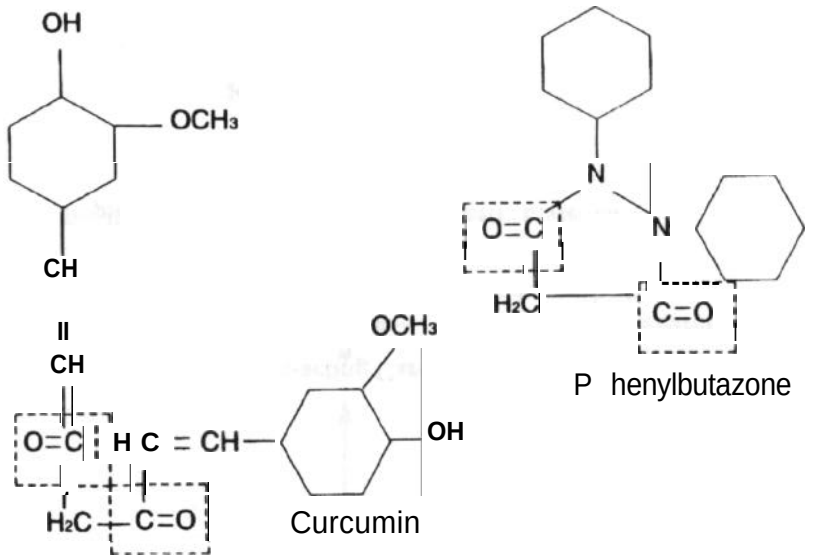


This enzymatic inhibition may be a result of diminishing inflammatory products of the arachidonic acid metabolism, e.g., prostaglandins, leukotrienes and 5-hydroxyeicostate-traenoic acid.[23,24,25] Curcumin has similar action-as that of aspirin and aspirin-like anti-inflammatory agents.[81] However, there is an important advantage of curcumin over aspirin since curcumin, unlike aspirin, selectively inhibits synthesis of inflammatory prostagalandin thromboxane (TXA_2), while not affecting the synthesis

of, prostacyclin (PGI_2).[81] Prostacyclin is an important factor preventing vascular thrombosis and any drug that affects its synthesis, particularly if used in large doses, may increase the risk of vascular thrombosis. Curcumin may, therefore, be preferable in patients who, for example, are prone to vascular thrombosis and require anti-inflammatory and a&arthritic therapy.

The antioxidant properties of curcumin may also contribute to the overall anti-inflammatory action of the compound. As an antioxidant, curcumin is known to scavenge hydroxyl radicals generated by the inflammatory-response cells neutrophils and it also inhibits the production of lipid peroxides, which fuel the inflammatory process.[25,26,27]

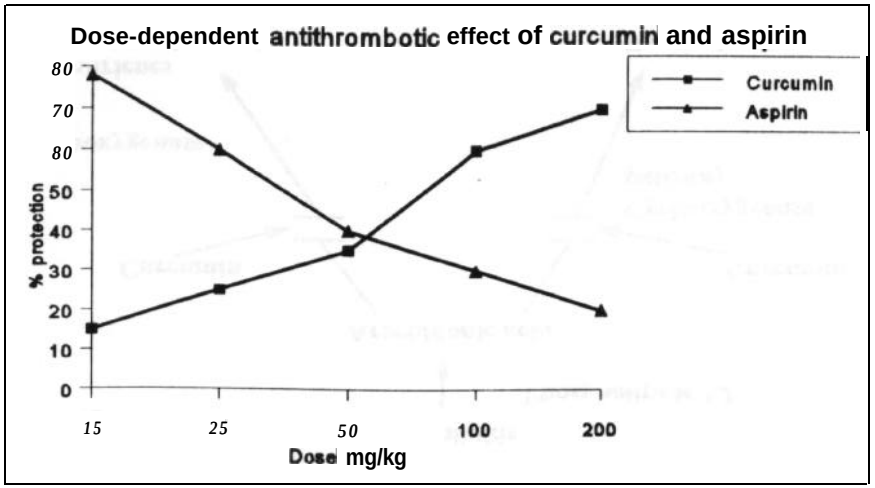
The anti-inflammatory mechanisms of curcumin in many ways compare to those of the NSAID drug phenylbutazone. Structurally, curcumin bears resemblance to phenylbutazone. Like phenylbutazone it has two aromatic groups, with two ketonic groups in the side chain."



3. Antithrombotic activity of curcumin

Platelet aggregation plays a vital role in initiation of thrombosis. In thrombosis, aggregation of platelets obstructs blood flow, leading to coronary diseases and hampered delivery of nutrients to the tissues. In a comparative study on the protective

effects of curcumin and aspirin in mice subjected to induced thrombotic challenge, curcumin was found to exhibit dose-related antithrombotic effect.[119] Curcumin, like aspirin, was found to inhibit cyclooxygenase activity of platelets and platelet thromboxane B_2 (TXB_2) levels. However, curcumin did not affect vascular prostacyclin (PGI,) synthesis. The protective effect of curcumin was directly proportional to the dose up to a level of 200 mg/kg as intraperitoneal administration, whereas the same was inversely proportional in the case of aspirin administration.



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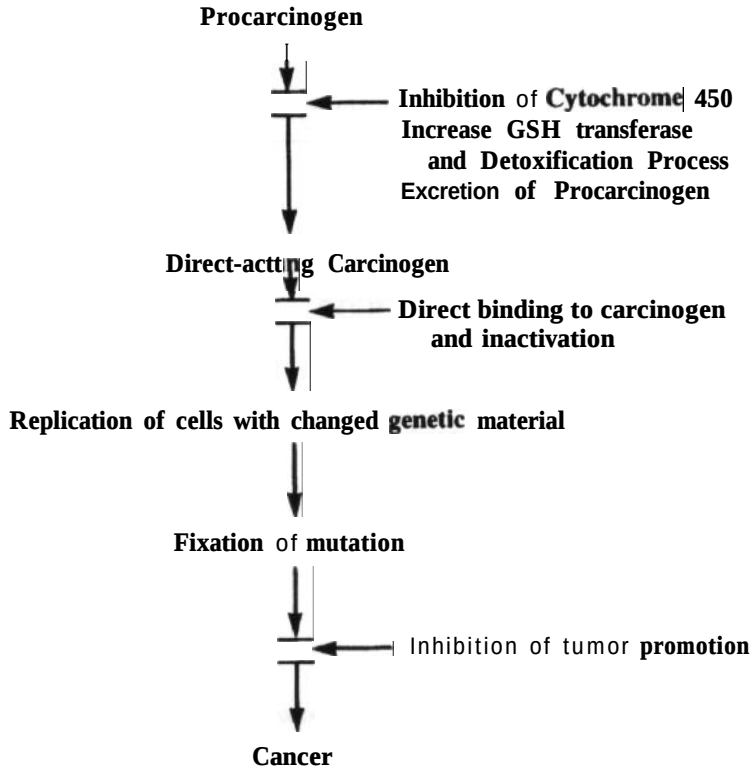
4. Turmeric extract and curcumin in prevention and treatment of cancer

Cancer-causing agents (carcinogens) belong to a diversified group of chemicals, originating from food, food contaminants, food additives, food processing agents, environmental pollutants, synthetic chemicals, pharmaceutical drugs and cosmetics. The development of cancer is a multistep process, involving many factors rather than a single one. Cancer-causing agents can either initiate **tumor,** promote tumor, or act as complete carcinogens having both tumor initiating and promoting activity. Likewise, cancer preventing agents or anticarcinogens, may prevent tumor initiation or promotion stages in tumor development.

Turmeric extracts and curcuminoids have been found to be cancer preventing compounds in different tumor models, as well as in limited human studies.

Their possible modes of action are outlined below:

Known Sites of Action of Curcumin in Prevention of Chemically-Induced Carcinogenesis In Experimental Animals



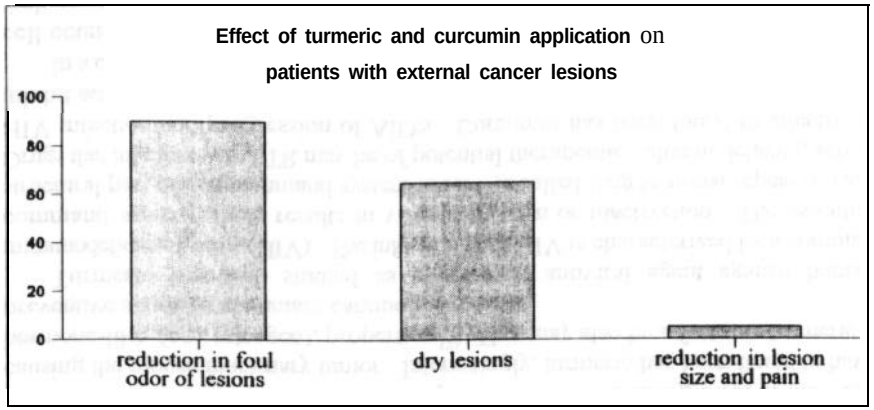
❑ e **Indicates Sites of Action of Curcumin**

Supplementation of 1% turmeric in the daily diet inhibited benzopyrene-induced stomach tumors and also spontaneous mammary tumors in mice.[28,29] Oral administration of turmeric water extract or curcumin inhibited benzopyrene-induced stomach tumors in mice.[29,82,23] In another study, mice fed OS%, 1% and 2% curcumin in daily diet at initiation and post initiation stages of benzopyrene-induced stomach cancer, showed reduction in rate of benign and malignant tumor development.[83] Rats

fed with 0.2% or curcumin in daily diet showed inhibition of azoxymethane-induced colon precancerous lesions and also reduced the development of colon carcinoma.[24,139]

Turmeric extract applied to mouse skin prevented dimethylbenzanthracene (DMBA) and methylcholanthrene induced skin tumors.[40] Skin painting with curcumin also prevented DMBA induced skin tumors in mice." This study confirmed that curcuminoids inhibit cancer at initiation, promotion and progression stages of development.

In clinical studies, curcumin 0.5% ointment applied topically on skin cancerous lesion in 62 patients, was found to reduce foul smell, itching, pain and exudate in majority of the patients.[86] Foul smell was considerably reduced in more than 90%, pain and itching in 50% and exudate was reduced in 70% of cases.



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Turmeric extract alone or in combination with betel leaf extract was effective against methyl acetoxy methyl nitrosamine induced oral tumors in hamster buccal pouch.[87] Curcumin also inhibited 4-nitroquinoline induced oral tumors in rats.[88] In a study done in India, anticarcinogenic effectiveness of curcumin was tested in patients with oral cancer. One hundred patients were given 500 mg of curcumin three times a day for 30 days. Some patients responded with a dramatic clinical improvement within 15 days, while others responded gradually, during the 30-day treatment.[32]

5. Turmeric extract and curcuminoids as antimutagens

The anticarcinogenic activity of turmeric extract and curcumin is related to their potential use as agents for preventing genetic mutation or mutagenesis. Mutation is a cellular genetic change, which is passed on to subsequent cell generations. The activity of turmeric and curcuminoids against mutagens like benzypyrene, dimethylbenzanthracene (DMBA), chili and capsaicin has been established using the standard Ames test[90] procedure. In animal studies, curcuminoids inhibited capsaicin-induced mutagenic changes in mouse bone marrow.[136] Additionally, mice maintained on turmeric or curcuminoid-enriched diets, when challenged with carcinogens, excreted low levels of mutagenic metabolites as well as carcinogens.[30,91]

It has been shown that turmeric and curcumin inhibited the mutagenicity of cigarette smoke condensate, *bidi* (a kind of cigarette used in India) smoke condensate, tobacco and *masheri* - a tobacco product used as dentifrice.[92,94,95] Aqueous extract of turmeric also inhibited the biological action of mutagens[83] In a comparative evaluation of efficacy of antimutagens toward cigarette smoke condensate and nitroquinoline- 1-oxide, curcumin was shown an effective inhibitor of both mutagens.[96]

Curcumin was evaluated as an antimutagen in a group of 16 chronic smokers in India." Curcumin was administered in a dose of 1.5 gm a day for 30 days. This regimen significantly reduced the urinary excretion of tobacco mutagens, and also enhanced the ability of drug metabolizing enzymes to detoxify carcinogens and mutagens found in cigarette smoke.

The nutritional quality of food is known to change with cooking. For example, upon cooking at high temperature, amino acids, undergo a physico-chemical change called pyrolysis. As a result, they may be converted into compounds with mutagenic, carcinogenic or diabetogenic properties. Turmeric extract and curcumin inhibited the formation of mutagenic pyrolysates.[30] These results validate the healthy practice of using turmeric as a food additive. It has also been found that turmeric extract and curcumin can inhibit formation and mutagenicity of aflatoxin, a very common toxin found in mold growing on poorly preserved foods.[97]

Among the carcinogenic and mutagenic substances produced form foods, nitrosamines are particularly important. Sodium nitrite is added as a preservative to enhance the color and texture of the meat. Under acidic conditions in the human stomach, amines and amides chemically react with nitrites, resulting in nitrosation reaction to form potent mutagenic and carcinogenic nitroso-compounds. This damaging reaction can be blocked by certain substances in food, including natural phenolics such as curcuminoids, vitamins C and E, and beta-carotenes.

Both turmeric extract and curcumin inhibited the nitrosation of methyl urea with sodium nitrite at acidic pH.[98] Turmeric extract and curcumin block the nitrosation

reaction of fish extract with sodium nitrite. The nitrosation inhibition by curcumin is mediated through its scavenging, or neutralizing, capability of nitrite ions from the media. The essential chemical structure in curcumin molecule to inhibit the nitrosation reaction has been shown to be the para hydroxy groups in the benzene rings.[98]

6. Antiviral, antimicrobial and antiparasitic activity or turmeric and curcumin

The development of mammary tumors in C3H (Jax) CRI strain of mice is a multifactorial event including four known components: mammary tumor virus (MMTV), hormonal factors, environmental factors and genetic predisposition to the disease. The C3H (Jax) CRI mice fed daily with diet containing 1% of turmeric had shown lower incidence of mammary tumors.[28] The inhibition of mammary tumors by turmeric could be a result of interference with some or all of the tumor contributing factors. It is believed that turmeric can prevent activation and growth of the virus causing the mouse mammary tumor. Interestingly, turmeric has been found to have hormone-like, (eg., estrogen), properties."' This may also be a factor in turmeric's preventive action in mammary carcinoma in mice.

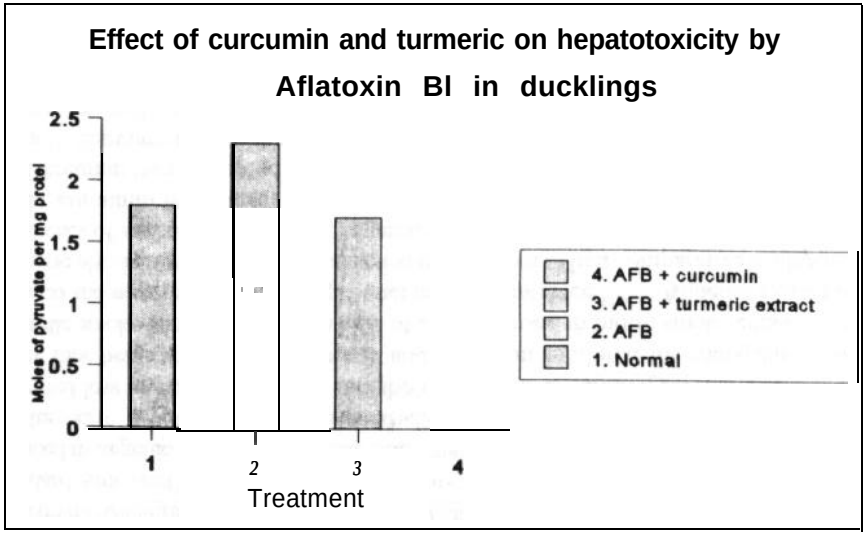
Tumeric was also studied as a potential antiviral agent against human immunodeficiency virus (HIV). The infection with HIV is characterized by a complex command system which results in virus activation or inactivation. The essential structural part of that command system in HIV is called long terminal repeat (LTR). Drugs that interfere with LTR may be of potential therapeutic value in delaying active HIV infection and progression of AIDS. Curcumin has been found to effectively inhibit activation of the LTR and to decrease HIV replication.[15]

In a controlled clinical study, a group of 18 HIV-seropositive patients with CD-4 cell counts ranging from 5 to 615 and CD-8 cell counts ranging from 283 to 1467, took an average of 2 gm of curcuminoids per day for an average of 127 days! This regimen resulted in the increase of CD-4 and CD-8 cell counts as compared to control treatment. No adverse effects of the treatment were noted. However, further studies are required to confirm the usefulness of these curcuminoids in AIDS management.

The antibacterial effects of alcoholic extract of turmeric, curcumin and oil from turmeric have been studied. [123,124,125] An alcoholic extract of turmeric at a dose of 50 mg/ml showed in vitro bactericidal activity. Turmeric oil was found to possess bacteriostatic action in vitro in dilutions of up to 1: 1000. The essential oil at a dose of 4.5 to 90 µl/lO0 ml was found effective in vitro against variety of microorganisms. Curcumin at concentration of 2.5 to 50 mg/100 ml inhibited in vitro growth of ***Staphylococcus aureus.***[123] Sodium curcuminate was found effective in vitro against ***Microcus pyogenes*** in a dilution of one part per million.[126]

Interestingly, the antibacterial and antiviral activities of curcumin were significantly enhanced by illumination with visible light.[128,129,130] The light-enhanced toxicity of the curcumin molecule to microorganisms has been found to be oxygen-dependent. It is mediated through curcumin generated superoxide, hydrogen peroxide and hydroxyl radicals.[131,132] These experimental data support the potential use of curcumin combined with light therapy (phototherapy) in the treatment of some bacterial and viral infections.

The crude ether and chloroform extracts of turmeric stem showed fungistatic activity against several dermatophytes in vitro.[133] The aqueous and alcoholic extracts of turmeric as well as curcumin inhibited production of aflatoxins by Aspergillus ***parasiticus in vitro."*** Aflatoxin is a metabolite of mold, which grows on food and contaminates various foods stored in unsanitary, humid conditions. Aflatoxins ingested with food, for example with moldy peanuts, may, over a long term, cause liver damage and cancer. The modifying effect of turmeric extract and curcumin on aflatoxin induced liver damage were studied in ducklings."



(Ref. **41)**

Aflatoxin-treated ducklings showed a decrease in body weight and white blood cell counts. They also had elevated levels of serum and liver glutamate pyruvate transaminase activity. Feeding turmeric extract and curcumin to ducklings which were exposed to aflatoxin, prevented the loss of body weight and decrease the damage to

white blood cells. The glutamate pyruvate transaminase activity in turmeric extract and curcumin fed animals was normalized to the control levels.

The ethanol extract of turmeric has been reported to have anti-amoebic activity against *Entamoeba histolytica,*[134] *in vitro.* Curcuminoids consisting of curcumin, demethoxycurcumin, bisdemethoxycurcumin and the newly isolated cyclocurcumin have been shown to be effective against nematode parasites *in vitro*.[60]

7. Bioprotective and chemopreventive activity of turmeric and curcumin

The broad biological activity of turmeric extract and its most researched compounds, curcuminoids, is basically related to protective mechanisms, which preserve the integrity and functioning of a biological system. Two important protective mechanisms of turmeric extract and curcuminoids have been studied: protection against environmental pollutants or bioprotection and protection against side effects produced by drug therapy or chemoprevention.

The state of health is a result of body's ability to recover from the continuous challenges posed by toxic substances entering through air, food and water. The most vivid example of this minute-by-minute struggle is our defense reaction against chemicals that cause cancer. Most cancer-causing compounds (carcinogens) undergo metabolic changes in the body and convert to 'activated' carcinogens. The 'activated' carcinogen binds to the cell DNA (genetic material), and damages it by forming the so-called 'DNA adducts.' Cells have mechanisms to repair the damaged DNA, but during this process errors may be introduced to the genetic code of repaired DNA. These errors result in permanent changes (mutations) to the process of building the essential elements of body cells, tissues and organs. These permanent changes may eventually give rise to cancer.

A potential preventive role of curcumin on DNA adduct formation with the carcinogen benzopyrene, has been studies *in vitro."* Rats kept on 0. 1 %, 0.5%, 3% turmeric supplemented diet and 0.03% curcumin supplemented diet for four weeks, were subsequently injected with benzypyrene. As compared to the control animals, rats fed with turmeric extract or curcumin showed decrease in the levels of DNA adduct in the liver cells.[34] This decrease could be explained by the competitive binding of curcumin to the active site of benzypyrene, preventing cellular DNA adduct formation.

The important protective role of curcumin in the biological systems may be due to its effect on polyamine biosynthesis. Increase in polyamine biosynthesis is associated with rapid cell growth, a characteristic of cancer tissue growth. Tumor promoters like tetradeconyl phorbol ester (TPA) are known to induce the activity of

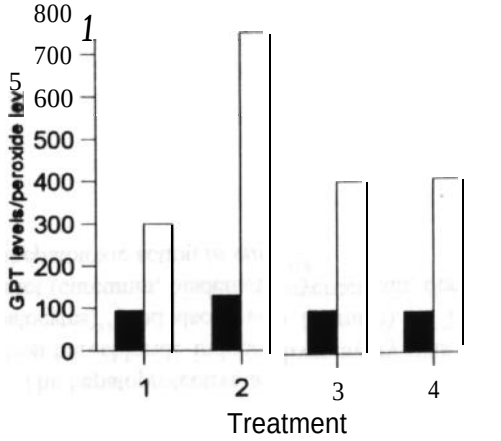
enzyme ornithine decarboxylase - one of the key enzymes which regulate polyamine biosynthesis and cell growth.

Addition of 0.5 to 10 micromoles of curcumin to an *in vitro* culture of mouse skin tissue and kerationcytes inhibited the TPA-induced activity of ornithine decarboxylase and DNA synthesis.[99] Several studies support the existence of a complex molecular mechanism for the bioprotective role of curcumin, involving a range of enzyme-inhibitory actions.[9]

Curcumin may also protect the integrity of genetic material, biological molecules and cells indirectly. This protective activity may be a result of altered levels of xenobiotic (drugs, environmental pollutants) metabolizing enzymes, e.g., gluthathione S-transferase and cytochrome P-450 are induced by curcumin.[36,37,38]

The potential of turmeric and curcumin in preventing tissue damage induced by xenobiotics has been studied. The effectiveness of any chemotherapeutic agent can be described by its chemotherapeutic index, that is the proportion of the effective dose to its toxic dose. Chemotherapeutic agents may be responsible for various degrees of collateral tissue damage, manifested as side effects. These side effects may often be more serious than the treated disease itself, disallowing long-term drug use.

Effect of turmeric extract and curcumin on liver toxicity induced by cyclophosphamide in mice



(Ref. 40)

Treatments as In the above chart:

p value for GPT

p value for Lipid peroxidation

1. Control

2. Cyclophosphamide	**p < 0.001**	p < 0.001
3. Cyclophosphamide + turmeric extract	**p < 0.001**	**p < 0.5**
4. Cyclophosphamide + curcumin	**p < 0.001**	p < 0.01

A serious health hazard associated with drug therapy can be illustrated by the current generation of anti-cancer drugs. Because of their cytotoxic nature, anti-cancer drugs do not discriminate between cancer cells and normal cells, and also could cause damage to non-cancerous tissue.

Turmeric extract and curcumin administered to mice along with the anti-cancer drug cyclophosphaminde, increased the life spans of animals and reduced bone marrow and liver toxicity of cyclophosphamide.[40] As an index of liver function, the liver alkaline phosphatase, glutamate pyruvate transaminase and Iipid peroxide levels were assayed. Animals receiving Cyclophosphamide alone has decreased levels of alkaline phosphatase, along with elevated levels of glutamate pyruvate transaminase and lipid peroxides. In contract, addition of turmeric extract or curcumin to the diet of cyclophosphamide-receiving animals normalized the liver enzymes to control levels.

Cyclophosphamide has been shown to undergo oxidation in the liver resulting in the production of damaging superoxides and free radicals. Curcuminoids, by virtue of their antioxidant activity, scavenge free radicals[132] as well as prevent their formation, thereby eliminating the toxic effects of the drug. Liver enzyme indices of toxicity therefore revert to normal levels on simultaneous administration of curcuminoids with the drug,

The effect of combining curcumin with cisplatin, a very potent anti-cancer drug, was evaluated.[102] Clinical use of cisplatin limited because of its severe toxicity leading to kidney failure. In an animal study, groups of 7 male ICR mice were treated with 2mg/kg cisplatin intraperitoneally for nine days and one group of mice received 50 mg/kg of curcumin, 30 minutes before receiving cisplatin. Analysis on the tenth day revealed statistically significant decrease in body weight, leucocyte count and increase in serum urea levels in the mice treated with cisplatin alone. On the other hand, mice treated with cisplatin and curcumin showed no such changes. Additionally, kidney lipid peroxidation was reduced by 59% in the cisplatin+curcumin-treated mice as compared with those treated with cisplatin alone.

Curcumin, as discussed previously, has defined antioxidant properties, and as such may exert protective effect against chemotherapeutics which either behave as free radicals or cause generation of tissue damaging free radicals. For example, cyclophosphamide undergoes oxidation in the body to form therapeutically active metabolites. During this oxidation process, various free radicals are generated as by-products of the oxidation reaction. Hydrogen peroxide is one of the free radicals

generated during a variety of oxidation reactions, including physiological processes. Unless neutralized, excess hydrogen peroxide oxidizes cellular membrane lipids and causes permanent damage to the cell resulting, for example, in hemolysis of red blood cells. Another oxidizing chemical generated during oxidation reactions and physiological processes is nitrite. Curcumin inhibited the hydrogen peroxide and nitrite-induced membrane lipid peroxidation and hemolysis of erythrocytes *in vitro*.[103,104]

The hepatoprotective action of an alcoholic extract of *Curcuma longa* against carbon tetrachloride-induced liver injury was tested in ***vitro*** (primary cultured rat hepatocytes)[118] and also *in* vivo (in mice).[140] The three major fractions from the extract (curcumin, bisdemethoxycrucumin, demethoxy-curcumin) exhibited similar antihcpatotoxic action *in vitro*.[118]