Research Journal of Pharmacognosy (RJP) 5(2), 2018: 71-89
Received: 22 Mar 2017
Accepted: 23 Sep 2017
Published online: 13 Mar 2018
DOI: 10.22127/RJP.2018.58511

Review article

Isothiocyanates: a Review

Chandra Kala[1*], Syed Salman Ali [2], Nabeel Ahmad[3], Sadaf Jamal Gilani[1], Najam Ali Khan[2]

[1]Glocal School of Pharmacy, The Glocal University, Uttar Pradesh, India.
[2]School of Pharmaceutical Sciences, IFTM University, Uttar Pradesh, India.
[3]Department of Biotechnology, IFTM University, Uttar Pradesh, India.

Abstract

Isothiocyanates (ITCs) are naturally occurring molecules belonging to highly reactive organosulphur synthons, with the general structure R–N=C=S. The precursor molecule glucosinolate anions are hydrolyzed enzymatically (under the effect of myrosinase enzymes) or unenzymatically to produce nitriles or isothiocyanates depending upon conditions such as pH and temperature. Brassicaceae Family is known to contain abundant ITCs. A significant number of isothiocyanates has been isolated from different plant sources and some of them have been synthesized. Several isothiocyanates have demonstrated significant pharmacological activities including anti-cancer, anti-inflammatory, anti-microbial activities, *etc*. Pharmacokinetic profiles of these sulphur containing compounds are well established. However, safety profiles of ITCs need consideration and a well-designed study with appropriate control, for their production as lead compounds. This review summarises the chemistry, sources, pharmacokinetics, pharmacological activity and toxicity profiles of the isothiocyanates.

Keywords: Brassicaceae; cruciferous vegetables; isothiocyanates; pharmacokinetics; pharmacological activity

Citation: Kala C, Ali SS, Ahmad N, Gilani SJ, Ali Khan N. Isothiocyanates: a review. Res J Pharmacogn. 2018; 5(2): 71-89.

Introduction

Isothiocyanates (ITCs) are naturally occurring molecules belonging to highly reactive organosulphur synthons [1], with the general structure R–N=C=S [2]. Isothiocyanates present in a vegetable are responsible for the plant sharp taste and active in its defence system [3]. They are derived from glucosinolates. Glucosinolates are thioesters, which upon loss of cellular integrity activate the enzyme glucosinolates myrosinase leading to generation of several unstable intermediates which rearrange into degradation products. These degradation products are transformed into isothiocyanate, oxozolidine-2-thiones, nitriles, *etc*. depending upon different factors like pH, presence of myrosinase-interacting protein, and availability of ferrous ion [4].

Isothiocyanates are found in cruciferous vegetables such as broccoli, rocket, cauliflower, Brussels sprouts, cabbage, radish, turnip and watercress [5]. Sixteen families of dicotyledonous angiosperms contain glucosinolates, which are precursor of ITCs. In several reviews, it has been mentioned that Brassicaceae family primarily contains ITCs [6].

Previously, these compounds were considered for having a negative impact on health as some anti-nutritional or goitrogenic glucosinolates were found in widely grown oilseed crops and in domesticated crops. However, more recent studies have reported that therapeutic and prophylactic properties of these compounds were revealed by numerous researches [6]. They possess strong anti-oxidant, anti-inflammatory activity, anti-microbial, neuroprotective and cardioprotective activity. Isothiocyanates are considered to be safe and no serious adverse effects have been reported in humans. They possess potent anti-carcinogenic activity and are

*Corresponding author: chandrakala@theglocaluniversity.in

active against many forms of cancers and tumours, but when given after chemical carcinogens in high concentration, *i.e*. 25-250 times more than normal dietary intake, they promote bladder cancer in rats [7]. However, there is some information showing that isothiocyantes act against bladder and prostate cancer [7]. Some ITCs are known to form adducts with DNA and induce gene mutation and aberration. Thus, there is need of considering the genotoxicity of ITC [8].

The present review summarizes the sources, chemistry, pharmacokinetics, pharmacological activity and toxicity profile of ITCs.

Methods

Available scientific databases including Science direct, Pub med, Elsevier, Scopus, Research scholar, Google scholar were explored for chemistry, pharmacokinetics and pharmacology of ITCs. US patents were also referred for details of *Lepidium meyenii* roots. Reports of toxicological profile of ITCs were obtained from Oriental mustard seed (OMS) US EPA office of pesticide program and Bureau of pest management, New York.

Results and Discussion

Chemistry of isothiocyanates

ITCs are basically esters of isothiocyanic acid (H-NCS). They may also be considered as sulfur analogues of isocyanates (R-NCO). They are isomeric with thiocyanates (R-SCN) and isoelectric with thioketones ($R_2C=C=S$) [1]. The hydrogen atom on the adjacent carbon to the isothiocyanate moiety is also responsible for the tautomeric structure of methylene-isothiocyanate moiety to beta-unsaturated thioketones [10]. It has been suggested that ITCs possess high electrophilicity due to the presence of the carbon atom and high nucleophilicity associated with sulfur atom. The dipole moments of ITCs as determined by several workers indicate that –NCS group possess electron accepting character and negative end of dipole and hence may serve as a versatile building block for preparation of wide class of nitrogen, sulphur and oxygen heterocycles. They possess extended pi-electron system contributing to its ability to variety of target molecules. [1]

There are various reports mentioning the health benefits of ITCs in ailments such as in cancer, cardiovascular diseases CVDs and diabetes. The pharmacological benefits of the ITCs are attributed to the electrophilic nature of carbon residue which reacts with biological nucleophiles [8] and with nutrients such as amines, amino acids, proteins, thiols and alcohols to give variety of compounds such as thioureas and dithicarbamates [10]. ITCs react with N-terminal residues of alpha-amino group of lysine through alkylation. [11]

Physicochemical properties such as solubility and acid-base property also exert effects on biological properties of isothiocyanates. The activity of ITCs against 4-(methylnitrosamino)-1-(3-pyridyl)-1- butanone (NNK)-induced lung tumorigenesis is favoured at their high lipophilicity and low reactivity against GSH [9].

Exchange of substituents does not result in remarkable changes in reactivity of NCS group. In ITCs of aryl alkyl type or substituted phenylsulphide and phenylsulphone type, lengthening of ITC does not affect NCS group reactivity [12].

Pharmacokinetics of ITCs

Absorption

ITCs are rapidly absorbed across intestinal cell membrane via passive diffusion [13]. Allyl isothiocyanate (AITC) is known to be absorbed from small bowel and colon and its metabolite can be easily detected using urine determination of [^{14}C] AITC (25 or 250 μmol/kg body weight) [14]. About 90% of orally administered AITC is absorbed, confirming its high bioavailability [15]. In a research, human intestinal effective permeability and intestinal metabolism of sulforaphane was studied in human jejunum (loc-1-gut). The percentage absorption of sulforaphane through the jejunum was found to be 74±29% [16].

In another study, it was found that broccoli sprouts ITCs were largely absorbed with a peak concentration of 0.943-2.27 μm/L in plasma, serum and erythrocytes 1 h after feeding [17]. One of the interesting facts is that, absorption of ITC is lower from cooked food compared to raw vegetables because the colon microflora is able to catalyse the hydrolysis of glucosinolate in a cooked form [7].

Distribution

In a study conducted in mice fed with purified

sulforaphane and erucin, both metabolites were found in skin, liver, kidney, bladder, lungs and plasma by HPLC-MS/MS analysis. Erucin was the favoured form in the liver, kidney and bladder, suggesting systemic benefits of both [18]. In another report regarding tissue distribution and metabolism of sulforaphane in wild type and Nrf2 knockout mice, sulforaphane metabolites were detected in all tissues at 2 and 6 h following administration, and the highest concentration was observed in the small intestine, prostate, kidney and lungs [19].

Metabolism

Organic ITCs are conjugated with glutathione (GSH) to form dithiocarbamates. These dithiocarbamates are metabolized by γ-glutamyl transpeptidase (GGT) to form cysteinyl glycine ITC conjugates. These are further metabolized to form cysteine-ITC conjugates and by N-acetyl transferase to form NAC-ITC conjugate [20,21].

GSTM1 and GSTT1 catalyse the conjugation and excretion of ITCs, and these conjugates are substrates of ATP-binding cassette (ABC) transporter [22]. P-glycoprotein (P-gp, ABCB1), multi drug resistance protein (MRP1, ABCC1), (MRP2, ABCC2) and breast cancer resistance protein (BCRP ABCG2) are known to interact with ITCs [22]. To support the above fact the kinetic property of the glutathione S-transferases (GSTs) GSTs A1-1, M1-1, M4-4 and P1-1 with 14 ITCs (propyl-, butyl-, pentyl-, hexyl-, (±)-2-hexyl, cyclohexyl-, cyclopropyl-, cyclooctyl-, benzyl-, phenethyl-, allyl-, erucin, sulforaphane, erysolin) was investigated. The result of that study showed that GST M1-1 and GST P1-1 were the main contributor of metabolism of ITC. Liver tissue was detected with 0.1 mM concentration of GST A1-1 and GSTM1-1 revealing their major role in conjugation of ITCs [22].

Four genotypes of GST (GSTP1, GSTM1-1, GST-A1-1 and GSTM2-2) were tested for conjugation with GSH for allyl-benzyl-PEITC and sulphoraphane. Among these four genotypes, GST-P1-1 and GSTM-1 were the most efficient catalysts and GSTM2-2 was least efficient. Among the four ITCs, benzyl ITC was the most rapidly conjugated while, sulforaphane was only slowly conjugated [23].

Steck *et al.* conducted a study with 114 individuals to assess the association of the GST with urinary ITC metabolite following ingestion of broccoli. The HPLC cyclocondensation assay was used for measuring urine ITC metabolites and matrix assisted laser desorption/ionization time of flight was used for GSTM1, GSTT1 deletion GSTP1 11e105Val and GSTA1*A/*B genotyping. The result of the study revealed that there were no substantial differences in ITC levels among genotypes, supporting the idea of alternative routes of ITC metabolism [24].

Comparative metabolism of different ITCs was studied on F344 rat liver microsomes [25]. The enzyme responsible for ITC metabolism was induced by treating the rats with Aroclor 1254, beta-napthoflavone, isosafrole or phenobarbital. The result of the study revealed that rate of metabolic conversion followed the order of 1-naphthyl>>phenyl>benzyl & phenethyl >>propyl, ethyl and methyl [25].

ITCs are potent inducers of phase-II enzymes such as UDP-glucoronosyltransferase (UGTs), sulphonyltransferase (SULTs) and quinone reductase (QRs) and hence detoxify activated carcinogens. For example, consumption of broccoli and watercress inhibit CYP450 enzymes and reduces the risk of lung cancer [25].

In a research, it was revealed that heme destruction and protein modification was responsible for the inactivation of CYP2E1 by phenyl ethyl isothiocyanate (PEITC). GSH metabolize PEITC to form PIC (phenethyl isocyanates). PIC bounds to CYP2E1 but does not inactivate them. Instead, the reactive sulphur atom generated during desulfurization of PEITC is involved in inactivation of CYP2E1, as revealed by LCMS of inactivated CYP2E1 apo-protein [26].

Excretion

The mercapturic acid pathway was found to be the major route of elimination of ITCs. Defecation, exhalation and perspiration were found to be the minor routes. Several studies support his fact e.g. pure benzyl isothiocyanate (BITC administered in a human was 54% recovered as mercapturic acid in the urine. Similarly PEITC from watercress was 47% recovered as mercapturic acid [27]. A study on metabolism and excretion of BITC revealed that 53.7% of BITC was excreted as metabolites by the renal route. The metabolite was completely excreted 10-12 h after administration [28].

A similar study was conducted to find out the disposition of AITC in rat and mouse. The urine was found to be the major route of excretion of ^{14}C AITC about 50-80%, with smaller amounts about 6-12% in faces and 3-7% in expired air [29]. Moreover, inorganic thiocyanate, allylthiocarbomoylmercapturic acid and allylthiocarbomoylcysteine were found in mice but no cysteine conjugate was found in rat urine. Also, approximately 80% of ^{14}C were present in mercapturic in rat, suggesting that hydrolysis is the major metabolic pathway of AITC in mouse whereas glutathione conjugation is the major metabolic pathway of AITC rats [29].

Borghoff *et al.* evaluated age related changes in excretion of AITC [30]. The study was conducted in male Fischer rats of age 3, 16 and 27 months. Difference in elimination of AITC in urine (which is a major route of excretion) was not significant. However, faecal excretion of AITC decreased with age and bile excretion increased at 16 month and decreased afterward as compared to 3 month age group. Age related decrease in $^{14}CO_2$ was also observed [30].

The relationship between dietary total ITC and GST M1/T1/P1 genotype was reported in one observational study by measuring total ITC in urine sample of 246 Singapore Chinese individuals [31]. The results revealed that there was no difference in ITC level of GSTM1-null and GSTM1-positive subjects. However, significant difference was observed in urine ITC level of GSTT1 null and positive individual. The null genotypes were associated with lower excretion levels [31].

Pharmacological activity of ITCs

Cytotoxic activity of isothiocyanates

Anti-carcinogenic activity of isothiocyanates has been known for decades. Recently anticancerous activity of synthetic ITC, CM9, and its fullerene derivative was studied in human jurkat T-leukemic cell [32]. CM9 is an naphthalenetetracarboxylic diimide (NDI) consisting of two side chains: a *N,N'-bis* [3,3'-(dimethylamino) propylamine]-naphthalene-1,4,5,8-tetracarboxylic diimide (N-BDMPrNDI) derived protonated dimethylaminoethyl side chain and an ITC group as a second side chain. The result of the study showed that CM9 reduced Jurkat leukemic cell viability in a concentration dependent manner [32].

The anticancer activity of phenethyl isothiocyanate (PEITC) was studied by several researchers [33]. The anti-proliferative and pro-apoptotic effect of PEITC was studied on human cervical HeLa cell line in cancer stem like cells using a cell proliferation assay, immunoblotting and flow cytometry [33]. It was found to possess anti-cancer activity.

The anticancer potency of SFN, along with other anti-proliferative agents was evaluated against Barrett oesophageal adenocarcinoma (BEAC) [34]. Various parameters including the effects on drug resistance by Rhodamine efflux assay and the induction of apoptosis using annexin V labelling and Western blot analysis of poly (ADP-Ribose) polymerase cleavage were evaluated. The study showed that SFN induced a decline in cell survival, cell cycle arrest and apoptosis. It also reduced tumour volume and increased the activity of other anti-proliferative agents [34]. Sulphoraphane fractions (standard SFN, extract and purified SFN) of *Brassica oleraceae* var *rubra* were also evaluated for anti-cancer activity against human epithelial carcinoma HEp-2 and Vero cells [35]. Sulphoraphane was found to be potent against pancreatic cancer cell Mia Paca-2, Panc-1, AsPc-1, and BxPc-3 and inhibited pancreatic cancer cell growth and induced apoptosis [36]. It was also found to be potent against ovarian cancer both in human ovarian cancer cell line SKOV3 and mouse ovarian cancer cell lines C3 and T3 [37]. Similarly, SFN was found to be a potent inhibitor of A-549 lung cancer cell growth [38].

Similarly, allyl isothiocyanate AITC which commonly occurs in cruciferous vegetables is a potent inhibitor of many types of cancer [39]. It has inhibited both human bladder cancer UM-UC-3 cells and rat bladder cancer AY-27 cells proliferation as evaluated using orthotropic rat bladder cancer model [40]. It has also exhibited potent anti-proliferative activity against MDA-MB-468 human breast cancer cells [41] and against human A-549 and H-1299 non-small cell lung cancer (NSCLC) cells *in vitro* [42].

Mechanism of action

1. Modulation of B-cell lymphoma-2 (Bcl-2), Bcl associated X protein (Bax), Tumor protein p53 ()p53, vascular endothelial growth factor (VEGF), cyclin B1 and capsases-3 expression leading to activation of

intrinsic pathway of apoptosis as determined western blotting and reverse transcription polymerase chain reaction (RT PCR) in human epithelial carcinoma HEp-2 and verocells, and orthotrophic rat bladder cancer model [32,35,40].

2. The mechanism of anti-proliferating effect of phenylethyl isothiocyanate (PEITC) was found to be mediated by up regulation of death receptor 4 (DR4) and death receptor 5 (DR5) of TNF-related apoptosis-inducing ligand (TRAIL)-mediated apoptotic pathways [33].
3. The anti-cancer activity of SFN was attributed to induction of caspase 8 and p21 and down regulation of heat shock protein 90 (hsp90) as determined by western blot and RT-PCR in human epithelial carcinoma HEp-2 and Vero cells [35]. It also targets Kelch-like ECH-associated protein 1 (Keap1)-Nuclear factor (erythroid-derived 2)-like 2 (Nrf2) complex and induced phase 2 genes. It targets Bcl-2 protein, death receptor, Mitogen-activated protein kinases (MAPK), mitochondria-associated apoptotic protein and induce programmed cell death, it targets cyclins, cyclin-dependent kinases (CDKs), Cdc25C (gene), histone deacetylases (HDAC), p21, microtubules and cause cell cycle arrest mainly G2/M phase. It targets matrix metalloproteinases (MMP)-2 and MMP-9 and inhibits invasion and metastasis [43]. Degradation of heat shock protein (HSP) 90 and blockade of interaction between HSP90 and its cochaperone p50 in pancreatic cancer cells are also targets of sulforaphane as determined by western blotting [36].
4. Inhibition of Akt (protein kinase - B) - signal transduction pathway as determined by effect of sulforaphane in human ovarian cancer cell line SKOV3 (IC_{50} 40 μmol/L) and mouse ovarian cancer cell lines C3 and T3 (IC_{50} 25 μmol/L each) using 2,3-bis(2-methoxy-4-nitro-5-sulfophenyl)-*2H*-tetrazolium-5-carboxanilide inner salt (XTT) assay [37]. Increased ubiquitination and degradation of α- and β-tubulin leading to inhibition of mitosis and induction of mitochondrial mediated apoptosis in bladder cancer cell as determined in orthotopic rat bladder cancer model [40].
5. γH2AX (phophorylated form of H2A histone family, member X) and Fanconi anemia (FA) complementation group proteins D2 (FANCD2) foci, ataxia telangiectasia mutated protein kinase (ATM) / ataxia telangiectasia and Rad3 relatde protein kinase (ATR) - and S and G2/M cell cycle arrest are identified as potential targets [42].

Antimicrobial activity

Antimicrobial activity of ITCs has been reported by several researchers. ITCs possess inhibitory activity against food spoilage bacteria species such as *Escherichia. coli*, *Klebsiella* spp., *Salmonella* spp., *Bacillus* spp., *Serratia* spp., *Staphylococcus* spp. and *Listeria* spp. Gram negative bacteria were found to be more sensitive to ITCs compared to Gram positive bacteria [44]. Isothiocyanate isolated from horseradish was evaluated for its anti-bacterial property against 4 strains of antibiotic-resistant bacteria: methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-resistant *S. aureus* (VRSA), multidrug-resistant *Acinetobacter baumanii* (MRAB), and multidrug-resistant *Pseudomonas aeruginosa* (MRPA), and 3 clinical strains of pathogenic bacteria: *S. aureus, A. baumanii*, and *Pseudomonas aeruginosa* by measuring minimum bactericidal concentrations (MBC) [45]. ITCs showed maximum activity against *A. baumanii* and *P. aeroginosa*, with MBC values substantially less than the standard antibiotics vancomycin, levofloxacin, and were comparable with that of norfloxacin [46].

4-(α-L-Rhamnosyloxy) benzyl isothiocyanate and 4-(4'-O-acetyl-α-L -rhamnosyloxy)-benzyl isothiocyanate isolated from *Moringa oleifera* was evaluated for antibacterial activity against *Staphylococcus aureus, S. epidermidis, Bacillus subtilis, Escherichia coli, Enterobacter aerogenes, Klebsiella pneumoniae*, and *Pseudomonas aeruginosa*, and for their antifungal activities against *Candida albicans, Trichophyton rubrum,* and *Epidermophyton floccosum* using the disk diffusion method. Both of these compounds exhibited moderate to good antimicrobial activity [47].

Phenethyl, benzyl, benzoyl ITCs are known to inhibit harmful intestinal bacteria *Clostridium difficile*, *Clostridium perfringes* and *E. coli*. SFN, BITC are known to be active against Gram

positive and negative bacteria of human faecal isolates. β-Phenylethyl ITCs are known to possess strong activity against *Vibrio parahaemolyticus* and *S. aureus*; considerable activity against *Bacillus cereus* and *Vibrio parahaemolyticus*. Allyl, benzyl, 4-methylthio-3-butenyl are also known to possess strong anti-bacterial activity against *E. coli* ATCC 11775, *Enterobacter cloacae* ATCC 13047, *Salmonella typhimurium* ATCC 13311, *Proteus vulgaris* ATCC 6380, *S. aureus* S-6, *Staphylococcus epidermidis* ATCC 14990, *Bacillus subtilis* ATCC 6633, *B. cereus var. mycoides* ATCC 11778 and *Campylobacter jejuni* [48].

Mechanism of action

1. The action is attributed to interaction of these compounds with sulfhydryl of glutathione containing enzymes followed by inhibition of sulfahydryl enzymes activities [49]. The linkage between ITC and enzyme limits enzymatic activity and leads to formation of oxygen and other free radicals thus reducing the content of important thiol group and reducing the viability of bacterial cell [50]. The rate of spontaneous degradation of ITC-thiol conjugates and bacterial detoxification mechanism determine the efficacy of particular ITC [51].
2. The effect on cell membrane and leakage of cellular metabolite and increase in 3-galactosidase activity [46].
3. ITCs interact with cytochrome-450 and get oxidized to produce more reactive isocyanates which are more toxic than the parent compound as determined by cytochrome-450 dependant oxidative desulfuration of 2-naphthylisothiocyanate to yield 2-naphthylisocyanate [50].

Activity against neurodegenerative diseases

Sulforaphane, one of the potent isothiocyanates which possesses various pharmacological activities, is also active in neurodegenerative disorders [52]. Alzheimer's disease is one of the most common neurodegenerative diseases. It is characterized by accumulation of amyloid beta peptides resulting in oxidative damage and inflammation [52]. Another neurodegenerative disease, Parkinson's disease (PD), is characterized by loss of dopaminergic (DA) neurons [52]. PD also involves autooxidation of dopamine, leading to the formation of neurotoxic species including electrophilic DA quinone, ROS and hydrogen peroxide, which mediate α-synuclein-associated neurotoxicity [52]. Loss of function and mutation in genes of phase-II detoxification pathway (especially glutathione metabolism) also contribute to neuronal death accompanying enhancement of α-synuclein expression. Sulforaphane is potent against both of these neurodegenerative conditions [52]. SFN has been able to overcome neuronal loss in various models of Parkinsonism like *Drosophila* model of alpha-synucleinopathy and *Drosophila* parkin mutants. [52] SFN is also protective against spinal cord injury [53]. Erucin, an isothiocyanate, possesses neuroprotective effects on dopaminergic like neuroblastoma SH-SY5Y cell line. [54] Neuroprotective effects of synthetic ITC 4-iodophenyl isothiocyanate (4-IPITC) was evaluated in several *in vitro* and *in vivo* models of neurodegenration including the exposure of primary cortical neurons to glutamate, oxidative stress, oxygen, glucose deprivation, 1-methylphenylpyridinium (MPP+) and experimental autoimmune encephalomyelitis (EAE). 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) induced Parkinson's disease [55]. That study revealed that 4-IPITC significantly reduced neuronal cell death and possessed neuroprotective and neurotrophic properties [55]. In light of these researches, it could be concluded that isothiocyanates possess potent neuroprotective activity.

Mechanism of action

1. Sulforaphane has induced proteasome expression in murine neuroblastoma Neuro2A cells via carbonyl formation and hydrogen peroxide induced cytotoxicity and ultimately protected neuronal cells from amyloid beta peptide mediated cytotoxicity in Aleizhmer's disease [54].
2. SFN has been able to enhance glutathione synthesis or glutathione conjugation activity. SFN also caused reduction of DA quinone level as revealed in various *in vitro* models [52].
3. SFN has prevented nigral dopaminergic neurons cell death and causes astrogliosis, microgliosis inhibition in basal ganglia as

revealed in 1-methyl-4'-phenyl-1,2,3,6-tetrahydropyridine mouse model of PD [55].

4. L-SFN has activated Nrf2-antioxidant response (ARE) transcriptional pathway which protects neuron degeneration in rat nigrostraital tissue slice model. [56]
5. SFN has also modulated extracellular signal–regulated kinases **(**ERK)1/2 neuronal survival pathway in brain of mice [57].
6. SFN is known to inhibit caspase-3 enzyme activation and has reduced the phosphorylation of mitogen activated protein kinase (MAPK) signaling pathway, oxidative stress. It has enhanced intracellular glutathione level, methylglyxalase detoxifying system and activity of glyoxalase 1 [58,59].
7. In spinal cord injury, treatment with sulforaphane has up regulated phase 2 antioxidant response, decreased mRNA level of Matrix MetalloProteinase (MMP)-9, an inflammatory cytokine and increased the serotonergic axon caudal counting to the lesion site [52].
8. Treatment with erucin has resulted in significant elevation of GSH levels and reduction of reactive oxygen species and $O\bullet^{-}$ formation resulting in resistant to neuronal cell death [53].

Radioprotective activity

Radioprotective activity of AITC and PITC was evaluated in Swiss albino mice. The whole body of the mice were irradiated by gamma radiation by a Co^{60} source (dose rate = 0.98 Gy/min). Parameters such as serum alkaline phosphatase (ALP), liver glutamate pyruvate transaminase (GPT), haemoglobin, total erythrocyte count and total leucocyte count were monitored. Both ITCs reduced the level of these enzymes significantly, which had been elevated due to irradiation. ITCs also enhance the GSH content of intestinal mucosa and liver. [60] Radioprotective activity of seed extract of *Brassica campteris* which is rich in glucosinolates and isothiocyanates was also evaluated and the extract was found to be effective against radiation induced heamtological and biochemical changes [61].

Radioprotective activity of seed extract of *Moringa oleifera*, which is rich in isothiocyanates, was studied against gamma radiation (8 gy) induced irradiation in mice. [62] The extract reduced RNA and acid phosphatase levels significantly, revealing its radioprotective activity.

Cardioprotective activity

Cardiovascular disease (CVD) is the largest cause of disability and morbidity in developing countries. Despite advancement in understanding the pathobiology and treatment of CVD, we are facing the global problem in alleviating the burden of CVD. At present, much research focuses on validation of nutraceuticals. Many studies have indicated that consumption of cruciferous vegetables is associated with decreased risk of CVD [63]. This category is found to be rich in isothiocyanates [63].

Consumption of broccoli (which is rich in sulforaphane) is associated with reduced risk of coronary artery disease in postmenopausal women. It has also reduced LDL and total cholesterol and increases HDL cholesterol. Furthermore, broccoli consumption has reduced ischemic reperfusion injury mediated cardiomyocyte death [64].

4-Carboxyphenyl isothiocyanate (4-CPI) was evaluated for its cardioprotective effect in Lagendroff perfused heart of wistar rat and C57BL/6J mice, an *ex vivo* model of ischemia reperfusion injury [65]. 4-CPI significantly improved functional parameters and reduced the diameter of ischemic areas. It showed vasorelaxation effects on the coronary artery, and membrane hyperpolarisation effects on vascular smooth muscle cells [65].

Mechanism of action

1. SFN activates the AKT / Protein kinase B and extracellular signal-regulated kinas 1 and 2 (ERK1/2) signaling pathway, these pathways are implicated in cardiac cell survival. It also modulates expression and activity of glutathione reductase, glutathione-S-transferases, thioredoxin reductase and NAD(P)H: quinone oxidoreductase via activation of phosphatidyl 3-kinase/AKT pathway [66].
2. Nuclear factor-E2-related factor 2 (Nrf2), a basic leucine zipper transcription factor, signaling pathway plays a significant role in regulation of gene expression of phase II cytoprotective enzymes. In absence of oxidative stress (Nrf2) pathway remains

inactive as Nrf2 is sequestrated and degraded in the cytoplasm. As soon as the cell experiences oxidative stress, Nrf2 is translocated to the nucleus where it induces transcription of antioxidant genes. It also promotes the transcription of many cytoprotective genes. Another factor named Kelch-like chicken erythroid-derived cap "n" collar homology (ECH) factor-associated protein 1- (Keap1) is a negative regulator of Nrf2. Keap1 forms complex with Nrf2 and promotes its rapid proteasomal degradation. But during oxidative stress, disruption of Keap 1-Nrf2 complex takes place (through transduction of phosphorylation and redox modification of the complex) which ultimately promotes translocation of Nrf2 to the nucleus. SFN, an isothiocyanate exerts cytoprotective effect by activation Nrf2 pathway in three ways: 1) Direct effect on Nrf2 related signaling pathway , as it is an electrophile that interact with Keap cysteine residue and block degradation and polyubiquitination of Nrf2 and induce significantly higher level of Nrf2 expression in cardiac cell nuclei. 2) The indirect effect on Nrf2 related signaling pathway, as it activates protein kinase B and extracellular signal regulated kinase 1 and 2 signaling pathway, which play role in cardiac cell survival. 3) SFN also mediate long term protection against free radical induced cell damage. Therefore, SFN is found to be potent against many cardiovascular diseases like hypertension, atherosclerosis, and cardiac ischemia reperfusion injury [63].

3. The cardioprotective effect of broccoli is mediated through inhibition of phase I enzyme and DNA adduct, angiogenesis, inflammation and induction of phase II enzymes and cell cycle arrest [64].
4. The mechanism of action of 4-CPI in cardioprotection was attributed to H_2S donor capacity (H_2S play pivotal role in regulation of blood pressure and cardiac function) and activation of mitochondrial K^+-ATP channel activation [65].

Anti-diabetic activity

Studies on antidiabetic activity of isolated isothiocyanates are scarce. Although crude extracts of plants rich in isothiocyanates are reported to possess antidiabetic activity, confirmation that the activity is due to the presence of isothiocyanates has not been established. There are few reports e.g. SFN affords protection against diabetes. It is reported to block the development of type 1 diabetes in streptozotocin treated mice by inhibiting cytokine and oxidative stress induced beta cell damage and protects against diabetes induced complications [63]. SFN is also known to protect aortic damage in streptozotocin (STZ) induced diabetes in T1D mice [66].

Mechanism of action

Activation of Nrf2 leading to suppression of NF-κB mediated pathway. These pathways make antioxidant defense system. Oxidative stress and inflammation plays pivotal role in diabetes and its complication [67].

Anti-platelet activity

1-Isothiocyanato-4-methylsulfinylbutane has been evaluated for its anti-platelet activity using *in vitro* techniques. In these studies, collagen, adenine diphosphate and thrombin were used for platelet aggregation. 1-Isothiocyanato-4-methylsulfinylbutane has inhibited collagen induced platelet aggregation significantly. It also inhibited collagen and epinephrine induced pulmonary embolism [68]. 1-Isothiocyanato-4-methylsulfinylbutane anti-platelet activity was confirmed *invivo* using ADP-induced acute pulmonary thromboembolism in mice [69]. Allyl isothiocyanate was evaluated for its anti-platelet activity via *in vitro* techniques [68]. The study suggested that it possessed remarkable anti-platelet activity.

Mechanism of action

1. Anti-platelet activity of 1-Isothiocyanato-4-methylsulfinylbutane may be attributed to activation of intrinsic pathway and inhibition of cycloxygenase and glycoprotein IIb/IIIa [69].
2. Activation of adenyl cyclase/cAMP pathway followed by inhibition of intracellular cascades such as PI3-kinase/Akt and collagen-induced phosphorylation of phospholipase C (PLC) γ2- protein kinase C (PKC) -p47 pathway and eventually inhibition of platelet activation and aggregation [68].
3. Anti-platelet activity of AITC was attributed

to the inhibition of TXA2 production, ATP release, cellular calcium increase and phosphorylation of PKCδ, p38, ERK, Akt [70].

Toxicity profile

The risk benefit ratio of isothiocyanates is controversial and hence the toxicity profile of ITCs needs to be elaborated. Various data from *in vitro* studies suggest that ITCs exhibit genotoxic potential as they form adducts with DNA. However, the dietary concentration of ITCs was found to be several orders of magnitude lower compared to the doses used in genotoxicity studies. A precise evaluation of toxicological profile is required to predict potential genotoxicity of ITC and for any recommended clinical use [71]. Listed here are the toxicity profiles of some individual ITCs on the basis of which some conclusion can be drawn.

Benzyl isothiocyanate (BITC)

Daily dose of BITC (12 µm) was found to be safe and did not possess any toxicity. However, in rats treated with a substantially higher dose (200 mg/kg), alteration in various parameters were observed including reduction in haemoglobin, lymphocyte count and elevation in neutrophils, eosinophils and platelets and increased cholesterol and renal dysfunction. Adverse effects were not observed at a dose of 50 mg/kg and hence that dose was considered to be safe at therapeutic dose. BITC induced mutagenesis in the Ames assay and also caused chromosomal aberration and DNA strands breaks. It is also known to induce bladder carcinogenesis [72].

In a study examining the sub-acute toxicity of BITC in male rats at oral doses of 0, 50, 100 and 200 mg/kg B.W/day for 4 weeks, BITC induced haematological alteration of toxicological relevance and decreased TG level at 200 mg/kg and increased serum cholesterol at all doses [73], indicating renal dysfunction.

Allyl isothiocyanate (AITC)

As per the report of agency vegetable oil RED, Dec 1993, AITC is not likely to cause adverse human health effects and the oral lethal dose in rats was found to be 339 mg/kg [74]. AITC at a dose of 20 mg/kg *s.c.* has reduced WBC counts (mainly lymphocytes and monocytes) and increased neutrophils count. At a dose of 100-150 mg/kg, AITC increased AST levels. However, the toxicity was suggested to be dose dependant [73]. AITC was moderately and acutely toxic and caused skin and eye irritation, although it has been systemically safe in subchronic feeding study in laboratory animals [75]. A dose of 25 mg/kg/day for 90 days was reported to be safe and no clinical effects were observed. According to one report, fourteen individual tests were conducted to determine the mutagenic effects of AITC. Among these, eight were considered to be negative as they did not develope any mutagenecity, three were positive (developed mutagenicity) and two were equivocal (ambigous) [74].

Mustard oil (containing 93-97% AITC) was used to access the developmental toxicity and mutagenecity. Low observed adverse effect level (LOAEL) of AITC was found to be 2.8 and 23.8 mg/kg/day (in rabbits and hamsters) for incidences of extra sternebrae and incomplete ossification of sternebrae [74].

Phenylethyl isothiocyanate (PEITC)

PEITC has been reported to be safe and does not possess any apparent toxicity when administered at high doses (as revealed by safety studies in rats and dogs) [33]. At doses of 80 and 160 mg/kg PEITC *i.p.*, increased weight of liver and spleen. Effective concentration of PEITC were 0.12 to 14 µM [75].

PEITC is currently in phase I trials and found to be active against lymphoproliferative disorders. The trial is conducted by University of Texas – M.D Anderson Cancer Centre. [75]

Sulforaphane

There is lack of confirmatory evidences whether SFN is safe or possesses some toxicity. SFN at a dose of 64 mg/kg (in mouse) has been reported to induce hepatotoxicity and there are studies which contradicts such toxicity [76].

Various isothiocyanates from plant sources and their potential benefits

Glucosinolate and their breakdown product, isothiocyanates, exhibit diverse therapeutic potentials. For decades, studies have been conducted to explore more and more potential

benefits of isothiocyanates and their toxicity. Numerous plants have been exploited by workers using different extraction methods to extract out ITCs of significant therapeutic potential. Table 1 has summarised those plants which have undergone studies for their isothiocyanate contents, method of extraction and their pharmacological activities.

Table 1. Various isothiocyanates from plant sources with their pharmacological and biological activity

Scientific name	Part	Extraction technique	Identification technique	ITCs	Pharmacological/biological activity	Ref
Brassica oleracea var. *capitata*	Leaves	Solid Phase micro extraction	GC/MS	Allyl ITC Phenyl ITC Benzyl ITC Phenylethyl ITC	Active against *Alteria rot* in Bell Pepper	77
Eruca sativa	Seeds	Steam distillation	GC, NMR, IR	methylthiobutylITC	Skin anti-inflammatory effect	78
	Seed oil	Steam distillation HS/SPME	GC/MS	Allyl ITC 3-butenyl ITC 2-phenylethyl ITC Sulphoraphane	Activity against harmful effect of Alfatoxin	79
Brassica oleracea var. *italica*	Sprout		GC/MS	R-sulforaphane Erucin	Inhibit quoram sensing in *pseudomonas aeroginosa*	80
Rorippa nasturtium-aquaticum (L.) Hayek	Tissue	Solvent extraction with 70% ethanol	LC-MS/ UV	Phenylethyl ITC 7-methylsulfinylheptyl ITC 8-methylsulfinyloctyl ITC	Inducer of Phase-II enzymes (quinone reductase)	81
Wasabia japonica Matsum	Root	Sulfadex G-15 Gel filtration and reverse HPLC	Preparative HPLC, FAB-MS, EI-MS, IR & NMR	6-(Methylsulfinyl) hexyl ITC	Anti-inflammatory activity	82
Moringa oleifera	Pods	---	---	4-[(4'-O-acetyl-α-L-rhamnosyloxy)benzyl] ITC 4(α-L-rhamnosyloxy) benzyl ITC	Hypotensive effect	83
Raphanus sativus	---	---	---	4-(methylthio)-3-butenyl ITC	Antimutagenic activity	84
Brassica alba	Seeds	---	---	Allyl ITC	Acaricidal activity	85
Brassica oleraceae var botrytis c.v. Calabrese	Seed	RP-HPLC	GC/MS	Sulforaphane	---	86
	Stem	Hydrodistillation	GC/MS	Allyl ITC	Anti-microbial activity	87
	Stem	Hydrodistillation	GC/MS	Butane-1-ITC	Anti-microbial activity	87
	Stem	Hydrodistillation	GC/MS	2-phenylethyl ITC	Anti-microbial activity	87
Cardariadraba (1) desv	Aerial pats	Hydrodistillation	GC-FID / GC/MS	4-(methylsulfanyl)butyl ITC	Anti-microbial activity	88
Brassica junica	Seeds	SPME	---	2-phenylethyl ITC Benzyl ITC 2-butenyl ITC	Fungicidal activity	89
Brassica nigra	Seeds	SPME		2-phenylethyl ITC	Fungicidal activity	89
Degeniavelebitica	Leaves	Hydrodistillation-adsorption	GC/MS	Pentyl-4-enyl ITC	Antimicrobial activity	90
Salvadoraperseca	Roots		GC/MS	Benzyl ITC	Activity against gram negative bacteria	91
Eruca sativa	Flowers, leaves, bean	Hydrodistillation-adsorption, hydroditillation	GC, GC-MS	*E*-2-hexenal methyl ITC 3-butenyl ITC Hexyl ITC Erucin	---	92
Wasabia japonica Matsum	Whole plant	Supercritical fluid extraction	Gas chromatography	Allyl ITC	---	93
Brassica juncea	Seed	Extraction from pretreated mustard meal by distillation	---	Allyl ITC	---	94

Table 1. Continued

Scientific name	Part	Extraction technique	Identification technique	ITCs	Pharmacological/biological activity	Ref
	Seed	Solvent extraction with *n*-hexane	GC/MS	Allyl ITC Benzyl ITC Phenethyl ITC	---	95
Alliaria petiolata	Stems, leaves and flower	Hydrodistillation	GC & GC/MS	Allyl ITC	---	96
Armoracia ruticana	Root	Steam distillation	GC/MS	Allyl ITC 3-butenyl ITC 2-pentyl ITC β-phenylethyl ITC Isopropyl ITC Butyl ITC 2-pentyl ITC Phenyl ITC 3-methylthiopropyl ITC Benzyl ITC	---	98
Brassica rapa subsp. *pekinensis*	Leaves	---	GC/MS & LC/MS	3-butenyl ITC	---	99
	Midrib	---	GC/MS & LC/MS	3-butenyl ITC	---	99
*Brassica campestris*spp. pekinensis	Flower	Solvent extraction with Dichloromethane	GC/MS	4-pentenyl ITC 2-phenylethyl ITC 4-methylpentyl ITC	---	100
Table 1. Continued	Seeds	---	GC/MS	3-butenyl ITC 4-pentenyl ITC 2-phenylethyl ITC	---	101
Brassica Rapa	---	---	---	Benzyl ethyl ITC	---	102
Brassica Rapa	Seeds/ sprout	HS/SPME	GC-IT-MS	Isopropyl ITC Allyl ITC 3-butenyl ITC 3-methylbutyl ITC Pentyl ITC 4-methylpentyl ITC Hexyl ITC 3-methylbutyl ITC Pentyl ITC 4-methylpentylhexyl ITC 3-methylthiopropyl ITC Phenylethyl ITC	---	102
Leptidium sativa	Seeds	---	---	Benzyl ITC	---	103
	Roots and non-flowering aerial parts	---	---	Benzyl ITC	---	103
Tropaeolum majus	Seeds	Solvent extraction using 90% methanol	HPLC	Benzyl ITC	---	104
Moringa oleifera Lam	Seeds & leaves		^{1}H-NMR & LC-MS	4-[(4'-O-acetyl-α-L-rhamnosyloxy)benzyl] ITC 4(α-L-rhamnosyloxy) benzyl ITC (4-[(2'-O-acetyl-α-L-rhamnosyloxyl)benzyl ITC (4-[(2'-O-acetyl-α-L-rhamnosyloxyl)benzyl ITC	---	105
Raphanus sativus L.		Hydrodistillation	HPLC	Benzyl ITC	---	106
	Roots	---	GC/MS	4-methylthio-3-butenyl ITC	---	107

Table 1. Continued

Scientific name	Part	Extraction technique	Identification technique	ITCs	Pharmacological/biological activity	Ref
Drypetes roxburghii	---	---	---	---	---	108
Carica papaya	Seeds	---	GC/MS	Benzyl ITC	---	109
Sisymbrium officinale	---	---	GC/MS	Isopropyl ITC Sec-butyl ITC	---	110
Lepidium meyenii	---	---	---	Benzyl ITC p-metoxybenzyl ITC	---	111
Capparis spinosa	Leaves oil	---	^{13}C-NMR	Isopropyl ITC Butyl ITC	---	112
	Leaves	---	GC/MS	Methyl ITC Benzyl ITC	---	113
	Ripe fruit and roots	---	---	Methyl ITC Isopropyl ITC Sec-butyl ITC	---	113
Capparis Ovata	Oil	---	GC/MS	Methyl ITC	---	113
Brassica hirta	Seed	---	GC/MS & GC-FID	4-hydroxybenzyl ITC	---	92
Capsella bursa pastoris	Seed	---	GC/MS	3-Butenyl ITC	---	92
Erysimum allionnii	Seed	---	GC/MS	Erucin Sulforaphane Erucin nitrile Sulforaphane nitrile	---	92
Lesquerella fendleri	---	---	---	3-butenyl ITC	---	92
Lobularia maritima	---	---	---	3-butenyl ITC	---	92
Matthiola longipelata	---	---	---	Sulforaphene nitrile	---	92
Brassica fruticulosa	Leaves & root	SPME	GC/MS	3-butenyl ITC	---	114
Morettia phillaeana	Aerial parts	Hydrodistillation	GC/MS	4-isothiocyanato-1-Butene Isothiocyanatomethyl Benzene	---	115
Broccolini (Brassica oleraceaItalica X Alboglabra)	Seeds	---	GC/MS	3-benzyl ITC 4-methylpentyl ITC 1-isothiocyanato butane Phenylethyl ITC Sulforaphane	---	116
Iberis sempervirens	Whole plant	Hydrodistillation	GC & GC/MS	Allyl ITC Butyl ITC 3-butenyl ITC 3-butenyl ITC 3-methylthippropyl ITC 4-methylthiobutyl ITC 2-phenylethyl ITC	---	117

Isothiocyanates (ITCs), Gas chromatography-Mass spectroscopy (GC/MS), Nuclear magnetic resonance (NMR), Infrared (IR), Liquid chromatography- mass spectroscopy (LC-MS), Ultra violet (UV), High performance- liquid chromatography (HPLC), Reverse phase-HPLC (RP-HPLC), Solid phase micro extraction (SPME), Gas chromatography – flame ionization detector (GC-FID). Fast atom bombardment mass spectroscopy (FAB-MS), Electron ionization mass spectroscopy (EI-MS)

Conclusion

Isothiocyanates which could be found abundantly in cruciferous vegetables have dragged lots of attention during the past years for their ability to exert beneficial effects on human health. Numerous epidemiological studies have proved their beneficial effects. An attempt has been made by authors to distinguish minor to major aspects of isothiocyanates because some aspects have not been discussed previously and thus still need a profound consideration and study. Pharmacological activities *e.g.* anti-carcinogenic, anti-tumour and anti-inflammatory effects have been known but accurate doses of ITC have not been established by researchers. Different researchers used different doses of ITC. Some displayed their therapeutic activity in micrograms while others much more concentrations. Also, the therapeutic window and toxicity profile of ITCs have not been defined.

Therapeutic activity of ITC is also influenced by the presence of aromatic and aliphatic side chain in their structure; thus, the structure activity relationship should be further studied thoroughly.

This review has provided insights of various aspects of ITCs like chemistry, pharmacokinetics, pharmacological activities and toxicological profile. It has flashed light on the hurdles that are still to be overcome. Exploring more about chemical and physical properties of isothiocyanates will contribute a great part in their isolation process. The present review has also provided safety and toxicity profile of ITCs and the mechanisms of their pharmacological activities for the probable production of more potent and safer drugs and nutraceuticals.

Author contributions

Chandra Kala: concept and manuscript writing; Syed Salman Ali: manuscript writing; Nabeel Ahmad: table drafting; Sadaf Jamal Gilani: editing manuscript; Najam Ali Khan: making closed access article available and supervision.

Declaration of interest

The authors declare that there is no conflict of interest. The authors alone are responsible for the content of the paper.

References

[1] Satyavan S. Sulphur reports: isothiocyanates in heterocyclic synthesis. Alexender S, Ed. Harwood: Academic Publishers GmBH United Kingdom, 1989.

[2] Wu X, Zhou QH, Xu K. Are isothiocyanates potential anti-cancer drugs? *Acta Pharmacol Sin*. 2009; 30(5): 501-512.

[3] Hecht SS. Chemoprotection by isothiocyanates. In: Kelloff GJ, Hawk ET, Sigman CC, Eds. Promising cancer chemopreventive agents. Totowa: Humana Press, 2004.

[4] Redovnikovic IR, Glivetic T, Delonga K, Furac JV. Glucosinolates and their potential role in plant. *Period Biol*. 2008; 110(4): 297-309.

[5] Food and Health Innovation Service. The Brassicas-an undervalued nutritional and health beneficial plant family. Accessed 2012]. Available from: http://www.foodhealthinnovation.com/media/5567/brassicas_report_.pdf.

[6] Fahey JW, Zalcmann AT, Talalay P. The chemical diversity and distribution of glucosinolates and isothiocyanates among plants. *Phytochemistry*. 2001; 56(1): 5-51.

[7] Linus pauling Institute, Oregan University. *Isothiocyantes. Micronutrient information centre*. [Accessed 2017]. Available from http://lpi.oregonstate.edu/mic/dietary-factors/phytochemicals/isothiocyanates.

[8] Fimognari C, Turrini E, Ferruzzi L, Lenzi M, Hrelia P. Natural isothiocyanates: genotoxic potential versus chemoprotection. *Mutat Res*. 2012; 750(2): 107-131.

[9] Jiao D, Eklind KL, Chsoi CI, Desai DH, Amin SG, Chung FL. Structure-activity relationships of isothiocyanates as mechanism-based inhibitors of 4-(methy initrosamino) -1-(3-pyridy1)-1-butanone-induced lung tumorigenesis in A/J Mice1. *Cancer Res*. 1994; 54(16): 4327-4333.

[10] Rahman AU. Studies in natural products chemistry: bioactive natural products (Part F). 1st ed. Amsterdam: Elsevier Science Ltd, 2001.

[11] Dufour V, Stahl M, Baysse C. The antibacterial properties of isothiocyanates. *Microbiol*. 2015; 161(2): 229-243.

[12] Tichy M. Quantitative structure-activity relationships**.** Proceedings of the chemical structure-biological activity relationships conference. 1973 June 27-29; Prague, Czechoslovakia.

[13] Johnson IT. Glucosinolates in human diet. Bioavailability and implication for health. *Phytochem Rev*. 2002; 1(2):183-188.

[14] Zhang Y. Allylisothiocyanate as cancer chemoprotective phytochemical. *Mol Nutr Food Res*. 2011; 57(1): 127-135.

[15] Elfoul L, Rabot S, Khelifa N, Quinsac A, Duguay A, Rimbault A. Formation of AITC from sinigrin in digestive tract of ratsmonoassociated with the human colonic strain bacteroids thetaiotaomicron. *FEMS Microbiol Lett*. 2001; 197(1): 99-103.

[16] Petri N, Tannergren C, Holst B, Mellon FA, Bao Y, Plumb GW, Bacon J, O'Leary KA, Kroon PA, Knutson L, Forsell P, Eriksson T, Lennernas H, Williamson G. Absorption/metabolism of sulforaphane and quercitin and regulation of phase II enzymes in human jejunum *in vivo*. *Drug metab Dispos*. 2003; 31(6): 805-813.

[17] Ye L, Dinkova-Kostova AT, Wade KL, Zhang Y, Shapiro TA, Talalay P. Quantitative determination of dithiocarbamates in human plasma serum,

erythrocytes and urine. pharmacokinetics of broclli sprout ITC in human. *Clin Chim Acta*. 2002; 321(1-2): 127-129.

[18] Bricker GV, Riedl KM, Ralston RA, Tober KL, Oberyszyn TM, Schwartz SJ. ITC metabolism, distribution, interconversion in mice following consumption of thermally processed broccoli sprouts or purified sulphoraphane. *Mol Nutr Food Res*. 2014; 58(70): 1991-2000.

[19] Clarke JD, Hsu A, Williams DE, Dashwood RH, Stevens JF, Yamamoto M, Ho E. Metabolism and tissue distribution of sulforaphane in nrf-2 knckout and wild type mice. *Pharm Res*. 2011; 28(12): 3171-3179.

[20] Zhang Y. The molecular basis that unifies metabolism and cellular uptake and chenmoprevention activites of dietary ITC. *Carcinogenesis*. 2012; 33(1): 2-9.

[21] Telang U, Ji Y, Morris ME. ABC transporters and ITC: potential for pharmacokinetic diet-drug interaction. *Biopharm Drug Dispos*. 2009; 30(7): 335-344.

[22] Kolm RH, Danielson H, Zhang Y, Talalay P, Mannervik B. Isothiocyanaytes as substrate for human glutathione transferase: structure activity studies. *Biochem J*. 1995; 311(2): 453-459.

[23] Gasper AV, Al-Janobi A, Smith JA, Bacon JR, Fortun P, Atherton C, Taylor MA, Hawkey CJ, Barrett DA, Mithen FZ. Glutathione S-transferase M1 polymorphism and metabolism of sulforaphane from standard and high-glucosinolate broccoli. *Am J Clin Nutr*. 2005; 82(6): 1283-1291.

[24] Steck SE, Gammon MD, Hebert JR, Wall DE, Zeisel SH. GSTM1, GSTT1, GSTP1 and GSTA1 polymorphisms in urinary ITC metabolite following broccoli consumption in humans. *J Nutr*. 2007; 137(4): 904-909.

[25] Lee Ms. Enzyme induction and comparative oxidation desulfuration of isothiocyanates to isocyantaes. *Chem Res Toxicol*. 1996; 9(7): 1072-1078.

[26] Yoshigae Y, Sridar C, Kent VM, Hollenberg PF. The inactivation of human CYP2E1 by PEITC, a naturally occurring chemoprotective agaents and its oxidative bioactivation. *Drug Metab Dispos*. 2013; 41(4): 858-869.

[27] Vermeulen M, Katelaars IWAAK, Berg RVD, Vaes WHJ. Biovavailbility and kinectics of sulforaphane in human after consumption of cooked versus raw broccoli. *J Agric Food Chem*. 2008; 56(22): 10505-10509.

[28] Mennicke WH, Gorler K, Krumbiegel G, Lorenz D, Rittmann N. Studies on the metabolism and excretion of benzyl isothiocyanates in man. *Xenobiotica*. 1988; 18(4): 441-447.

[29] Bollard M, Stribbling S, Mitchell S, Caldwell J. The disposition of AITC in rat and mouse. *Food Chem Toxicol*. 1997; 35(10-11): 933-943.

[30] Borghoff J. Age related changes in metabolism and excretion of allyl isothiocyanate. A model compound for glutathione conjugation. *Drug Metab Dispos*. 1986; 14(4): 417-422.

[31] Seow A, Shi CY, Chung FL, Jiao D, Hankin JH, Lee HP, Coetzee GA, Yu MC. Urinary total isothiocyanate (ITC) in a population-based sample of middle-aged and older Chinese in Singapore: relationship with dietary total ITC and glutathione S-transferase M1/T1/P1 genotypes. *Cancer Epidemiol Biomarkers Prev*. 1988; 7(9): 775-781.

[32] Gianni ED, Turrini E, Milelli A, Maffei F, Carini M, Minarini A, Tumiatti V, Ros TD, Prato M, Fimognari C. Study of the cytotoxic effects of the new synthetic isothiocyanate CM9 and its fullerene derivative on human t-leukemia cells. *Toxins*. 2015; 7(2): 535-552.

[33] Wang D, Upadhyaya B, Liu Y, Knudsen D, Dey M. Phenethyl isothiocyanate upregulates death receptors 4 and 5 and inhibits proliferation in human cancer stem-like cells. *BMC Cancer*. 2014; Article ID 4148558.

[34] Qazi A, Pal J, Maitah M, Fulciniti M, Pelluru D, Nanjappa P, Lee S, Batchu RB, Prasad M, Bryant CS, Rajput S, Gryaznov S, Beer DG, Weaver DW, Munshi NC, Goyal RK, Shammas MA. Anticancer activity of a broccoli derivative, sulforaphane, in barrett adenocarcinoma. Potential use in chemoprevention and as adjuvant in chemotherapy. *Transl Oncol*. 2010; 3(6): 389-399.

[35] Devi JR, Thangam EB. Mechanisms of anticancer activity of sulforaphane from *Brassica oleracea* in HEp-2 human epithelial

carcinoma cell line. *Asian Pac J Cancer Prev*. 2012; 13(5): 2095-2100.

[36] Li Y, Karagoz GE, Seo YH, Zhang T, Jiang Y, Yu Y, Duarte AM, Schwartz SJ, Boelens R, Carroll K, Rüdiger SG, Sun D. Sulforaphane inhibits pancreatic cancer through disrupting Hsp90– p50 Cdc37 complex and direct interactions with amino acids residues of Hsp90. *J Nutr Biochem*. 2012; 23(12): 1617-1626.

[37] Chaudhuri D, Orsulic S, Ashok BT. Antiproliferative activity of sulforaphane in Akt-overexpressing ovarian cancer cells. *Mol Cancer Ther*. 2006; 6(1)*:* 334-345.

[38] Farag MA, Motaal AAA. Sulforaphane composition, cytotoxic and antioxidant activity of crucifer vegetables. *J Adv Res*. 2010; 1(1): 65-70.

[39] Geng F, Tang L, Li Y, Yang L, Choi KS, Kazim AL, Zhang Y. Allyl isothiocyanate arrests cancer cells in mitosis, and mitotic arrest in turn leads to apoptosis via Bcl-2 protein phosphorylation. *J Biol Chem*. 2011; 286(37): 32259-32267.

[40] Bhattacharya A, Li Y, Wade KL, Paonessa JD, Fahey JW, Zhang Y. Allyl isothiocyanate-rich mustard seed powder inhibits bladder cancer growth and muscle invasion. *Carcinogenesis*. 2010; 31(12): 2105-2110.

[41] Tsai SC, Huang WW, Huang WC, Lu CC, Chiang JH, Peng SF, Chung JG, Lin YH, Hsu YM, Amagaya S, Yang JS. ERK-modulated intrinsic signaling and G(2)/M phase arrest contribute to the induction of apoptotic death by allyl isothiocyanate in MDA-MB-468 human breast adenocarcinoma cells. *Int J Oncol*. 2012; 41(6): 2065-2072.

[42] Tripathi K, Hussein UK, Anupalli R, Barnett R, Bachaboina L, Scalici J, Rocconi RP, Owen LB, Piazza GA, Palle K. Allyl isothiocyanate induces replication-associated DNA damage response in NSCLC cells and sensitizes to ionizing radiation. *Oncotargets*. 2015; 6(7): 5237-5252.

[43] Zhang Y, Tang L. Discovery and development of sulforaphane as a cancer chemopreventive phytochemical. *Acta Pharmacol Sin*. 2007; 28(9): 1343-1354.

[44] Taylor M. Handbook of natural antimicrobials for food safety and quality, 1st ed. Cambridge: Woodhead Publishing, 2014.

[45] Lin CM, Preston JF, Wei CI. Antibacterial mechanism of allyl isothiocyanate. *J Food Prot*. 2000; 63(6): 727-734.

[46] Kim HY, Phan-a-god S, Shin IS. Antibacterial activities of isothiocyanates extracted from horseradish (*Armoracia rusticana*) root against antibiotic-resistant bacteria. *Food Sci Biotechnol*. 2015; 24(3): 1029-1034.

[47] Padla EP, Solis LT, Levida RM, Shen CC, Ragasa CY. Antimicrobial isothiocyanates from the seeds of *Moringa oleissfera* Lam. *Z Naturforsch C*. 2012; 67(11-12): 557-564.

[48] Dufour V, Stahl M, Baysse C. The antibacterial properties of isothiocyanates. *Microbiol*. 2015; 161(2): 229-243.

[49] Dufour V, Alazzam B, Ermel G, Thepaut M, Rossero A, Tresse O, Baysse C. Antimicrobial activities of isothiocyanates against *Campylobacter jejuni* isolates. *Front Cell Infect Microbiol*. 2012; Article ID 3417524.

[50] Dias C, Aires A, Saavedra MJ. Antimicrobial activity of isothiocyanates from cruciferous plants against methicillin-resistant *Staphylococcus aureus* (MRSA). *Int J Mol Sci*. 2014; 15(11): 19552-19561.

[51] Aires A, Mota VR, Saavedra MJ, Rosa EAS, Bennett RN. The antimicrobial effects of glucosinolates and their respective enzymatic hydrolysis products on bacteria isolated from the human intestinal tract. *J Appl Microbiol*. 2009; 106(6): 2086-2095.

[52] Tarozzi A, Angeloni C, Malaguti M, Morroni F, Hrelia S, Hrelia P. Sulforaphane as a potential protective phytochemical against neurodegenerative diseases. *Oxid Med Cell Longe*. 2013; Article ID 415078.

[53] Benedict AL, Mountney A, Hurtado A, Bryan KE, Schnaar RL, Dinkova-Kostova AT, Talalay P. Neuroprotective effects of sulforaphane after contusive spinal cord injury. *J Neurotrauma*. 2012; 29(16): 2576-2578.

[54] Tarozzi A, Morroni F, Bolondi C, Sita G, Hrelia P, Djemil A, Cantelliforti G. Neuroprotective effects of erucin against 6-hydroxydopamine-induced oxidative damage in a dopaminergic-like neuroblastoma cell line. *Int J Mol Sci*. 2012; 13(9): 10899-10910.

[55] Wellejus A, Elbrond-Bek H, Kelly NM, Weidner MS, Jorgensen SH. 4-iodophenyl isothiocyanate: a neuroprotective compound. *Restor Neurol Neurosci*. 2012; 30(1): 21-38.

[56] Tarozzi A, Angeloni C, Malaguti M, Morroni F, Hrelia S, Hrelia P. Sulforaphane as a potential protective phytochemical against neurodegenerative diseases**.** *Oxid Med Cell Longev*. 2013; Article ID 415078.

[57] Kelsey N, Wilkins HM, Linseman DA. Nutraceutical antioxidants as novel neuroprotective agents. *Molecules*. 2010; 15(11): 7792-7814.

[58] Morroni F, Tarozzi A, Sita G, Bolondi C, Manuel J, Moraga Z, Cantelliforti G, Hrelia P. Neuroprotective effect of sulforaphane in 6 hydroxydopamine-lesioned mouse model of Parkinson's disease. *Neurotoxicology.* 2013; 36: 63-71.

[59] Angeloni C, Malaguti M, Rizzo B, Barbalace MC, Fabbri D, Hrelia S. Neuroprotective effect of sulforaphane against methylglyoxal cytotoxicity. *Chem Res Toxicol*. 2015; 28(6): 1234-1245.

[60] Chittezhath M, Kuttan G. Radioprotective activity of naturally occurring organosulfur compounds. *Tumori*. 2006; 92(2): 163-169.

[61] Soni AK, Samarth RM, Kumar M, Shukla S, Meena PD, Kumar M, Kumar A. Modulation of radiation induced alterations in Swiss albino mice by *Brassica compestris* (var sarason). *Pharmacologyonline.* 2006; 2: 190-199.

[62] Karami M, Nosrati A, Naderi M, Makhloogh M, Shahani S. Protective effects of *Nasturtium officinale* against gamma-irradiation-induced hepatotoxicity in C57 mice. *Res J Pharmacogn*. 2015; 2(2): 19-25.

[63] Bai Y, Wang X, Zhao S, Ma C, Cui J, Zheng Y. Sulforaphane protects against cardiovascular disease via Nrf2 activation. *Oxid Med Cell Longe*. 2015; Article ID 407580.

[64] Owis AI. Broccoli; the green beauty: a review. *J Pharm Sci Res*. 2015; 7(9): 696-703.

[65] Testai L, Marino A, Tomita K, Citi V, Martelli L, Breschi MC, Levi R, Calderone V. Carboxyphenyl isothiocyanate (4-CPI), a novel hydrogen sulfide-donor endowed with cardioprotective property. *Nitric Oxide*. 2015; 47(S): 14-60.

[66] Leoncini E, Malaguti M, Angeloni C, Motori E, Fabbri D, Hrelia S. Cruciferous vegetable phytochemical sulforaphane affects phase II enzyme expression and activity in rat cardiomyocytes through modulation of Akt signaling pathway. *J Food Sci*. 2011; 76(7): 175-181.

[67] Luo J. Investigating the potential anti-diabetic effect of sulforaphane. M.Sc. thesis. Faculty of the Virginia Polytechnic Institute and State University, Blacksburg, Virginia, 2014.

[68] Jayakumar T, Chen WF, Lu WJ, Chou DS, Hsiao G, Hsu CY, Sheu JR, Hsieh CY. A novel antithrombotic effect of sulforaphane via activation of platelet adenylate cyclase: *ex vivo* and in *vivo studies*. *J Nutr Biochem*. 2013; 24(6): 1086-1095.

[69] Oh CH, Shin JI, Mo SJ, Yun SJ, Kim SH, Rhee YH. Antiplatelet activity of L-sulforaphane by regulation of platelet activation factors, glycoprotein IIb/IIIa and thromboxane A2. *Blood Coagul Fibrinolysis*. 2013; 24(5): 498-504.

[70] Lee DS, Kim TH, Jung YS. Inhibitory effect of allyl isothiocyanate on platelet aggregation. *J Agric Food Chem*. 2014; 62(29): 7131-7139.

[71] Masutomi N, Toyoda K, Shibutani M, Niho N, Uneyama C, Takahashi N, Hirose M. Toxic effects of BITC and AITC and benzyl ioform specific metabolites in urinary bladder after a single intravesical application to rats. *Toxicol Pathol*. 2001; 26(2): 617-622.

[72] Bathaie SZ, Tamanoi F. Mechanism of the anticancer effect of phytochemicals. 1st ed. Waltham: Academic Press, 2015.

[73] Lewerenz HJ. Subacute oral toxicity study of BITC in rats. *Food.* 1992; 36(2): 190-198.

[74] Health Canada Pest Management Regulatory Agency. Oriental mustard seed (OMS). Ottawa: Pest Management Regulatory Agency, 2011.

[75] Fofaria NM, Ranjan A, Kim S, Srivastava SK. Mechanism of anticancer effects of isothiocyanates. *Enzymes.* 2015; 37:111-137.

[76] Gupta P. Molecular targets of ITC in cancer: Recent Advances. *Mol Nutr Food Res.* 2014; 58(8): 1685-1707.

[77] Troncoso R, Espinoza C, Estrada

AS, Tiznado ME, García HS. Analysis of the isothiocyanates present in cabbage leaves extract and their potential application to control *Alternaria rot* in bell peppers. *Food Res Int.* 2005; 38(6): 701-708.

[78] Yehuda H, Khatib S, Sussan I, Musa R, Vaya J, Tamir S. Potential skin anti-inflammatory effects of 4-methylthiobutylisothiocyanate (MTBI) isolated from rocket (*Eruca sativa*) seeds. *Biofactors*. 2009; 35(3): 295-305.

[79] Hanafi EM, Hegazy EM, Riad RM. Bio-protective effect of *Eruca sativa* seed oil against the hazardus effect of aflatoxin b1 in male-rabbits. *Int J Acad Res*. 2010; 2(2): 67-74.

[80] Ganin H, Rayo J, Amara N, Levy N, Krief P, Meijler M. Sulforaphane and erucin, natural isothiocyanates from broccoli, inhibit bacterial quorum sensing. *Med Chem Commun*. 2013; 4: 175-179.

[81] Rose P, Faulkner K, Williamson G, Mithen R. 7-Methylsulfinylheptyl and 8-methylsulfinyloctyl isothiocyanates from watercress are potent inducers of phase II enzymes. *Carcinogenesis.* 2000; 21(11): 1983-1988.

[82] Uto T, Hou DX, Morinaga O, Shoyama Y. Molecular mechanisms underlying anti-inflammatory actions of 6-(methylsulfinyl)hexyl isothiocyanate derived from Wasabi (*Wasabia japonica*). *Adv Pharmacol Sci*. 2012; Article ID 614046.

[83] Faizi S, Siddiqui BS, Saleem F, Gilani AH. Hypotensive constituents from the pods of *Moringa oleifera*. *Planta Med*. 1998; 64(3): 225-228.

[84] Nakamura Y, Iwahashi T, Tanaka A, Koutani J, Matsuo T, Okamoto S, Sato K, Ohtsuki K. 4-(methylthio)-3-butenyl isothiocyanate, a principal antimutagen in daikon (*Raphanus sativus*; Japanese white radish). *J Agric Food Chem*. 2001; 49(12): 5755-5760.

[85] Lim JH, Kim HW, Jeon JH, Lee HS. Acaricidal constituents isolated from *Sinapis alba* L. seeds and structure-activity relationships. *J Agric Food Chem.* 2008; 56(21): 9962-9966.

[86] Naser SA, Besheli BA, Mehr SS. The isolation and determination of sulforaphane from broccoli tissues by reverse phase-high performance liquid chromatography. *J Chin Chem Soc*. 2011; 58(7): 906-910.

[87] Hifnawy MS, Salam RMS, Rabeh MA, Aboseada MA. Glucosinolates, glycosidically bound volatiles and antimicrobial activity of *Brassica oleraceae* Var. *botrytis*, (Soultany cultivar). *J Biol Agric Healthcare*. 2013; 3(17): 66-80.

[88] Radonic A, Blazevic I, Mastelic J, Zekic M, Skocibusic M, Maravic A. Phytochemical analysis and antimicrobial activity of *Cardaria draba* (L.) Desv. volatiles. *Chem Biodivers.* 2011; 8(6): 1170-1181.

[89] Olivier C, Vaughn SF, Mizubuti ESG, Loria R. Variation in allyl isothiocyanate production within *Brassica* species and correlation with fungicidal activity. *J Chem Eco*. 1999; 25(12): 2687-2701.

[90] Mastelic J, Blazevic I, Kosalec I. Chemical composition and antimicrobial activity of volatiles from *Degenia velebitica*, a European stenoendemic plant of the Brassicaceae family. *Chem Biodivers*. 2010; 7(11): 2755-2765.

[91] Sofrata A, Santangelo EM, Azeem M, Borg-karlson AK, Gustafsson A, Putsep K. Benzyl isothiocyanates, a major compound from roots of *Salvadora persica* is highly active against gram positive bacteria. *Plos One*. 2011; 6(8): 1-10

[92] Mastelic J, Jerkovic I, Blazevi I, Radoni A, Krstulovi L. Hydrodistillation–adsorption method for the isolation of water-soluble, non-soluble and high volatile compounds from plant materials. *Talanta*. 2008; 76(4): 885-891.

[93] Li L, Lee W, Lee WJ, Auh JH, Kim SS, Yoon J. Extraction of allyl isothiocyanate from wasabi (*Wasabia japonica* Matsum) using supercritical carbon dioxide. *Food Sci Biotechnol*. 2010; 19(2): 405-410.

[94] Sharma HK, Ingle S, Singh C, Sarkar BC, Upadhyay A. Effect of various process treatment conditions on the allyl isothiocyanate extraction rate from mustard meal. *J Food Sci Technol.* 2012; 49(3): 368-372.

[95] Marton MR, Lavric V. A simple method for the quantification of isothiocyanates from mustard. *UPB Sci Bull Series B.* 2013; 75(1): 63-72.

[96] Ivica B, Josip M. Free and bound volatiles of garlic mustard (*Alliaria petiolata*). *Croat Chem Acta*. 2008; 81(4): 607-613.
[97] Vaughn SF, Berhow MA. Allelochemicals isolated from tissues of the invasive weed garlic mustard (*Alliaria petiolata*). *J Chem Ecol*. 1999; 25(11): 2495-2504.
[98] Jiang ZT, Li R, Yu JC. Pungent components from thioglucosides in *Armoracia rusticana* grown in China, obtained by enzymatic hydrolysis. *Food Technol Biotechnol*. 2006; 44(1): 41-45.
[99] Kim YK, Kin GH. Determination of 3-butenyl isothiocyanate in different part and cultivators of Chinese cabbage. *Food Sci Biotech*. 2005; 14(4): 466-469.
[100] Hong E, Kim GH. GC-MS analysis of extracts from Korean cabbage. (*Brassica campeteris* L. ssp *pekinensis*) and its seed. *Prev Nutr Food Sci*. 2013; 18(3): 218-221.
[101] Jones G, Sander OG, Grimm C. Aromatic compounds in three varieties of turnip greens harvested at three maturity leaves. *J Food Quality*. 2007; 30(2): 218-227.
[102] Taveira M, Fernades F, Pinho PGD, Andrade PB, Pereira JA, Valentao P. Evaluation of *Brassica rapa* var *rapa* L. volatile composition by HS-SPME and GC/IT-MS. *Microchem J*. 2009; 93(2): 140-146.
[103] Sharma S, Agrawal N. Nourishing and healing power of garden cress. *Indian J Nat Prod Resour*. 2011; 2(3): 292-297.
[104] Lykkesfeldt J, Moller BL. Synthesis of benzylglucosinolate in *Tropaeolum majus* L. *Plant Physiol*. 1993; 102(2): 609-613.
[105] Goyal BR, Agrawal BB, Goyal RK, Mehta AA. Phytopharmacology of *Moringa oleifera* Lam. an overview. *Nat Prod Rad*. 2001; 6(4): 347-353.
[106] Santhi S, Shanthipriya S. High-performance liquid chromatography in the study of benzylisothiocyante from *Raphanus sativus* (white radish). *SIRJ-APBBP*. 2014; 1(4): 28-33.
[107] Li J, Xie B, Yan S, Li H, Wang Q. Extraction and determination of 4-methylthio-3-butenyl isothiocyanate in Chinese radish (*Raphanus sativus L.*) roots. LWT *Food Sci Technol*. 2015; 60(2): 1080-1087.
[108] Puntambekar SV. Mustard oil and mustard oil glycoside occuring in seed kernels of Putranjiva roxburghii. Banglore: Indian Academy of Sciences, 1950.
[109] Li W, Du Y, Zhang Y, Chi Y, Shi Z, Chen W, Ruan M, Zhu H. Optimized formation of benzyl isothiocyanate by endogenous enzyme and its extraction from *Carica Papaya* seed. *Trop J Pharm Sci*. 2014; 13(8): 1303-1311.
[110] Biro-Sandor Z. Assessment report on *Sisymbrium officinale* (L.) Scop. herba. London: European Medicine Agency, Committee on herbal medicinal product, 2014.
[111] Zheng BL, Kim CH, Wolthoff S, He K, Rogers L, Shao Y, Zheng QI, inventors; Pure World Botanicals, Inc, assignee. Extract of *Lepidium meyenii* roots for pharmaceutical applications. United States Patent US 6267995 B1. 2001 July 31.
[112] Rajesh P, Selvamani P, Latha S, Saraswathy A, Kannan VR. A review on chemical and medicobiological applications of capparidaceae family. *Pharmacog Rev*. 2009; 3(6): 378-387.
[113] Muhaidat R, Al-Qudah MA, Al-Shayeb A, Jacob JH, Al-Jaber H, Hussein E, Al Tarawneh IN, Orabi STA. Chemical profile and antibaceterial activity of crude fractions and essential oils of *Capparis ovata* Desf. and *Capparis spinosa* L. (capparaceae). *Int J Integr Biol*. 2013; 14(1): 39-47.
[114] Tripodia G, Verzeraa A, Dimaa G, Condursoa C, Ragusab S. *Brassica fruticulosa* Cyr. and *Brassica incana* Ten. (Brassicaceae) as Mediterranean traditional wild vegetables: a valuable source of bioactive compounds. *J Essent Oil Res*. 2012; 24(6): 539-545.
[115] El-Egami AA, Burham BO, El-Kamali HH. Essential oil composition of the flowering aerial parts of Sudanese *Morettia phillaeana* (Del.) DC. *Curr Res J Biol Sci*. 2011; 3(2): 100-103.
[116] Zhang B, Wang X, Yang Y, Zhang X. Extraction and identification of isothiocyanates from broccolini seeds. *Nat Prod Commun*. 2011; 6(1): 65-66.
[117] Blazevic I. Glucosinolates: novel sources and biological potential. *Austin J Bioorg Org Chem*. 2014; 1(1): 4.

Abbreviations

4-CPI: 4-carboxyphenyl isothiocyanate; 4-IPITC:

4-iodophenyl isothiocyanate; ABC: ATP-binding cassette; AITC: allyl isothiocyanate; ALP: alkaline phosphatase; ARE: activated Nrf2-antioxidant response; ATM: ataxia telangiectasia mutated protein kinase; ATR: ataxia telangiectasia and Rad3 related protein kinase; Bax: Bcl associated X protein; Bcl-2: B-cell lymphoma-; BCRP: breast cancer resistance protein; BEAC: barrett oesophageal adenocarcinoma; BITC: benzyl isothiocyanate; BITC: benzyl isothiocyanate; CDK: cyclin-dependent kinases; CVD: cardiovascular diseases; DA: dopaminergic; DR4: death receptor 4 ; DR5: death receptor 5; EAE: experimental autoimmune encephalomyelitis; EI-MS: electron ionization mass spectroscopy; ERK: extracellular signal–regulated kinase; FAB-MS: fast atom bombardment mass spectroscopy; FANCD2: Fanconi anemia complementation group proteins D; GC/MS: gas chromatography-Mass spectroscopy; GC-FID gas chromatography - flame ionization detector; GGT: γ-glutamyl transpeptidase; GPT: glutamate pyruvate transaminase; GSH: glutathione; GST: glutathione S-transferas; HDAC: histone deacetylases; HPLC: high performance- liquid chromatograph; HSP: heat shock protein; IR: infrared; ITC: isothiocyanate; Keap 1: Kelch-like ECH-associated protein 1; LC-MS: liquid chromatography- mass spectroscopy; MAPK: mitogen-activated protein kinases; MBC: minimum bactericidal concentration; MMP: matrix MetalloProteinas; MMP: matrix metalloproteinases; MPP+: methylphenylpyridinium; MPTP: 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridin; MRAB: multidrug-resistant *Acinetobacter baumanii*; MRP: multi drug resistance protein; MRPA: multidrug-resistant *Pseudomonas aeruginosa*; MRSA: methicillin-resistant *Staphylococcus aureus*; NDI: naphthalenetetracarboxylic diimide; NMR: nuclear magnetic resonanc; Nrf2: nuclear factor (erythroid- derived2)-like 2; Nrf2: nuclear factor-E2-related factor 2; NSCLC: non-small cell lung cancer; OMS: oriental mustard see; PD: Parkinson's disease; PEITC: phenyl ethyl isothiocyanate; P-gp: P-glycoprotein; PIC: phenethyl isocyanates; PKC: protein kinase C; PLC: phospholipase ; QR: quinone reductase ; RP-HPLC: reverse phase-HPLC ; RT-PCR: reverse transcription polymerase chain reaction; SFN: Sulphoraphane; SPME: solid phase micro extraction ; STZ: streptozotocin; SULT: sulphonyltransferase; TRAIL: TNF-related apoptosis-inducing ligand; UGT: UDP-glucoronosyltransferase; UV: ultra violet; VEGF: vascular endothelial growth factor; VRSA: vancomycin-resistant *S. aureus*; XTT: 2,3-bis(2-methoxy-4-nitro-5-sulfophenyl)-2H-tetrazolium-5 carboxanilide inner salt