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## Garlic in health and disease

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## Abstract

The present article reviews the historical and popular uses of garlic, its antioxidant, haematological, antimicrobial, hepatoprotective and antineoplastic properties and its potential toxicity (from sulfoxide). Garlic has been suggested to affect several cardiovascular risk factors. It has also been shown that garlic and its organic allyl sulfur components are effective inhibitors of the cancer process. Since garlic and its constituents can suppress carcinogen formation, bioactivation and tumour proliferation, it is imperative that biomarkers be established to identify which individuals might benefit most. Garlic powder, aged garlic and garlic oil have demonstrated antiplatelet and anticoagulant effects by interfering with cyclo-oxygenase-mediated thromboxane synthesis. Garlic has also been found to have synergistic effects against *Helicobacter pylori* with a proton pump inhibitor. The active compound allicin may affect atherosclerosis not only by acting as an antioxidant, but also by other mechanisms, such as lipoprotein modification and inhibition of LDL uptake and degradation by macrophages. Freshly prepared garlic homogenate protects against isoniazid+rifampicin-induced liver injury in experimental animal models. Several mechanisms are likely to account for this protection.

## Keywords

Garlic      Antioxidants      Antimicrobials      Antineoplastics

Hepatoprotection

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## Introduction

Garlic (*Allium sativum*) is a popular spice, a remedy for a variety of ailments and is also known for its medicinal uses as an antibiotic, anti-thrombotic and antineoplastic agent. It has been used for thousands of years for culinary, medicinal and spiritual purposes.

## Biochemistry

Potentially active chemical constituents of garlic are:

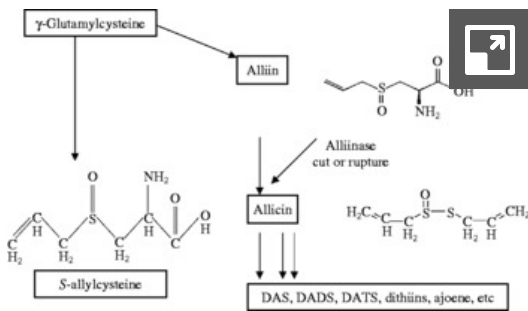
- (a)** Sulfur compounds: alliin, allicin, ajoene, allylpropyl disulfide, diallyl trisulfide (DATS), *S*-allylcysteine (SAC), vinylidithiins, *S*-allylmercaptocysteine and others.
- (b)** Enzymes: alliinase, peroxidases, myrosinase and others.
- (c)** Amino acids and their glycosides: arginine and others.
- (d)** Se, Ge, Te and other trace minerals.

Garlic contains at least thirty-three sulfur compounds, several enzymes and seventeen amino acids<sup>(1)</sup>. Additional constituents of intact garlic include steroidal glycosides and lectins<sup>(2, 3)</sup>. It contains higher concentrations of sulfur compounds than any other *Allium* species. The sulfur

compounds are responsible both for garlic's pungent odour and many of its medicinal effects.

### Chemical changes in garlic

When garlic is 'damaged', i.e. attacked by microbes, crushed, cut, chewed, dehydrated, pulverised or exposed to water, the vacuolar enzyme alliinase rapidly lyses the cytosolic cysteine sulfoxides (alliin). The transiently formed compound, allicin, comprises 70–80 % of the thiosulfinates. Typically, alliin is converted to allicin by alliinase (Fig. 1). Allicin instantly decomposes to other compounds, such as diallyl sulfide (DAS), diallyl disulfide (DADS), dithiins and ajoene. At the same time,  $\gamma$ -glutamyl cysteine is converted to SAC, via a pathway other than the alliin–allicin pathway.



**Fig. 1** Chemical changes in garlic. DAS, diallyl sulfide; DADS, diallyl disulfide; DATS, diallyl trisulfide.

The dozens of brands of garlic on store shelves can be classified into four groups, i.e. garlic essential oil, garlic oil macerate, garlic powder and aged garlic extract (Table 1)<sup>(4)</sup>.

**Table 1** Main brands of garlic on the market

Type of product	Main compounds and characteristics
Garlic essential oil	Only 1 % oil-soluble sulfur compounds (for example, DAS or DADS) in 99 % vegetable oil. No water-soluble compounds
Garlic oil macerate	Oil-soluble sulfur compounds and alliin, no allicin
Garlic powder	Alliin and a small amount of oil-soluble sulfur compounds
Aged garlic extract	Mainly water-soluble compounds (for example, SAC or saponins), small amount of oil-soluble sulfur compounds

DAS, diallyl sulfide; DADS, diallyl disulfide; SAC, S-allylcysteine.

Garlic oil, aged garlic and steam-distilled garlic do not contain significant amounts of alliin or allicin, but contain various products of allicin transformation, among which none appears to have as much physiological activity as fresh garlic or garlic powder<sup>(5)</sup>. In rats, alliin is well absorbed orally, reaching maximum serum concentrations within 10 min, and is excreted within about 6 h. Allicin and vinylthiols are absorbed more slowly, reaching peak levels between 30 and 120 min and persisting in the body for up to 4 d<sup>(6)</sup>. In rats, mice and dogs, SAC is well absorbed (98–100 %) orally<sup>(7)</sup>. Excretion occurs through the renal system and through hepatic breakdown, faecal excretion and exhalation.

## **Anti-carcinogenic actions of garlic**

Epidemiological and animal studies have shown that consumption of garlic and its allyl sulfur compounds reduces the incidence of cancer, for example, stomach, oesophagus, colon, breast, cervix, skin, uterine and lung cancers<sup>(8)</sup>.

The diverse array of compounds and target tissues involved suggests either that garlic or its associated constituents have multiple mechanisms of action or, more logically, influence a fundamental step in the overall cancer process. Metabolic activation is a necessary event for many of these carcinogens used in animal studies, and possibly for environmental exposures faced by humans. Thus, phase I and II enzymes involved in carcinogen bioactivation and removal may be key in explaining the response to garlic and allyl sulfur compounds. However, few studies have noted significant changes in cytochrome P450 1A1, 1A2, 2B1 or 3A4 activities after supplementation with garlic or related sulfur

compounds<sup>(9, 10)</sup>. Therefore, other enzymes involved in the bioactivation or removal of carcinogenic metabolites may play a role.

Singh *et al.* provided evidence that the efficacy of various organosulfides to suppress benzo(a)pyrene tumorigenesis correlated with their ability to suppress NAD(P)H:quinone oxidoreductase, an enzyme involved with the removal of quinones associated with this carcinogen<sup>(11)</sup>. Depressed carcinogen bioactivation because of reduction in cyclooxygenase and lipoxygenase activity may also account for some of the lower incidence of tumours after treatment with some carcinogens<sup>(12, 13)</sup>. Enhanced glutathione availability and an elevation in the activity of specific glutathione S-transferase (GST), both factors involved in phase II detoxification, may also be significant in the protection provided by garlic and associated allyl sulfur components. Ingestion of garlic by rats increases the activity of GST in both liver and mammary tissue<sup>(9, 14)</sup>. It should be noted that not all GST isozymes are influenced equally. Hu *et al.* provided evidence that the induction of GST  $\pi$  may be particularly important in the anticarcinogenic properties associated with garlic and allyl sulfur components<sup>(15)</sup>.

## Medical uses of garlic

More than 3000 publications in the past have confirmed the efficacy of garlic for the prevention and treatment of a variety of diseases, acknowledging and validating its traditional uses. It may influence the risk of heart disease and is also used for the treatment of fatigue, although the mechanism involved remains unclear. The anti-fatigue function of garlic may be

closely related to its many favourable biological and pharmacological effects<sup>(16)</sup>. Garlic has historically been used to treat aches and pains, leprosy, deafness, diarrhoea, constipation, parasitic infection and fever and to relieve stomachache<sup>(17)</sup>. It has also been used to lower blood pressure, food poisoning, tumours and as a mild anticoagulant<sup>(18)</sup>. Arabian herbalists use garlic to treat abdominal pain, infantile colic, diarrhoea, diabetes, eye infections, snake bites, dandruff and tuberculosis<sup>(19)</sup>. In Ayurvedic medicine, garlic is used to treat respiratory problems, colic and flatulence. Garlic oil drops are used to treat earache<sup>(20)</sup>. Louis Pasteur demonstrated garlic's antiseptic activity in 1858. In African countries, it is also used to treat dysentery<sup>(21)</sup>. Garlic is commonly used to treat chronic bronchitis, recurrent upper respiratory tract infections and influenza<sup>(22)</sup>. In India and Europe, garlic remedies are used to treat coughs, colds, hay fever and asthma<sup>(23)</sup>.

## Antioxidant actions of garlic

Protection with aged garlic extract and *Nigella sativa* oil has been shown to prevent most of the haematological and biochemical changes and to markedly improve the antioxidant capacity of schistosomiasis-infected mice compared with infected-untreated ones. The antioxidant and antischistosomal action of aged garlic extract and *N. sativa* oil was greatly diverse according to treatment regimen<sup>(24)</sup>.

Garlic oil could be an effective agent or food supplement in reducing the toxicity of tributyltin<sup>(25)</sup>. Allicin-mediated lipoperoxide production in fungal plasma membranes is the

cause of the enhancement in the cellular uptake of polymyxin B as well as its action against the vacuole<sup>(26)</sup>.

Supplementation of garlic oil at 5 mg/kg body weight has been shown to have an anticoagulation effect in an animal study<sup>(27)</sup>.

Whole garlic and aqueous garlic extract exhibit direct antioxidant effects and enhance the serum levels of two antioxidant enzymes: catalase and glutathione peroxidase<sup>(28)</sup>. Garlic extract and allicin efficiently scavenged exogenously generated hydroxyl radicals in a dose-dependent fashion<sup>(29)</sup>. Other garlic constituents, such as SAC, also demonstrated significant antioxidant effects *in vitro*<sup>(30)</sup>. The sulfur compounds found in fresh garlic appear to be nearly 1000 times more potent as antioxidants than those found in aged garlic extract<sup>(31)</sup>.

Garlic (both the 10 % homogenate in physiological saline solution and its supernatant fraction) was able to reduce the radicals that were generated by the Fenton reaction and trapped by phenyl-butyl-nitron; radicals present in cigarette smoke were also reduced by garlic<sup>(32)</sup>. In rat liver microsomes, garlic extract prevented the formation of thiobarbituric acid-reactive substances in cell membranes during lipid peroxidation in a dose dependent fashion<sup>(33)</sup>. An aqueous extract obtained from 1 mg of a garlic preparation was as effective as an antioxidant as 30 nmol ascorbic acid and/or 3.6 nmol  $\alpha$ -tocopherol<sup>(34)</sup>. Glycation endproduct formation is increased under conditions of oxidative stress, such as glutathione depletion, and glycated proteins produce 50-fold more toxic free radicals than non-glycated proteins. A



recent study suggested that aged garlic extract inhibits the formation of glycation endproducts *in vitro* and the formation of glycation-derived free radicals<sup>(35)</sup>.

Garlic and its constituents protect tissue against oxidative damage and improve organ functions in various animal models<sup>(36)</sup>. In male Wistar rats, administration of garlic extract against nicotine hydrogen bitartrate for 21 d increased glutathione levels and decreased malondialdehyde levels in aorta, heart, kidney and urinary bladder tissues<sup>(37)</sup>. Aqueous garlic administration protects the tissues against nicotine-induced oxidative damage and improves renal function and histological damage. Similarly, oral administration of garlic oil in carbon tetrachloride-intoxicated rats significantly decreases the tissue lipid profile, lipid peroxidation levels, alkaline phosphatase, serum transaminases and protects the liver from damage by carbon tetrachloride as effectively as vitamin E. These changes induced by garlic oil in these rats are comparable with that of vitamin E<sup>(38)</sup>. DAS, DADS and DATS are principal constituents of garlic. The effect of these sulfides on phase II drug-metabolising enzymes and on the rat model of acute liver injury induced by carbon tetrachloride has been studied. A highly purified form of each sulfide was administered to rats at a concentration of 10 or 100 µmol/kg body weight for 14 consecutive days. DATS (10 µmol/kg) and DADS (100 µmol/kg) significantly increased the activities of GST and quinone reductase, whereas DAS did not. In the rat model of carbon tetrachloride-induced acute liver injury, DATS (10 µmol/kg) significantly suppressed the increase in plasma lactate dehydrogenase and serum glutamate oxaloacetate

transaminase activities. Therefore, hepatic phase II enzymes were induced strongly by the trisulfide and weakly by the disulfide, but not by DAS. DATS significantly reduced the liver injury caused by carbon tetrachloride. DATS may be one of the important factors in garlic oil that protects our body against the injury caused by radical molecules<sup>(39)</sup>.

Another study showed the modifying effects of aqueous extracts of garlic and neem leaf during the pre-initiation and post-initiation phases of gastric carcinogenesis induced by *N*-methyl-*N'*-nitro-*N*-nitrosoguanidine in rats. Enhanced lipid peroxidation in the liver and blood of tumour-bearing animals was accompanied by significant decreases in the activities of glutathione-dependent antioxidants in the pre-initiation as well as in the post-initiation phases, but these alterations were reduced by combined supplementation of garlic and neem. It was suggested that the modulatory effects of garlic and neem leaf on hepatic and blood oxidant-antioxidant status may play a key role in preventing cancer development at extra-hepatic sites<sup>(40)</sup>. SAC, a water-soluble compound of garlic, also has been shown to have antioxidant activity in experimental animals. Much experimental evidence suggests that the actions of quinolinic acid also involve reactive oxygen species (ROS) formation and oxidative stress as major features of its pattern of toxicity. The effect of a garlic-derived compound and well-characterised free radical scavenger, SAC, on quinolinic acid-induced striatal neurotoxicity and oxidative damage has been studied in rats. SAC at a dose of 150 mg/kg resulted effective to prevent quinolinate-induced lipid peroxidation, whereas the systemic administration of 300 mg/kg of this compound to rats

decreased effectively the quinolinic acid-induced oxidative injury measured as striatal ROS formation and lipid peroxidation. SAC (300 mg/kg) also prevented the striatal decrease of Cu/Zn-superoxide dismutase (SOD) activity produced by quinolinate. In addition, SAC, at the same dose tested, was able to reduce the quinolinic acid-induced neurotoxicity evaluated as circling behaviour and striatal morphological alterations. SAC ameliorates the *in vivo* quinolinate striatal toxicity by a mechanism related to its ability to: (a) scavenge free radicals, (b) decrease oxidative stress and (c) preserve the striatal activity of Cu/Zn-SOD. This antioxidant effect seems to be responsible for the preservation of the morphological and functional integrity of the striatum<sup>(41)</sup>. Aged garlic extract, through its scavenging property on oxidants, increases SOD, catalase, glutathione peroxidase and glutathione levels, and inhibits lipid peroxidation and inflammatory prostaglandins in CVD and dementia<sup>(42)</sup>.

Garlic increases cellular glutathione in a variety of cells including those in normal liver and mammary tissues<sup>(43)</sup>. The ability of garlic to increase glutathione peroxidase and other ROS-scavenging enzymes is important in radioprotection and UV suppression of certain forms of immunity<sup>(44)</sup>.

Oxidative stress and increased systemic inflammation may contribute to endothelial dysfunction. Aged garlic extract containing antioxidant compounds increases NO production and decreases the output of inflammatory cytokines from cultured cells. A recent study showed the effect of treatment with aged garlic extract on brachial artery flow-mediated

endothelium-dependent dilation and circulating markers of oxidative stress and systemic inflammation in fifteen men with 2 weeks of treatment. During aged garlic extract supplementation, endothelium-dependent dilation increased (44 %) significantly from baseline and mainly in men with lower baseline endothelium-dependent dilation. Levels of endothelium-dependent dilation at the end of aged garlic extract treatment were significantly higher compared with the corresponding levels at the end of placebo treatment. Although markers of oxidative stress (plasma oxidised LDL and peroxides), systemic inflammation (plasma C-reactive protein and IL-6) did not change significantly during the study, short-term treatment with aged garlic extract may improve impaired endothelial function in men with coronary artery disease treated with aspirin and statin<sup>(45)</sup>.

Short-term garlic supplementation may have a protective role in hypertensive patients and on indices of oxidative stress. Garlic pearls at a dose of 250 mg/d for 2 months in hypertensive patients were found to decrease lipids and lipoprotein subfractions, plasma-oxidised LDL, plasma and urinary concentrations of 8-iso-PG F2 $\alpha$  as a biomarker of oxidative stress and increase total antioxidant status. With 2 months of garlic pearl supplementation, there was also a significant decline in both systolic and diastolic blood pressures. It could be suggested that dietary supplementation of garlic may be beneficial in reducing blood pressure and oxidative stress in hypertensive individuals<sup>(46)</sup>.

Epidemiological as well as laboratory studies have shown that garlic consumption reduces the incidence of cancer in the stomach, colon, mammary glands, cervix, etc. Some of the hypothesised mechanisms of the anticarcinogenic effects of garlic are: (i) scavenging of free radicals, (ii) increasing glutathione levels, (iii) increasing the activities of GST and catalase, (iv) inhibition of cytochrome p450 2E1, (v) DNA repair and prevention of chromosomal damage<sup>(47)</sup>. Durak *et al.*<sup>(48)</sup> showed that the ingestion of garlic extract (1 ml garlic extract/kg body weight daily for 6 months) leads to significantly lowered plasma and erythrocyte malondialdehyde levels in atherosclerotic patients, even in the absence of changes in antioxidant enzyme activities. They also demonstrated the presence of oxidant stress in blood samples from patients with atherosclerosis which was eliminated by ingesting garlic extract. Thus, it is possible that reduced peroxidation processes may play a part in some of the beneficial effects of garlic in atherosclerotic diseases. In addition, the relationship between the effects of garlic extract supplementation on the blood lipid profile and oxidant-antioxidant status in hypertensive patients suggests a protective effect.

## **Antilipaemic actions of garlic**

Garlic has demonstrated effects on several risk factors for CVD-hyperlipidaemia, hypertension and platelet aggregation<sup>(49)</sup>. Its anti-atherosclerotic properties are mainly attributed to allicin, which is produced upon crushing of the garlic clove. By using a pure allicin preparation, Gonen *et al.* were able to show that allicin may affect atherosclerosis not only by acting as an antioxidant, but also by other mechanisms, such as

lipoprotein modification and inhibition of LDL uptake and degradation by macrophages<sup>(50)</sup>.

In rat hepatocytes, garlic paste, garlic oil, allicin and ajoene were found to significantly reduce cholesterol biosynthesis by inhibiting 3-hydroxy-3-methylglutaryl (HMG)-CoA reductase and 14- $\alpha$ -demethylase<sup>(51)</sup>. Some authors postulate that garlic's trace minerals, such as Te, also inhibit hepatic cholesterol synthesis<sup>(52)</sup>. But most attribute garlic's antilipaemic effects to DADS, a decomposition product of allicin<sup>(53)</sup>.

Garlic lowers hyperlipidaemia in animal studies<sup>(38)</sup>. In rats, both garlic and garlic oil have been shown to exhibit significant lipid-lowering effects, primarily through a decrease in hepatic cholesterologenesis<sup>(54)</sup>. Rats that were on an atherogenic diet were supplemented with freeze-dried garlic powder; garlic exerted a dose-effect response, with the highest doses lowering serum cholesterol significantly, enhancing the ratio of HDL-cholesterol:LDL-cholesterol<sup>(55)</sup>. Chickens whose diets were supplemented with garlic powder had significant reductions of plasma and tissue cholesterol and plasma TAG<sup>(56)</sup>. Garlic supplementation also significantly decreased HMG-CoA reductase activity and cholesterol 7 $\alpha$ -hydroxylase activity<sup>(57)</sup>. In an experimental model, where rabbits were fed with a high-cholesterol diet supplemented with garlic or allicin, hypercholesterolaemia was significantly inhibited by 50 %, indicated by decreased tissue cholesterol, LDL concentrations, raised HDL concentrations, and reduced atheromatous changes<sup>(58)</sup>. Garlic also provided significant protection for the enzymes of the glutathione-dependent

peroxide detoxification system, which is strongly impaired under hypercholesterolaemia<sup>(59)</sup>. In another study, rats were given garlic, ginger or garlic and ginger supplements for 4 weeks. All groups exhibited significant decreases in blood glucose, serum total cholesterol and serum alkaline phosphatase. Cholesterol was significantly decreased in animals fed with a combination of the two, compared with either alone, indicating that a combination of garlic and ginger is much more effective in reducing blood glucose and serum lipids<sup>(60)</sup>. Since 1975, many human studies have evaluated garlic's lipid-lowering effects. Controlled trials in healthy adults given garlic supplementation along with cholesterol-rich diets suggest that garlic can reduce mean serum cholesterol levels and increase fibrinolytic activity<sup>(48)</sup>. Although the quality of randomised trials has been only modest, recent meta-analyses have concluded that in over a dozen trials published between 1979 and 1993, there was an average improvement in serum cholesterol concentrations of 9–12 % and a significant reduction in serum TAG in hyperlipidaemic patients taking standardised garlic powder supplements of 600–900 mg daily, with improvement evident within 1 month<sup>(61)</sup>. In a single-blind, placebo-controlled cross-over study, forty hypercholesterolaemic adults were assigned to either placebo for 1 month or fish oil with garlic powder (1200 mg) capsules daily for 1 month. Supplementation with garlic resulted in an 11 % decrease in cholesterol, a 34 % decrease in TAG, and a 10 % decrease in LDL levels, as well as a 19 % decrease in cholesterol/HDL risk<sup>(62)</sup>.

In cell cultures, aqueous solutions of dried garlic powder containing allicin and ajoene have been found to significantly

inhibit the proliferative activity of smooth muscle cells from atherosclerotic aortic plaques<sup>(63)</sup>. In hypercholesterolaemic rabbits, garlic supplements significantly reduced the aortic lesions and lipid content of existing fatty plaques<sup>(64)</sup>. In a randomised placebo-controlled trial in ten healthy adults, there was a significant improvement in plasma viscosity and capillary blood flow within 5 h of taking 900 mg of standardised garlic powder<sup>(65)</sup>. In a prospective 4-year clinical trial of patients treated with 900 mg daily of standardised garlic powder, there was a 9–18 % reduction in plaque volume, a 4 % decrease in LDL levels, an 8 % increase in HDL concentrations and a 7 % decrease in blood pressure<sup>(66)</sup>.

## Hepatoprotective actions of garlic

Garlic oil constituents, particularly DAS, activate constitutive androstane receptor (CAR) and nuclear factor E2-related factor 2 (Nrf2) to induce drug-metabolising enzymes<sup>(67)</sup>.

SAC decreases CCl<sub>4</sub>-induced liver injury by attenuation of oxidative stress, and can be a useful regimen for chronic liver disease<sup>(68)</sup>. Freshly prepared garlic homogenate has been shown to protect against isoniazid+rifampicin-induced liver injury in an experimental animal model<sup>(69)</sup>. In rat liver cultures, garlic administration before exposure to hepatotoxins (such as carbon tetrachloride, galactosamine and doxorubicin) provided protection against histological and biochemical evidence of damage<sup>(70)</sup>.

In mice, rats and chickens, pretreatment with oral garlic supplements provided significant protection against the toxicity of known hepatotoxins, including heavy metals<sup>(71, 72)</sup>.



In toads and rats, pretreatment with garlic protected against aflatoxin- and chemically induced liver tumours<sup>(73)</sup>. Aged garlic and garlic's diallyl sulfur compounds protected against acute chemically induced hepatotoxicity in rats<sup>(74)</sup>.

In rats, daily pretreatment with garlic oil against buthionine sulfoximine and carbon tetrachloride effectively prevented increases in plasma aminotransferases and lactate dehydrogenase activities, liver injuries as evidenced by elevations in central necrosis, hepatocyte degeneration and inflammation and increases in plasma TAG contents<sup>(36)</sup>. In the carbon tetrachloride-induced acute liver injury model of rats, DADS and DATS given at a higher dose (100 µmol/kg), by increasing the activities of GST, quinone reductase, and the antioxidative enzyme glutathione peroxidase, reduced the injury caused by the induction of phase II enzymes with carbon tetrachloride<sup>(39)</sup>. Garlic reduced the toxic effects exerted by carbon tetrachloride in other organs in rats, through inhibition of the cytochrome P450 system that activates carbon tetrachloride into its active metabolite, the trichloromethyl radical<sup>(75)</sup>. In ischaemia and reperfusion injury, garlic has been shown to have a protective effect. Aqueous garlic extract (1 ml/kg) was given to Wistar albino rats during an ischaemia and reperfusion period. Liver transaminases, malondialdehyde levels, glutathione levels, myeloperoxidase activity and hepatic collagen content, as a fibrosis marker, were increased in the ischaemia and reperfusion group. But these changes were reduced back to control levels by the aqueous garlic extract treatment. Therefore, aqueous garlic extract administration alleviated the ischaemia-reperfusion-induced injury of the liver and

improved hepatic structure and function. It seems likely that garlic extract, with its antioxidant and oxidant-scavenging properties, may be of potential therapeutic value in protecting the liver against oxidative injury due to ischaemia-reperfusion<sup>(37)</sup>.

In another study, induced liver fibrosis in male Wistar albino rats by bile duct ligation was reduced by aqueous garlic extract. It seems that aqueous garlic extract with its antioxidant and antifibrotic properties protects the liver from oxidative injury resulting in fibrosis<sup>(76)</sup>. In one case report, a patient with severe hepatopulmonary syndrome, who failed somatostatin therapy and declined liver transplantation, began taking large daily doses of powdered garlic. She experienced partial palliation of her symptoms and some objective signs of improvement over 18 months of continuous self-medication<sup>(77)</sup>.

## **Other potential effects of garlic**

### **Haematological effects**

In a study on horses, the ability of a herbal composite containing garlic, white horehound, boneset, aniseed, fennel, licorice, thyme and hyssop to reduce the clinical signs of recurrent airway obstruction was investigated; it was hypothesised that the product would safely reduce signs and would improve the inflammatory cell profile within the lungs<sup>(78)</sup>. It has been reported by Wojcikowski *et al.*<sup>(79)</sup> that garlic oil should not be relied on in individuals in which reduction in platelet aggregation is desired.

Fresh garlic, garlic powder, aged garlic and garlic oil have demonstrated antiplatelet and anticoagulant effects by interfering with cyclo-oxygenase-mediated thromboxane synthesis<sup>(33)</sup>. Garlic compounds contributing to the anti-thrombotic effect were: alliin, ajoene, allicin, vinyldithiols and DADS<sup>(80)</sup>. These effects appear to be important contributors to garlic's beneficial role in atherosclerotic conditions<sup>(81)</sup>.

## Antimicrobial effects

Garlic oil and allyl alcohol, both derived from alliin in garlic using different preparation methods, cause potent growth inhibition in yeasts<sup>(82)</sup>. The Gram-negative diarrhoeagenic pathogens from stool samples were highly sensitive to garlic, while ciprofloxacin was most effective against *Escherichia coli*<sup>(83)</sup>. The antimicrobial activity was compromised by storage and heating; therefore it is advisable to use fresh garlic and avoid boiling it for more than 5 min during cooking<sup>(84)</sup>.

Chopped garlic has a slowing-down effect on microbiological growth in ground meat depending on the garlic concentration, but this effect was not at an expected level even at the highest concentration, because potential antimicrobial agents in chopped garlic were probably insufficiently extracted<sup>(85)</sup>. Garlic cloves have antimicrobial properties *in vitro* against streptococci and anticarcinogenic properties against oral micro-organisms in spite of their adverse effects<sup>(86)</sup>. Fresh garlic and garlic powder, through their combined antioxidant and antimicrobial effects, are potentially useful in preserving meat products<sup>(87)</sup>. A wide variety of these organosulfur compounds, whether naturally occurring or synthetic, exhibit antibacterial properties. Mechanistically, organosulfur groups in garlic compounds

can act as metal chelators, powerful nucleophiles or electrophiles depending on the local environment in which a given reaction occurs<sup>(88)</sup>.

Garlic is nicknamed 'Russian penicillin' for its widespread use as a topical and systemic antimicrobial agent<sup>(89)</sup>. Crude garlic extracts exhibited activity against both Gram-negative and Gram-positive bacteria at room temperature, but there were no significant effects if the garlic had been boiled for 5 min before testing<sup>(90)</sup>. Aged garlic extract demonstrated dose-dependent antimicrobial activity against three different reference strains of *Helicobacter pylori* at concentrations of 2–5 mg/ml; however, heat treatment of the extracts reduced the anti-bacterial effects<sup>(91)</sup>. Garlic was also found to have synergistic effect against *H. pylori* with a proton pump inhibitor<sup>(92)</sup>. Garlic inhibited the growth of twenty different strains of intracellular *Mycobacterium avium* isolated from AIDS patients and non-AIDS patients<sup>(93)</sup>. In rabbits, aqueous garlic extract and allicin had significant antibacterial activity against *Shigella flexneri*, fully curing the infected rabbits within 3 d compared with a mortality rate of 80 % within 48 h in the untreated rabbits; antibacterial activity against the challenge strain was observed in the sera of the treated rabbits within 30–60 min of administration of the agent<sup>(94)</sup>.

## Antineoplastic effects

S-allylmercaptocysteine (SAMC) may promote docetaxel-induced cell death through promoting G2/M cell cycle arrest and apoptosis. The study by Howard *et al.* implies a potential role for SAMC in improving docetaxel-based chemotherapy for the treatment of hormone-refractory prostate cancer<sup>(95)</sup>.

With the dynamic expression of cyclin B1, DADS induces reversible cell cycle arrest in the G2/M phase of HCT-116 cells through a p53-independent mechanism<sup>(96)</sup>.

Garlic has been known to have anti-inflammatory, anti-cancer and antioxidant effects. Owing to these effects, garlic and its preparations have been used for the treatment of prostate cancer and relief of benign prostatic hyperplasia symptoms for decades<sup>(97)</sup>. DAS can effectively check the mutations induced by environmental toxicants, as reported by Nigam & Shukla<sup>(98)</sup>. The protective effects of high doses of garlic oil were more pronounced as compared with those in a vitamin E-administered group<sup>(99)</sup>.

The results demonstrated that DADS induced the expression of signal transducer and activator of transcription 1 (STAT1), which was also confirmed using Western blotting. STAT1 decoy oligonucleotides were also used to block STAT1 mRNA and led to a decrease in the levels of STAT1 and to a subsequent decrease in the percentage of apoptosis induced by DADS in examined colo 205 cells<sup>(100)</sup>.

Polybutylcyanoacrylate nanoparticles of DATS (DATS-PBCA-NP) were found to have a good prolonged release effect *in vivo* and hepatic-targeted activity, and a significant anti-tumour effect on the orthotopic transplantation hepatocellular carcinoma model in mice in association with the suppression of proliferation and the induction of apoptosis of tumour cells. These advantages are probably due to their liver-targeting characteristics and consequently bring a higher anti-tumour activity<sup>(101)</sup>. Garlic compounds induced apoptosis in glioblastoma cells due to the production

of ROS, increase in endoplasmic reticulum stress, decrease in  $\Delta\Psi_m$ , and activation of stress kinases and cysteine proteases<sup>(102)</sup>.

The promising chemopreventive nature of liposomal DAS may form the basis for establishing effective means of controlling various forms of cancer, including skin papilloma<sup>(103)</sup>. DAS is capable of acting efficiently and selectively only in the liver and can be used for hepatoprotection during chemotherapy<sup>(104)</sup>. DADS is an effective antifungal agent able to trigger cell death in *Candida*, most probably by eliciting oxidative stress as a consequence of thiol depletion and impaired mitochondrial function<sup>(105)</sup>. The ubiquitous efficiency of Z-ajoene, a new compound, has been demonstrated to fight against cancers of various origins including those that harbour viruses<sup>(106)</sup>. The role of oxidative stress and its relation to tumour promotion suggest protective effects of garlic oil against ferric nitrilotriacetate-induced hepatic toxicity and that it can serve as a potent chemopreventive agent to suppress oxidant-induced tissue injury and carcinogenesis<sup>(107)</sup>.

Garlic and its extracts appear to protect against DNA damage *in vitro*. In animal and epidemiological studies, garlic is associated with a reduced risk of cancer<sup>(108)</sup>. At least two anti-carcinogenic agents have been identified in garlic: DAS and GST<sup>(109)</sup>. In the glandular stomach mucosa of rats, pretreatment with DAS significantly and dose dependently inhibited chemically induced nuclear aberrations and ornithine decarboxylase activity<sup>(110)</sup>. Similarly, oral administration of a garlic supplement inhibited the

development of benzopyrene-induced neoplasia and induced increased GST activity in the forestomach<sup>(111)</sup>. In a study, supplementation with high-Se garlic before exposure to a known carcinogen significantly reduced DNA damage, normalised hepatic enzymes, and increased the concentrations of GST and uridine 5'-diphosphate-glucuronyltransferase in the liver and kidney, thus suggesting that garlic may suppress tumour development by enhancing detoxification<sup>(112)</sup>.

Effects of garlic have been summarised in Fig. 2.



**Fig. 2** Summary of the effects of garlic. GPX, glutathione peroxidase; HMG, 3-hydroxy-3-methylglutaryl; Nrf2, nuclear

factor E2-related factor 2.

## Potential toxicity of garlic

At present, there are no known toxic compounds in garlic and its extracts. Chronic administration of garlic powder (50 mg/d) resulted in inhibition of spermatogenesis in rats. Reduced concentration of sialic acid in the testes, epididymis and seminal vesicles together with decreased leydig cell function reflect anti-androgenic effects of garlic<sup>(113)</sup>. A higher concentration of garlic powder (200 mg/ml) or allicin isolated from garlic caused considerable cell injuries in the portal hepatic zone in isolated perfused rat liver<sup>(114)</sup>, which was not observed at a lower concentration.

Another *in vitro* study showed that DAS at 5 mM significantly decreased cell viability in the liver<sup>(115)</sup>. This evidence suggests

the cautious use of garlic in animals to monitor blood levels simultaneously. In another study in dogs and rats, raw garlic caused severe mucosal damage<sup>(6)</sup>. In another study, toxic effects of oral and intraperitoneal administration of garlic extracts on lung and liver tissue of rats were studied. Administration of low doses of garlic (50 mg/kg) to rats either orally or intraperitoneally had no effect on lung and liver tissue as compared with control animals. Administration of higher dose of garlic (500 mg/kg) resulted in profound changes in lung and liver tissue of rats, indicating dose-related toxicity<sup>(116)</sup>. The toxic effect of a high dose of garlic was further confirmed in another experimental model where 300 and 600 mg of garlic bulb extract was administered to both male and female rats for 21 d, which resulted in growth retardation and effects on biological parameters and histological structures<sup>(117)</sup>.

Banerjee *et al.*<sup>(118)</sup> investigated long-term effects of a low-dose garlic extract in an animal model using endogenous antioxidant enzymes and lipid peroxidation in the liver and kidney. Rats were fed with fresh garlic homogenate daily in three different doses (250, 500 and 1000 mg/kg per d) for 30 d. In comparison with saline-treated rats, the 250 mg/kg per d dose resulted in significantly reduced lipid peroxidation and glutathione peroxidase, no change in catalase and reduced glutathione; SOD was increased significantly. Both the 500 and 1000 mg/kg per d doses significantly reduced endogenous antioxidants (catalase and SOD) without altering lipid peroxidation level. Animals treated with 1000 mg/kg per d showed morphological changes in the liver both at light microscopy and ultrastructural levels. Histologically, liver



showed focal non-specific injury to the hepatocytes. The reason for such type of injuries could be the sulfoxides present in the garlic extract that can undergo exchange reactions with –SH groups of enzymes and proteins in the body, spontaneously at physiological pH and temperature, inhibiting their activity<sup>(119)</sup>. Therefore, it seems that garlic in low doses has the potential to enhance the endogenous antioxidant status, although at higher doses a reversal of these effects is observed.

Intraperitoneal and oral administration of high doses (5 ml garlic juice/kg) led to weight loss, and hepatic and pulmonary toxicity in rats<sup>(120)</sup>. However, in one study, hypertensive rats given garlic supplements four times daily developed erratic pulses, dehydration, weight loss and lethargy<sup>(121)</sup>.

In humans garlic is considered as safe by the US Food and Drug Administration but is also known to cause gastric irritation if taken in high doses by sensitive individuals. In randomised controlled trials, side effects included heartburn, nausea, vomiting, diarrhoea, flatulence, bloating, mild orthostatic hypotension, flushing, tachycardia, headache, insomnia, sweating and dizziness as well as offensive body odour<sup>(122)</sup>.

## Doses of garlic and its compounds

Yadav *et al.*<sup>(123)</sup> have shown that garlic in low doses is safe in frogs. They suggested that garlic extract has some beneficial effects on heart rate, rhythm and force of contraction, but very high doses may exert non-desirable effects as well. The comparative toxic effects of oral and intraperitoneal

administration of garlic extracts on lung and liver tissue of rats have also been studied. Administration of a low dose of garlic (50 mg/kg per d) to rats either orally or intraperitoneally had little effect on lung and liver tissues as compared with control animals<sup>(116)</sup>. In contrast, administration of a high dose of garlic (500 mg/kg per d) resulted in profound changes in lung and liver tissues of rats. Moreover, intraperitoneal administration of a high dose of garlic was more damaging to lung and liver tissues of rats than oral administration. Therefore, these reports highlight some of the adverse and toxic effects of high doses of garlic in animals. In support to this, it has been shown that higher concentrations of garlic extract are clastogenic in mice<sup>(124)</sup>, which was appreciably reduced at lower concentrations. Female and male rats were given 300 and 600 mg/kg per d of a garlic aqueous extract for 21 d and the results showed that garlic extract causes toxic effects affecting weight growth, biological parameters and histological structures in rats<sup>(117)</sup>. Banerjee *et al.*<sup>(118)</sup> also studied the effects of chronic fresh garlic homogenate in three different doses (250, 500 and 1000 mg/kg per d) for 30 d on various endogenous antioxidant enzymes and lipid peroxidation in two major organs, the liver and kidney. It was observed that in comparison with saline-treated rats, the 250mg/kg per d dose significantly reduced the lipid peroxidation levels and increased SOD activity significantly. But higher doses of garlic (500 and 1000 mg/kg per d) significantly reduced endogenous antioxidants (catalase and SOD) without altering lipid peroxidation levels. A 1000 mg/kg per d dose of garlic caused marked histopathological and ultrastructural changes in both liver and kidneys. Thus, garlic in low doses has the

potential to enhance antioxidant status, although at higher doses a reversal of these effects is observed. Garlic has the potential ability with a high dose to induce morphological changes in the liver and kidneys, indicating the need to identify a safe dose range for garlic. Prolonged feeding of high levels of raw garlic in rats has resulted in anaemia, weight loss and failure to grow due to the lysis of erythrocytes<sup>(38)</sup>. Raw garlic juice at a dose of 5 ml/kg has resulted in the death of rats due to stomach injury<sup>(125)</sup>. Surviving rats exhibited swelling of the liver, hypertrophy of the spleen and adrenal glands and the decrease of erythrocyte count with various morphological changes after 3 and 8 d. Chen *et al.*<sup>(126)</sup> have also reported that treatment of rats with fresh garlic homogenate for 7 d caused a significant decrease in liver catalase activity in doses of 2 and 4 g/kg per d. Garlic oil fed to rats at a dose of 100 mg/kg per d in fasting conditions has also been found to be lethal. The cause of death appears to be acute pulmonary oedema with severe congestion<sup>(127)</sup>. Garlic oil and DADS (200 mg/kg) significantly reduced the body-weight gain of rats<sup>(128)</sup>. One possible explanation for all the above-mentioned toxicity reports is that the sulfoxides present in garlic extract undergo exchange reactions with –SH groups of enzymes and other proteins in the body at physiological pH and temperature, so inhibiting their activity<sup>(129)</sup>.

The effect of raw garlic at a dietary dose on platelet function has been studied by Scharbert *et al.* who reported that single consumption of fresh garlic up to 4.2 g/d has no inhibitory effect on platelet function in healthy volunteers<sup>(130)</sup>. Gadkari *et al.*<sup>(131)</sup> showed increased clotting times from 4.15 to

5·02 min by investigating the *in vivo* effect of raw garlic intake of 10 g for 2 months. Asdaq & Inamdar have concluded that garlic in a moderate dose (250 mg/kg) with added hydrochlorothiazide possesses synergistic cardioprotective and antihypertensive properties against fructose- and isoproterenol-induced toxicities<sup>(132)</sup>. A dose of garlic homogenate (250 mg/kg per d) either alone or with propranolol showed significant increase in activities of antioxidant enzymes such as SOD and catalase during ischaemia-reperfusion injury<sup>(133)</sup>. Satitvipawee *et al.*<sup>(134)</sup> showed no statistically significant changes in serum total cholesterol, TAG, LDL-cholesterol and HDL-cholesterol after 12 weeks of treatment with enteric-coated Thai garlic extract tablet once daily (standardised to 1·12 % allicin or 5·6 mg/tablet). In addition, no changes in plasma glucose, liver and renal functions were found<sup>(134)</sup>.

## Summary

In the present review, antioxidant, antilipaemic, hepatoprotective, haematological, antimicrobial and antineoplastic actions of garlic have been shown. The studies reviewed reveal that the benefits of garlic are not limited to a specific species, a particular tissue, or a specific carcinogen. Oil-soluble compounds such as DADS have been observed to be effective in reducing the proliferation of neoplasms. However, it has also been reported that higher concentrations of garlic powder cause considerable cell injury in the liver of rats, which is not observed at lower concentrations.

Thus, although the evidence supports the benefits of garlic, additional evidence is needed to determine the quantity

required by humans to minimise cancer, cardiovascular risks and haematological diseases, and to provide hepatoprotection and antilipaemic and antimicrobial effects.

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