

We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,900

Open access books available

186,000

International authors and editors

200M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities



WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com



Clinical Applications of Pomegranate

Sally Elnawasany

Additional information is available at the end of the chapter

<http://dx.doi.org/10.5772/intechopen.75962>

Abstract

Pomegranate, *Punica granatum* L., is an ancient, unique fruit borne on a small, long-living tree in the Mediterranean region, Southeast Asia, and tropical Africa. Pomegranate was mentioned in ancient times in the Old Bible, the Jewish Torah, and mentioned three times in the holy Quran where it was described as one of the paradise fruits. In ayurvedic medicine, pomegranate is used in the treatment of parasitic infection, diarrhea, and ulcers. Recently, pomegranate has been studied in several systems of medicine for its pharmacological actions: anti-inflammatory, antioxidant, and anticarcinogenic. The aim of the chapter is to summarize pomegranate efficacy in many preclinical and clinical studies.

Keywords: pomegranate, ayurvedic medicine, pharmacological activities, preclinical, clinical studies

1. Introduction

Pomegranate, (*Punica granatum* L.), a paradise fruit, has a great value throughout history. It had been mentioned in Judaism, Christianity, and Islamic religions [1]. From ancient times, pomegranate was used in treatment of diarrhea [2], parasitic infections [3], and diabetes mellitus [4]. Greco-Arab and Islamic medicine prescribed pomegranate for sore throat, inflammation, and rheumatism [5]. Various pomegranate activities (anti-inflammatory, antioxidant, and anticancer) encouraged growing number of studies to apply it in solving multiple medical problems [6]. Pomegranate plant is a small tree (**Figure 1**) that is cultivated in the Middle East, Mediterranean region, China, India, California, and Mexico. The fruit (**Figure 2**) is composed of many parts such as seeds, peels (pericarp), pulp, and juice [6].



Figure 1. Pomegranate tree.



Figure 2. Pomegranate fruit.

2. Clinical applications

For its multiple pharmacological potential, pomegranate has been investigated by variable preclinical and clinical studies in a wide variety of health disorders:

2.1. Inflammation

Pomegranate exhibits a potent anti-inflammatory effect through inhibition of cyclooxygenase (COX) and lipoxygenase (important inflammatory mediators) [7].

2.1.1. Gastrointestinal inflammation

2.1.1.1. Gastric inflammation

2.1.1.1.1. Preclinical studies

Helicobacter pylori (*H. pylori*) are major etiological agents in peptic ulcer. Pomegranate methanol extract produced a remarkable anti-*H. pylori* activity with mean diameter of inhibition zone 39 at 100 µg disc⁻¹ [8]. This activity is explained by altering bacterial cell surface hydrophobicity and prevention of bacterial adhesion to gastric mucosa [9]. Moreover, pomegranate revealed gastroprotective potential via antioxidant mechanism in aspirin- and ethanol-induced gastric ulceration in animal models [10]. Gastroprotective property of pomegranate is attributed to its constituents (saponin, tannins, and flavonoids) as demonstrated in another study on wistar rats where oral administration (490 and 980 mg/kg body weight) of pomegranate aqueous methanolic extract significantly reduced gastric ulcer index in alcohol-, indomethacin-, and aspirin-induced ulcers [11]. Tannins are high molecular weight phenolic compounds present in many plants, including pomegranate fruit pericarp (peels). These compounds have the capacity to form complexes mainly with proteins [12, 13]. Pomegranate tannins form a protective layer (tannin-protein/tannin-polysaccharide complex), upon damaged epithelial tissues, thus allowing the healing process below to occur naturally through prevention of bleeding and acceleration of ulcer healing [14, 15]. All parts of the pomegranate tree have been used as a source for tannin in the leather industry, changing animal hide into leather. About 10–25% of tannin are present in the trunk bark and were important in leather production in Morocco. In this process, collagen chains in the hide are cross-linked by tannin to give leather. The formation of various complex bonds helps the tannin-protein polymer combination [16, 17]. These facts take our attention to the Islamic advice “To eat pomegranate with its pericarp as it is tanning for the stomach”.

2.1.1.2. Intestinal inflammation

2.1.1.2.1. Preclinical studies

Inflammatory response is induced by transduction cascades initiated by many inflammatory mediators, that is, tumor necrosis factor α (TNF- α) and nuclear factor κ B (NF- κ B). Pomegranate inhibited TNF- α -induced NF- κ B activation and COX-2 expression in colon cell line. This effect was highly presented by pomegranate juice compared to single constituents, that is, tannin and punicalagin. This highlights the synergism between all bioactive pomegranate compounds [18]. Prebiotics are food agents that stimulate the growth or activity of beneficial microorganisms. Pomegranate peel extract (6 mg/d for 4 weeks) increased the cecal pool of beneficial bifidobacteria when given to high-fat diet mice. Additionally, it counteracted the high-fat-induced expression of inflammatory markers both in the colon and in the visceral adipose tissue [19]. Through its antioxidative action, pomegranate elagic acid (EA) (10 mg/kg) in colonic-delivering microsphere significantly ameliorated the severity of colonic lesions and reduced myeloperoxidase (MPO) activity and lipid peroxidation. This effect was obtained by orally administering it to rat model of dextran sulfate sodium (DSS)-induced ulcerative colitis [20]. Mast cells are

important inflammatory cells that release histamine. Mast cell stabilizing is an additional anti-inflammatory mechanism of pomegranate where its hydroalcoholic extract significantly lowered DSS-induced elevated histamine level in mice colon tissue [21].

2.1.1.2.2. Clinical studies

The only human trial is the ongoing phase I study on the role of pomegranate juice ellagitannins in the modulation of inflammation in inflammatory bowel diseases. This has been registered since December 2016. Available online: <http://www.clinicaltrials.gov>

2.1.2. Joint inflammation

2.1.2.1. Preclinical studies

A pomegranate compound, delphinidin, attenuated the inflammatory signaling that results in rheumatoid arthritis. This mechanism was mediated by inhibition of the histone acetyl transferase and NF- κ B activation in human rheumatoid arthritis synovial cell line [22]. Pomegranate alleviated features of arthritis in collagen-induced arthritic mice (CIA). This effect was associated with histopathological evidence of reduced inflammatory cells and joint tissue damage. Moreover, pomegranate decreased the interleukin 6 (IL-6) level and suppressed inflammatory signal transduction pathways in mouse macrophages [23].

2.1.2.2. Clinical studies

Pomegranate (2 capsules of 250 mg pomegranate extract/day for 8 weeks) improved disease activity, some inflammatory blood biomarkers and oxidative stress (increased glutathione peroxidase) in 30 rheumatoid arthritis patients in a double-blind, placebo-controlled, randomized study [24].

2.1.3. Respiratory inflammation

2.1.3.1. Preclinical studies

Pomegranate peel aqueous extract attenuated lipopolysaccharide (LPS)-induced lung inflammation in mice. Furthermore, it inhibited the production of human neutrophil reactive oxygen species (ROS) and myeloperoxidase [25]. Synergistic anti-inflammatory effect of pomegranate extract (encapsulated into microparticles) with dexamethasone was demonstrated in asthma model mice. The microparticles attenuated leukocytes' recruitment to bronchoalveolar fluid, particularly eosinophils, reduced cytokines (IL-1 β and IL-5), and reduced protein levels in the lungs. These findings supported the alternative/complementary use of pomegranate in treatment of lung inflammation [26]. Pomegranate (80 μ mol/kg/day) significantly attenuated the expression of inflammatory mediators, apoptosis, and oxidative stress that were induced by acute mice exposure to cigarette smoke (for 3 days). Additionally, on chronic cigarette smoke exposure (1–3 months) pomegranate reduced expression of TNF- α and normalized lung cell architectures. Moreover, pomegranate juice attenuated the damaging effects of cigarette smoke extract on cultured human alveolar cells [27]. Pomegranate juice diminished

inflammatory changes in pulmonary tissue via its antioxidative capacity in a study that was carried out on 27 streptozotocin-induced diabetic rats, which were given either pomegranate or saline for 10 weeks [28].

2.2. Cancer

2.2.1. Prostate cancer

2.2.1.1. Preclinical studies

Prostate cancer suppression was exerted by different pomegranate fruit parts (juice, peel, and seed oil) on LNCaP, PC-3, and DU 145 human cancer cell lines. This effect was manifested by inhibition of proliferation, invasion, phospholipase A2 (PLA2) expression, and apoptosis induction [29, 30]. Pomegranate fruit extract inhibited cell growth and induced apoptosis via remodeling of apoptosis regulating proteins in prostate cancer PC-3 cell line. In addition, oral administration of pomegranate fruit extract to mice implanted with CWR22Rnu1 cells significantly suppressed tumor growth and decreased prostate-specific antigen (PSA) in the serum [31, 32]. Oral pomegranate fruit extract (100 mg/kg) for 4 weeks inhibited testosterone-induced prostatic hyperplasia, prostate weight, prostatic acid phosphatase activity, and total glutathione in rats [33].

2.2.1.2. Clinical studies

A two-stage phase-II clinical trial on 46 subjects with recurrent prostate cancer and rising serum prostate-specific antigen (PSA) after surgery or radiotherapy was carried out. The participants consumed daily eight ounces of pomegranate juice (570 mg of total polyphenol gallic acid equivalents) until meeting the disease progression endpoints. About 35% of patients achieved a significant decrease in serum (PSA). There was a significant increase in mean PSA doubling time from baseline of 15–54 months post-treatment. In a parallel in vitro study of patients' serum on LNCaP cell growth, there was a significant reduction in cell proliferation and induction of apoptosis after treatment with pomegranate juice [34].

2.2.2. Breast cancer

Pomegranate constituents have been proved to be antiproliferative, noninvasive [35], apoptotic [36] angiogenesis [37], and tumor growth inhibitors [38]. Pomegranate seed oil and fermented juice polyphenols exhibited antiangiogenesis potential by suppression of vascular endothelial growth factor in MCF-10A and MCF-7 and upregulated migration inhibitory factor (MIF) in MDA-MB-231 breast cancer cell lines [38].

2.2.3. Colon cancer

Pomegranate juice derived ellagitannins and their intestinal bacterial metabolites, urolithins, exhibited dose- and time-dependent decreases in cell proliferation, and clonogenic efficiency of HT-29 cells. The half maximal inhibitory concentration, IC₅₀ values, ranged from 56.7 μ M for urolithin A to 74.8 μ M for urolithin C [39].

2.2.4. Hepatocellular carcinoma

Oxidative stress is a precipitating factor of hepatocellular carcinoma (HCC), one of the most lethal cancers. Pomegranate emulsion (1 or 10 g/kg) was given 4 weeks before dietary carcinogen diethylnitrosamine (DENa)-induced rat hepatocarcinogenesis and 18 weeks thereafter. Pomegranate revealed chemopreventive activity manifested by reduced incidence, number, multiplicity, size, and volume of hepatic nodules. This effect was mediated by pomegranate antioxidant activity and inhibition of nuclear factor-kappaB (NF- κ B) (a potent stimulant of Wnt/ β catenin signaling which is involved in cell proliferation, cell survival, and apoptosis) [40, 41].

2.2.5. Bladder cancer

Transitional cell carcinoma results in most of the bladder tumors [42]. The tumor suppressor gene p53 which is essential for cell cycle arrest and apoptosis [43] was believed to be inactivated in more than 50% of carcinogenesis of bladder cancers [44]. Polyphenols in pomegranate rind extract was shown to inhibit bladder cancer cell EJ proliferation via p53/miR-34a axis [45].

2.3. Cardio vascular disorders

2.3.1. Preclinical studies

Pomegranate protected against cardiovascular injury initiated by cigarette smoking in rats through its antioxidative property [46]. Moreover, antioxidative and anti-inflammatory effects of pomegranate extract reduced the size of atherosclerotic plaques in the aortic sinus and reduced the proportion of coronary arteries with occlusive atherosclerotic plaques when it was given orally in a dose of 307.5 μ L of drinking water/day for 2 weeks to mice model of coronary heart disease [47]. Furthermore, pomegranate extract supplementation (625 mg/day) for 10 days to pigs prevented hyperlipemia-induced coronary endothelial dysfunction via a stimulation of the Akt/endothelial nitric oxide-synthase pathway [48].

2.3.2. Clinical studies

Natural pomegranate juice (150 ml/day) succeeded to significantly lower systolic and diastolic blood pressure 4–6 h post-consumption in 13 hypertensive patients [49]. Furthermore, a 1 year consumption of pomegranate juice by 10 atherosclerotic patients with carotid artery stenosis significantly reduced common carotid intima-media thickness (IMT), systolic blood pressure, and serum lipid peroxidation. Whereas after 3 years of pomegranate consumption, no additional beneficial effects occurred except for further reduction of serum lipid peroxidation by up to 16% [50].

2.4. Metabolic disorders

2.4.1. Preclinical studies

High level of low-density lipoprotein (LDL) is a risk factor for cardiovascular disease. The esterase paraoxonase1 (PON1) prevents oxidation of LDL. Decreased levels of PON1 increase the incidence of cardiovascular disease. Pomegranate juice (12.5 mL/L of juice in 1 l of water/day for 4 months) significantly induced PON1 gene expression and activity when given daily

to streptozotocin-induced diabetic mice fed with a high-fat diet. Furthermore, pomegranate reduced blood glucose level and body weight [51]. Metabolic syndrome includes common clinical disorders such as obesity, hypertension, dyslipidemia, and diabetes. Pomegranate juice and fruit extract induced a significant decrease in vascular inflammation markers; thrombospondin (TSP), and cytokine TGF β 1 and increase in plasma nitrate, nitrite levels, and nitric oxide-synthase expression (important factors for arterial function enhancement) in a metabolic syndrome rat model [52]. Pomegranate extract (300 mg/kg/day for 8 weeks) reduced the levels of high-fat diet-induced elevated serum interleukin 6 (IL6) and corticosterone in rats [53]. Nonalcoholic fatty liver disease (NAFLD) is one of the most common liver diseases in the world [54]. The pathogenesis of NAFLD includes the increased accumulation of triglyceride in hepatocytes, which progresses to nonalcoholic steatohepatitis (NASH) due to oxidative stress. In high-fat, high-sugar-diet-fed rats, pomegranate juice (60 ± 5 ml/day for 7 weeks) exhibited a significant modulation in hepatic steatosis, ballooning, lobular and portal inflammation, as well as significant attenuation of hepatic pro-inflammatory and pro-fibrotic gene expression. It significantly decreased plasma levels of alanine, aspartate aminotransferase, insulin, triglycerides, and glucose with respect to control [55]. A study comparing the anti-diabetogenic effect of glibenclamide (5 mg/kg) and pomegranate juice (1 ml/day) was carried out on 40 streptozotocin (STZ)-nicotinamide (NAD)-induced type 2 diabetes mellitus rats for 21 days. Pomegranate juice (1 mL/day) showed significant repair and restoration signs in islets of Langerhans. Additionally, it significantly lowered the level of plasma total cholesterol, triglyceride, and inflammatory biomarkers, which were actively raised in diabetic rats [56].

2.4.2. Clinical studies

Concentrated pomegranate juice (50 g daily for 4 weeks) exerted a significant increase in total and high-density lipoprotein cholesterol from baseline levels in 40 type 2 diabetic patients. Only serum interleukin-6 (IL-6) was significantly reduced among other tested inflammatory markers. There was about 75% increase in mean value of serum total antioxidant capacity (TAC) [57]. In a double-blinded, randomized crossover controlled study, daily 500 mL of pomegranate juice was introduced to 30 individuals with a metabolic syndrome for a week. Systolic and diastolic blood pressure as well as high sensitivity C-reactive protein was significantly reduced. However, pomegranate consumption significantly increased the level of triglyceride and low-density lipoprotein cholesterol which is attributed by the authors to the more lipogenic effect of fructose than glucose after hepatic metabolism into triglycerides [58]. On the other hand, administration of 400 mg of pomegranate seed oil capsules twice daily for 4 weeks to 25 dyslipidemic patients insignificantly reduced serum of TNF- α level [59].

2.5. Infections

2.5.1. Bacterial and fungal infection

2.5.1.1. Preclinical studies

Antimicrobial activity of pomegranate has been widely investigated in many studies. *Escherichia coli* (E. coli O157:H7) is associated with many disorders: diarrhea, hemorrhagic colitis, thrombocytopenic purpura, and hemolytic uremic syndrome. Pomegranate ethanolic

extract was shown to be bacteriostatic and bacteriocidal against *E. coli* with minimal inhibitory concentration (MICs) from 0.49 to 1.95 mg/ml and minimal bactericidal concentration (MBCs) from 1.95 to 3.91 mg/ml [60]. Tuberculosis is an infectious disease with a time long emergence of drug resistance. Pomegranate *juice*, and *peel extracts* prepared with methanol/water, and its polyphenolic byproducts namely caffeic acid, ellagic acid, epigallocatechin-3-gallate (EGCG) and quercetin, were examined against drug-resistant clinical isolates of *Mycobacterium tuberculosis* and β -lactamase producing *Klebsiella pneumoniae*. The peel extracts exerted higher anti-mycobacterial activity (MIC 64–1024 $\mu\text{g}/\text{mL}$) than the juice (MIC 256 - > 1024 $\mu\text{g}/\text{mL}$). EGCG and quercetin showed more antitubercular and antibacterial activity than caffeic acid and ellagic acid [61]. Biofilm is a protective layer made of extracellular polymeric substances where the pathogen hides with subsequent modulation of its virulence and pathogenicity. Pomegranate methanolic extract was believed to counteract the formation of biofilms by *Staphylococcus aureus*, methicillin-resistant *S. aureus*, *E. coli*, and *Candida albicans*. Moreover, pomegranate extract disrupted the preformed biofilms with inhibition of germ tube formation, a virulence trait, in *C. albicans*. Further studies revealed the ability of ellagic acid to inhibit the growth of all species in suspension at higher concentrations (>75 $\mu\text{g ml}^{-1}$) and biofilm formation at lower concentrations (<40 $\mu\text{g ml}^{-1}$) [62]. Besides single antifungal activity, pomegranate extract showed a synergistic effect with other antimicrobial agents. Punicalagin synergism with fluconazole against *C. albicans* and *C. parapsilosis* was demonstrated in an in vitro study with a twofold decrease of MIC for fluconazole [63]. Pomegranate methanolic extract showed synergistic effect with five antibiotics: chloramphenicol, gentamicin, ampicillin, tetracycline, and oxacillin against methicillin-resistant *S. aureus* strains (MRSA) and methicillin-sensitive *S. aureus* (MSSA). Most potent synergism was noticed when pomegranate was combined with ampicillin. This combination increased the post-antibiotic effect (PAE) of ampicillin from 3 to 7 h. as well as it reduced cell viability by 99.9 and 72.5% in MSSA and MRSA populations, respectively [64]. Methanol extract of pomegranate showed a synergistic action with ciprofloxacin against extended-spectrum β -lactamase (ESBL) producing *E. coli* and metallo- β -lactamase (MBL) producing *Pseudomonas aeruginosa*; that effect was attributed to bacterial efflux pump inhibitor (EPI) activity of the pomegranate polyphenolic constituents [65]. In an ongoing study of our work where we are testing the antifungal activity of some medicinal herbs against clinical isolates of *C. albicans* strain, pomegranate methanolic extract showed an inhibitory zone of 12 mm and synergistically augmented the action of fluconazole by increasing the inhibition zone from 25 to 35 mm after combination. The potent antifungal action of pomegranate led some researchers [66] to design a new antifungal peptide, pomegranin, with an N-terminal sequence from fresh pomegranate peels by ion exchange chromatography. Pomegranin suppressed mycelial growth in the fungi *Botrytis cinerea* and *Fusarium oxysporum* with half maximal inhibitory concentration (IC₅₀) of 2 and 6.1 μM , respectively.

2.5.2. Virus infection

2.5.2.1. Preclinical studies

Pomegranate showed antiviral action against many viruses: influenza, human immunodeficiency virus (HIV), herpes simplex virus (HSV), and adenoviruses in multiple studies. Of pomegranate polyphenol extract (PPE) constituents (ellagic acid, caffeic acid, luteolin, and

punicalagin), punicalagin had the highest affect against influenza A virus through suppression of viral RNA replication and agglutination of chicken RBCs. In addition, pomegranate polyphenol extract augmented the anti-influenza effect of oseltamivir when given together [67]. Pomegranate juice prevented HIV-1 binding to CD4 and blocked viral entry [68]. Moreover, agents present in pomegranate juice (polyphenols, beta-sitosterol, sugars, and ellagic acid) and fulvic acid were demonstrated as envelope virus neutralizing compounds that neutralize the viral infectivity by binding to the envelope lipid or sugar moieties [69]. Adenoviruses are a group of non-enveloped viruses that give rise to in a wide range of illnesses. Pomegranate peel ethanol extract exhibited anti-adenovirus activity on HeLa cell line where the half maximal inhibitory concentration (IC₅₀) and 50% cytotoxicity concentration (CC₅₀) of the extract were 165 ± 10.1 and 18.6 ± 6.7 $\mu\text{g/ml}$, respectively. The selectivity index (SI), the ratio of CC₅₀ and IC₅₀, was 8.89 [70]. Moreover, pomegranate tannins were shown to have anti-HSV-1, HSV-2 effect via blocking of virus adsorption to African green monkey kidney and human adenocarcinoma cells [71]. Hepatitis C virus (HCV) is the leading cause of end-stage liver disease. Ellagitannins from pomegranate peel crude extract, punicalagin, punicalin, and ellagic acid, specifically blocked the HCV NS3/4A protease activity in an in vitro study. Furthermore, punicalagin and punicalin significantly suppressed HCV replication in cell culture system. Moreover, these compounds were well tolerated ex vivo and “no-observed adverse effect level” (NOAEL) was established up to an acute dose of 5000 mg/kg in BALB/c mice. Additionally, these components were bio-available by pharmacokinetics study [72].

2.5.3. Parasitic infection

2.5.3.1. Preclinical studies

From ancient times, pomegranate was described as an antihelminthic agent. Malarial infection represents a public health and economic burden in tropical and subtropical regions of the world [73]. Pomegranate gallagic acid and punicalagin exerted an antiplasmodial activity against *Plasmodium falciparum* D6 and W2 clones with IC₅₀ values of 10.9, 10.6, 7.5, and 8.8 μM , respectively [74].

Schistosomiasis is a morbid widely distributed tropical disease [75]. Blood flukes of the genus *Schistosoma* pass a complex life cycle including multiple morphologically distinct phenotypes in definitive human and intermediate snail hosts [76]. In vitro and in vivo studies were designed to evaluate pomegranate impact on *Schistosoma mansoni* (*S. mansoni*), one of the three major species infecting humans. Pomegranate peels and leaves extracts significantly affected both adult *S. mansoni* worms and schistosomules with 100% death rate, after 24 h of exposure to plant extracts. Oral administration of the pomegranate extract to mice at a dose of 800 mg/kg, 45 days post-infection and on three consecutive days yielded a high percentage of dead adult worms (77.30 and 72.2) with either leaves or peels extract, respectively. In addition, reduction in tissue egg load, liver, and intestinal ova counts was observed. This antiparasitic effect was confirmed by electron microscopic examination that revealed ultrastructural alterations in the tegument and the male genital systems of the worms. Bone marrow examination of pomegranate-treated *S. mansoni*-infected mice showed eosinophilic degranulation that indicates reduced *S. mansoni* activity [77].

2.6. Central nervous system disorders

2.6.1. Cognitive disorders

2.6.1.1. Preclinical studies

Cognitive disorders affect learning, memory, perception, and problem-solving. These disorders include amnesia, dementia, and delirium. Pomegranate ellagic acid (30 and 100 mg/kg) ameliorated scopolamine- (0.4 mg/kg, i.p.) and diazepam (1 mg/kg, i.p.)-induced amnesia in mice. Furthermore, chronic administration of ellagic acid (30 mg/kg) improved the memory deficit induced by diazepam (1 mg/kg) in rats [78]. Memory impairment, a feature of Alzheimer's disease (AD), is initiated by neuroinflammation and impairments in synaptic plasticity. These disorders are induced by the effect of extracellular amyloid-beta ($A\beta$) deposits called senile plaques. The generation of $A\beta$ is dependent on the proteolytic processing of amyloid precursor protein (APP) [79]. Pomegranate is believed to slow the rate of neurodegeneration in Alzheimer's disease. At a cellular level, pomegranate compound, punicalagin, was examined for its memory protective anti-inflammatory effect on lipopolysaccharide (LPS)-induced neuroinflammation in astrocytes and microglial BV-2 cells. In a dose of 1.5 mg/kg punicalagin attenuated LPS (250 μ g/kg daily 7 times) induced memory impairment and blocked the LPS-induced expression of inflammatory proteins via suppression of NF- κ B activation [80]. In addition, freeze-dried pomegranate (25–200 μ g/ml) in a dose-dependent manner reduced COX-2-dependent prostaglandin E2 (PGE2) production in SK-N-SH cells stimulated with IL-1 β [81]. The neuroprotective action of pomegranate was obscured in an animal study in which dietary supplementation of 4% pomegranate extract to APP^{sw}/Tg2576 mice for 15 months ameliorated the loss of synaptic structure proteins, inhibited neuroinflammatory activity, and enhanced autophagy (degradation and recycling of cellular components). Moreover, it reduced β -site cleavage of APP [82]. Along with figs and dates, pomegranate dietary intake attenuated the levels of inflammatory cytokines in APP^{sw}/Tg2576 mice a model of Alzheimer disease, as well as delayed the formation of senile plaques [83].

2.6.2. Ischemic stroke

2.6.2.1. Preclinical studies

Ischemic stroke is one of the neurodegenerative diseases. An in vitro study utilized serum glucose deprivation (SGD) as a model for ischemia-induced brain injury in PC12 cells. Pretreatment with different pomegranate extracts, namely, pulp hydroalcoholic extract (PHE), pulp aqueous extract (PAE), and pomegranate for 2 h significantly and concentration-dependently, increased cell viability and decreased DNA damage initiated by SGD insult [84].

2.6.3. Multiple sclerosis

2.6.3.1. Preclinical studies

Multiple sclerosis (MS) is a chronic inflammatory disease of the central nervous system and is associated with demyelination, neurodegeneration, and sensitivity to oxidative stress.

Pomegranate seed oil (PSO) in nanodroplet formulation induced more significant beneficial effects than in mice model of multiple sclerosis (MS) than natural pomegranate seed oil. This effect was evident by dramatic alleviation of lipid demyelination and oxidation in mice brains [85].

2.6.4. Neonatal hypoxic-ischemic brain injury

2.6.4.1. Preclinical studies

Neonatal hypoxic-ischemic (HI) brain injury is a fatal condition that affects preterm very low birth-weight infants. After administration to pregnant mice, pomegranate juice revealed antioxidant-driven neuroprotective effect in experimentally induced HI brain injured neonatal offsprings [86–87].

2.7. Miscellaneous disorders

2.7.1. Skin disorders

2.7.1.1. Preclinical studies

Prolonged human exposure to sun's ultraviolet (UV) radiation, especially its UV-B, causes many adverse effects. Pomegranate fruit extract was proved to be a photo-chemo preventive agent on human epidermal keratinocytes. It alleviated ultraviolet A and B radiation-induced cell damage in a dose- and time-dependent manner [88, 89].

2.7.1.2. Clinical studies

Oral ellagic acid-rich pomegranate extract either in high (200 mg/d ellagic acid) or low doses (100 mg/d ellagic acid) improved ultraviolet-induced skin pigmentation of 26 subjects in 4 weeks double-blind placebo-controlled trial [90].

2.7.2. Male infertility and erectile dysfunction

2.7.2.1. Preclinical studies

Pomegranate juice improved epididymal sperm concentration, spermatogenic cell density, diameter of seminiferous tubules, and sperm motility. It decreased the number of abnormal sperms compared to control rat animals. Moreover, pomegranate juice resulted in improvement of antioxidant enzyme activity in both rat plasma and sperm [91]. Pomegranate juice significantly increased intracavernous blood flow and smooth muscle relaxation in a rabbit model of arteriogenic erectile dysfunction [92].

2.7.2.2. Clinical studies

In a randomized, double-blind, placebo-controlled, 10-week crossover trial, pomegranate juice (1.5 mmol polyphenols daily) showed insignificant improvement when introduced to 53 men with mild-to-moderate erectile dysfunction [93].

2.7.3. Dental disorders

2.7.3.1. Preclinical studies

Bacterial and fungal co-infection initiates oral diseases. Pomegranate phytotherapeutic gel was shown to be superior to miconazole in attenuation of microbial adherence with three and four associated organisms: *Streptococci* strains (*mutans* ATCC 25175, *sanguis* ATCC 10577 and *mitis* ATCC 9811) and *C. albicans* [94]. Elagic acid exerted a moderate inhibitory effect at 12.5 mg/mL with inhibition to adherence <50% against different strains of *Streptococcus mutans* bacteria that induced dental caries [95].

2.7.3.2. Clinical studies

In a human study, pomegranate hydroalcoholic extract was superior to chlorhexidine (standard and positive control) in decreasing the colony forming unit (CFU)/ml by 84 and 79%, respectively, of dental plaque microorganisms [96]. Pomegranate along with *Centella asiatica* extracts significantly improved clinical signs of chronic periodontitis and IL-1 β level when it was applied with biodegradable chips on periodontal disease in 20 patients with remaining probing pocket depths after conventional periodontal therapy [97]. Pomegranate gel was compared to miconazol gel (3 times daily for 15 days) in 60 patients suffering from denture stomatitis. The patients were randomly distributed into two groups of 30 patients each. Clinical response was statistically better in miconazol group ($P < 0.01$) with similar fungal negativity in both groups. Clinical response and fungal negativity was achieved in 21 and 23 patients of pomegranate group as compared to 27 and 25 subjects who received miconazol, respectively. Side effects were only reported from all miconazol-treated patients. The authors explained the better miconazol clinical response by the bigger number of subjects with good oral hygiene score in the miconazol group and the longer duration of miconazol (sticky formulation) in the mouth than pomegranate gel that was washed away on mixing with saliva [98].

3. Pharmacokinetic studies

Pomegranate ellagitannins release ellagic acid in the gut, and this compound is poorly absorbed in the small intestine, while it is largely metabolized by human gut microflora into urolithins, such as urolithins A and B and urolithin-8-methyl ether in the large intestine [99]. Pomegranate anthocyanins (the 3-glucosides and 3, 5-diglucosides of delphinidin, cyanidin, and pelargonidin) are stable in the stomach. While in the neutral pH of the small and large intestines, anthocyanins become less stable and are converted into a variety of metabolites [100–102].

The maximum plasma concentration (C_{max}) of ellagic acid was 33 ng/mL and time of maximum concentration (T_{max}) was 1 h [103]. A pharmacokinetic study on 18 healthy volunteers proved the rapid absorption and plasma clearance of ellagitannins as well as long persistence

(48 h) of urinary excreted urolithin metabolites after 180 ml of pomegranate juice consumption. Prolonged stay of urolithins in the human body is responsible for the health benefits of chronic pomegranate consumption [104]. A 1 liter pomegranate juice containing 4.37 g/L punicalagins and 0.49 g/L anthocyanins was introduced to six healthy individuals for 5 days; urolithin A, urolithin B, and a third unidentified minor metabolite were detected in plasma as well as in urine analysis at 24 h besides an aglycone metabolite corresponding to each of three plasma metabolites. Maximum excretion rates occurred 3–4 days after juice ingestion. The concentrations of urinary metabolites varied significantly in the subjects which may be attributed to colonic microflora variability and the site of ellagitannins metabolism [105]. A crossover pharmacokinetic study reported that higher free ellagic acid EA intake does not enhance its bioavailability in healthy volunteers who consumed two pomegranate extracts of 130 mg punicalagin+524 mg ellagic acid or 279 mg punicalagin+25 mg ellagic acid. The study showed high inter-individual variability; C_{max} ranged from 12 to 360 nM that may be attributed to the ellagitannin pH and protein environment [106].

4. Safety

Pomegranate is safe when it is used in normal doses [107]. The median lethal dose, LD₅₀ of the whole fruit extract, was 731 mg/kg after intra-peritoneal administration to OF-1 mice [108]. Standardized pomegranate extract of 30% punicalagins showed acute oral LD₅₀ in wistar rats and in Swiss albino mice it was more than 5000 mg/kg. Subchronic no-observed adverse effect level (NOAEL) was 600 mg/kg body weight/day [109]. Pomegranate ellagitannin-enriched polyphenol extract in a daily dose of 1420 mg (870 mg of gallic acid equivalents,) for 28 days showed no adverse effects in 64 overweight subjects [110].

5. Conclusion

Pomegranate's uncountable beneficial pharmacological properties encourage more and more studies to discover other secrets for solving mankind health problems.

Acknowledgements

First and foremost, my deepest gratitude to GOD, for his uncountable gifts including pomegranate. Second, I would like to thank my parents and family for continuous encouragement. I would also like to express my thanks to Dr. Farid Badria, Professor of Pharmacognosy, Faculty of Pharmacy, Mansoura University, Egypt, and Dr. Khalil Mahfouz, Assistant Professor of Botany, Faculty of Science, Tanta University, Egypt, for their generous advice and help. My great appreciation to Professor Dr. Said Shalaby, Vice President, Academy of Scientific Research and Technology, Cairo, Egypt, for his unforgettable support.

Author details

Sally Elnawasany

Address all correspondence to: elnawasany_s@hotmail.com

Faculty of Medicine, Tanta University, Tanta, Egypt

References

- [1] Langley P. Why a pomegranate? British Medical Journal. 2000;**321**(7269):1153
- [2] Cáceres A, Girón LM, Alvarado SR, Torres MF. Screening of antimicrobial activity of plants popularly used in Guatemala for the treatment of dermatomucosal diseases. Journal of Ethnopharmacology. 1987;**20**(3):223-237
- [3] Naqvi S, Khan M, Vohora S. Anti-bacterial, anti-fungal and anthelmintic investigations on Indian medicinal plants. Fitoterapia. 1991;**62**:221-228
- [4] Saxena A, Vikram NK. Role of selected Indian plants in management of type 2 diabetes: A review. The Journal of Alternative and Complementary Medicine. 2004;**10**(2):369-378
- [5] Zaid H, Silbermann M, Ben-Arye E, Saad B. Greco-Arab and Islamic herbal-derived anticancer modalities: From tradition to molecular mechanisms. Evidence-based Complementary and Alternative Medicine. 2012;**2012**
- [6] Lansky EP, Newman RA. Punica granatum (pomegranate) and its potential for prevention and treatment of inflammation and cancer. Journal of Ethnopharmacology. 2007;**109**(2):177-206
- [7] Schubert SY, Lansky EP, Neeman I. Antioxidant and eicosanoid enzyme inhibition properties of pomegranate seed oil and fermented juice flavonoids. Journal of Ethnopharmacology. 1999;**66**(1):11-17
- [8] Hajimahmoodi M, Shams-Ardakani M, Saniee P, Siavoshi F, Mehrabani M, Hosseinzadeh H, et al. In vitro antibacterial activity of some Iranian medicinal plant extracts against *helicobacter pylori*. Natural Product Research. 2011;**25**(11):1059-1066
- [9] Voravuthikunchai SP, Limsuwan S, Mitchell H. Effects of *Punica granatum* pericarps and *Quercus infectoria* nutgalls on cell surface hydrophobicity and cell survival of *helicobacter pylori*. Journal of health science. 2006;**52**(2):154-159
- [10] Ajaikumar K, Asheef M, Babu B, Padikkala J. The inhibition of gastric mucosal injury by *Punicagranatum* L.(pomegranate) methanolic extract. Journal of Ethnopharmacology. 2005;**96**(1):171-176
- [11] Alam MS, Alam MA, Ahmad S, Najmi AK, Asif M, Jahangir T. Protective effects of *Punica granatum* in experimentally-induced gastric ulcers. Toxicology Mechanisms and Methods. 2010;**20**(9):572-578

- [12] Hassanpour S, Sadaghian M, MaheriSis N, Eshratkhah B, ChaichiSemsari M. Effect of condensed tannin on controlling faecal protein excretion in nematode-infected sheep: In vivo study. *Journal of American Science*. 2011;**7**(5):896-900
- [13] Hassanpour S, Maheri-Sis N, Eshratkhah B, Mehmandar FB. Plants and secondary metabolites (tannins): A review. *International Journal of Forest, Soil and Erosion*. 2011;**1**(1):47-53. ISSN 2251-6387
- [14] de Jesus NZT, Falcão HDS, Gomes IF, Leite TJDA, Lima GRDM, Barbosa-Filho JM, et al. Tannins, peptic ulcers and related mechanisms. *International Journal of Molecular Sciences*. 2012;**13**(3):3203-3228
- [15] Chidambara Murthy K, Reddy VK, Veigas JM, Murthy UD. Study on wound healing activity of *Punica granatum* peel. *Journal of Medicinal Food*. 2004;**7**(2):256-259
- [16] Prabhu K, Bhute AS. Plant based natural dyes and mordants: A review. *Journal of Natural Product and Plant Resources*. 2012;**2**(6):649-664
- [17] Morton J. Pomegranate. In: Morton JF, editor. *Fruits of Warm Climate*. Miami, FL: Florida Flair Books; 1987. pp. 352-355
- [18] Adams LS, Seeram NP, Aggarwal BB, Takada Y, Sand D, Heber D. Pomegranate juice, total pomegranate ellagitannins, and punicalagin suppress inflammatory cell signaling in colon cancer cells. *Journal of Agricultural and Food Chemistry*. 2006;**54**(3):980-985
- [19] Neyrinck AM, Van Hée VF, Bindels LB, De Backer F, Cani PD, Delzenne NM. Polyphenol-rich extract of pomegranate peel alleviates tissue inflammation and hypercholesterolaemia in high-fat diet-induced obese mice: Potential implication of the gut microbiota. *British Journal of Nutrition*. 2013;**109**(5):802-809
- [20] Ogawa Y, Kanatsu K, Iino T, Kato S, Jeong Y, Shibata N, et al. Protection against dextran sulfate sodium-induced colitis by microspheres of ellagic acid in rats. *Life Sciences*. 2002;**71**(7):827-839
- [21] Singh K, Jaggi AS, Singh N. Exploring the ameliorative potential of *Punica granatum* in dextran sulfate sodium induced ulcerative colitis in mice. *Phytotherapy Research*. 2009;**23**(11):1565-1574
- [22] Seong AR, Yoo JY, Choi K, Lee MH, Lee YH, Lee J, et al. Delphinidin, a specific inhibitor of histone acetyltransferase, suppresses inflammatory signaling via prevention of NF- κ B acetylation in fibroblast-like synoviocyte MH7A cells. *Biochemical and Biophysical Research Communications*. 2011;**410**(3):581-586
- [23] Shukla M, Gupta K, Rasheed Z, Khan KA, Haqqi TM. Consumption of hydrolyzable tannins-rich pomegranate extract suppresses inflammation and joint damage in rheumatoid arthritis. *Nutrition*. 2008;**24**(7):733-743
- [24] Ghavipour M, Sotoudeh G, Tavakoli E, Mowla K, Hasanzadeh J, Mazloom Z. Pomegranate extract alleviates disease activity and some blood biomarkers of inflammation and oxidative stress in rheumatoid arthritis patients. *European Journal of Clinical Nutrition*. 2017;**71**(1):92

- [25] Bachoual R, Talmoudi W, Boussetta T, Braut F, El-Benna J. An aqueous pomegranate peel extract inhibits neutrophil myeloperoxidase in vitro and attenuates lung inflammation in mice. *Food and Chemical Toxicology*. 2011;**49**(6):1224-1228
- [26] de Oliveira JF, Garreto DV, da Silva MC, Fortes TS, de Oliveira RB, Nascimento FR, et al. Therapeutic potential of biodegradable microparticles containing *Punica granatum* L. (pomegranate) in murine model of asthma. *Inflammation Research* 2013;**62**(11):971-980
- [27] Husari A, Hashem Y, Bitar H, Dbaiibo G, Zaatari G, El Sabban M. Antioxidant activity of pomegranate juice reduces emphysematous changes and injury secondary to cigarette smoke in an animal model and human alveolar cells. *International Journal of Chronic Obstructive Pulmonary Disease*. 2016;**11**:227
- [28] Cukurova Z, Hergünel O, Eren G, Gedikbaşı A, Uhri M, Demir G, et al. The effect of pomegranate juice on diabetes-related oxidative stress in rat lung. *Turkiye Klinikleri Journal of Medical Sciences*. 2012;**32**(2):444-452
- [29] Lansky EP, Jiang W, Mo H, Bravo L, Froom P, Yu W, et al. Possible synergistic prostate cancer suppression by anatomically discrete pomegranate fractions. *Investigational New Drugs*. 2005;**23**(1):11-20
- [30] Albrecht M, Jiang W, Kumi-Diaka J, Lansky EP, Gommersall LM, Patel A, et al. Pomegranate extracts potently suppress proliferation, xenograft growth, and invasion of human prostate cancer cells. *Journal of Medicinal Food*. 2004;**7**(3):274-283
- [31] Malik A, Afaq F, Sarfaraz S, Adhami VM, Syed DN, Mukhtar H. Pomegranate fruit juice for chemoprevention and chemotherapy of prostate cancer. *Proceedings of the National Academy of Sciences of the United States of America*. 2005;**102**(41):14813-14818
- [32] Malik A, Mukhtar H. Prostate cancer prevention through pomegranate fruit. *Cell Cycle (Georgetown, Tex)*. 2006;**5**(4):371-373
- [33] Ammar AE, Esmat A, Hassona MD, Tadros MG, Abdel-Naim AB, Guns EST. The effect of pomegranate fruit extract on testosterone-induced BPH in rats. *The Prostate*. 2015;**75**(7):679-692
- [34] Pantuck A, Leppert J, Zomorodian N, Aronson W, Hong J, Barnard RJ, Seeram N, Liker H, Wang H, Elashoff R, Heber D, Aviram M, Ignarro L, Belldegrun A. Phase II study of pomegranate juice for men with rising prostate-specific antigen following surgery or radiation for prostate cancer. *Clinical Cancer Research*. 2006;**12**:4018-4026
- [35] Kim ND, Mehta R, Yu W, Neeman I, Livney T, Amichay A, et al. Chemopreventive and adjuvant therapeutic potential of pomegranate (*Punica granatum*) for human breast cancer. *Breast Cancer Research and Treatment*. 2002;**71**(3):203-217
- [36] Jeune ML, Kumi-Diaka J, Brown J. Anticancer activities of pomegranate extracts and genistein in human breast cancer cells. *Journal of Medicinal Food*. 2005;**8**(4):469-475
- [37] Toi M, Bando H, Ramachandran C, Melnick SJ, Imai A, Fife RS, et al. Preliminary studies on the anti-angiogenic potential of pomegranate fractions in vitro and in vivo. *Angiogenesis*. 2003;**6**(2):121-128

- [38] Mehta R, Lansky E. Breast cancer chemopreventive properties of pomegranate (*Punica granatum*) fruit extracts in a mouse mammary organ culture. *European Journal of Cancer Prevention*. 2004;**13**(4):345-348
- [39] Kasimsetty SG, Bialonska D, Reddy MK, Ma G, Khan SI, Ferreira D. Colon cancer chemopreventive activities of pomegranate ellagitannins and urolithins. *Journal of Agricultural and Food Chemistry*. 2010;**58**(4):2180-2187
- [40] Bishayee A, Bhatia D, Thoppil RJ, Darvesh AS, Nevo E, Lansky EP. Pomegranate-mediated chemoprevention of experimental hepatocarcinogenesis involves Nrf2-regulated antioxidant mechanisms. *Carcinogenesis*. 2011;**32**(6):888-896
- [41] Bhatia D, Thoppil RJ, Mandal A, Samtani KA, Darvesh AS, Bishayee A. Pomegranate bioactive constituents suppress cell proliferation and induce apoptosis in an experimental model of hepatocellular carcinoma: Role of Wnt/ β -catenin signaling pathway. *Evidence-based Complementary and Alternative Medicine*. 2013;**2013**
- [42] Pignot G, Vieillefond A, Vacher S, Zerbib M, Debre B, Lidereau R, et al. Hedgehog pathway activation in human transitional cell carcinoma of the bladder. *British Journal of Cancer*. 2012;**106**(6):1177-1186
- [43] Li C, Johnson DE. Liberation of functional p53 by proteasome inhibition in human papilloma virus-positive head and neck squamous cell carcinoma cells promotes apoptosis and cell cycle arrest. *Cell Cycle*. 2013;**12**(6):923-934
- [44] Hilton WM, Svatek RS. Re: Phase III study of molecularly targeted adjuvant therapy in locally advanced urothelial cancer of the bladder based on p53 status. *European Urology*. 2012;**61**(5):1062-1063
- [45] Zhou B, Yi H, Tan J, Wu Y, Liu G, Qiu Z. Anti-proliferative effects of polyphenols from pomegranate rind (*Punica granatum* L.) on EJ bladder cancer cells via regulation of p53/miR-34a Axis. *Phytotherapy Research*. 2015;**29**(3):415-422
- [46] Al Hariri M, Zibara K, Farhat W, Hashem Y, Soudani N, Al Ibrahim F, et al. Cigarette smoking-induced cardiac hypertrophy, vascular inflammation and injury are attenuated by antioxidant supplementation in an animal model. *Frontiers in Pharmacology*. 2016;**7**
- [47] Al-Jarallah A, Igdoura F, Zhang Y, Tenedero CB, White EJ, MacDonald ME, et al. The effect of pomegranate extract on coronary artery atherosclerosis in SR-BI/APOE double knockout mice. *Atherosclerosis*. 2013;**228**(1):80-89
- [48] Vilahur G, Padró T, Casaní L, Mendieta G, López JA, Streitenberger S, et al. Polyphenol-enriched diet prevents coronary endothelial dysfunction by activating the Akt/eNOS pathway. *Revista Española de Cardiología (English Edition)*. 2015;**68**(3):216-225
- [49] Asgary S, Keshvari M, Sahebkar A, Hashemi M, Rafieian-Kopaei M. Clinical investigation of the acute effects of pomegranate juice on blood pressure and endothelial function in hypertensive individuals. *ARYA Atherosclerosis*. 2013;**9**(6):326
- [50] Aviram M, Rosenblat M, Gaitini D, Nitecki S, Hoffman A, Dornfeld L, et al. Pomegranate juice consumption for 3 years by patients with carotid artery stenosis reduces common

- carotid intima-media thickness, blood pressure and LDL oxidation. *Clinical Nutrition*. 2004;**23**(3):423-433
- [51] Betanzos-Cabrera G, Guerrero-Solano J, Martínez-Pérez M, Calderón-Ramos Z, Belefant-Miller H, Cancino-Diaz JC. Pomegranate juice increases levels of paraoxonase1 (PON1) expression and enzymatic activity in streptozotocin-induced diabetic mice fed with a high-fat diet. *Food Research International*. 2011;**44**(5):1381-1385
- [52] de Nigris F, Balestrieri ML, Williams-Ignarro S, D'Armiento FP, Fiorito C, Ignarro LJ, et al. The influence of pomegranate fruit extract in comparison to regular pomegranate juice and seed oil on nitric oxide and arterial function in obese Zucker rats. *Nitric Oxide* 2007;**17**(1):50-54
- [53] Dushkin M, Khrapova M, Kovshik G, Chasovskikh M, Menshchikova E, Trufakin V, et al. Effects of rhaponticum carthamoides versus glycyrrhiza glabra and *punica granatum* extracts on metabolic syndrome signs in rats. *BMC Complementary and Alternative Medicine*. 2014;**14**(1):33
- [54] Farrell GC, Larter CZ. Nonalcoholic fatty liver disease: From steatosis to cirrhosis. *Hepatology*. 2006;**43**:S1
- [55] Noori M, Jafari B, Hekmatdoost A. Pomegranate juice prevents development of non-alcoholic fatty liver disease in rats by attenuating oxidative stress and inflammation. *Journal of the Science of Food and Agriculture*. 2017;**97**(8):2327-2332
- [56] Rouhi SZT, Sarker MMR, Rahmat A, Alkahtani SA, Othman F. The effect of pomegranate fresh juice versus pomegranate seed powder on metabolic indices, lipid profile, inflammatory biomarkers, and the histopathology of pancreatic islets of Langerhans in streptozotocin-nicotinamide induced type 2 diabetic Sprague-Dawley rats. *BMC Complementary and Alternative Medicine*. 2017;**17**(1):156
- [57] Shishehbor F. Effects of concentrated pomegranate juice on subclinical inflammation and cardiometabolic risk factors for type 2 diabetes: A quasi-experimental study. *International Journal of Endocrinology and Metabolism*. 2016;**14**(1)
- [58] Moazzen H, Alizadeh M. Effects of pomegranate juice on cardiovascular risk factors in patients with metabolic syndrome: A double-blinded, randomized crossover controlled trial. *Plant Foods for Human Nutrition*. 2017;**72**(2):126-133
- [59] Asghari G, Sheikholeslami S, Mirmiran P, Chary A, Hedayati M, Shafiee A, et al. Effect of pomegranate seed oil on serum TNF- α level in dyslipidemic patients. *International Journal of Food Sciences and Nutrition*. 2012;**63**(3):368-371
- [60] Voravuthikunchai SP, Limsuwan S. Medicinal plant extracts as anti-*Escherichia coli* O157: H7 agents and their effects on bacterial cell aggregation. *Journal of Food Protection*. 2006;**69**(10):2336-2341
- [61] Dey D, Ray R, Hazra B. Antimicrobial activity of pomegranate fruit constituents against drug-resistant mycobacterium tuberculosis and β -lactamase producing *Klebsiella pneumoniae*. *Pharmaceutical Biology*. 2015;**53**(10):1474-1480

- [62] Bakkiyaraj D, Nandhini JR, Malathy B, Pandian SK. The anti-biofilm potential of pomegranate (*Punica granatum* L.) extract against human bacterial and fungal pathogens. *Biofouling*. 2013;**29**(8):929-937
- [63] Endo EH, Cortez DAG, Ueda-Nakamura T, Nakamura CV, Dias Filho BP. Potent antifungal activity of extracts and pure compound isolated from pomegranate peels and synergism with fluconazole against *Candida albicans*. *Research in Microbiology*. 2010;**161**(7):534-540
- [64] Braga L, Leite AA, Xavier KG, Takahashi J, Bemquerer M, Chartone-Souza E, et al. Synergic interaction between pomegranate extract and antibiotics against *Staphylococcus aureus*. *Canadian Journal of Microbiology*. 2005;**51**(7):541-547
- [65] Dey D, Debnath S, Hazra S, Ghosh S, Ray R, Hazra B. Pomegranate pericarp extract enhances the antibacterial activity of ciprofloxacin against extended-spectrum β -lactamase (ESBL) and metallo- β -lactamase (MBL) producing gram-negative bacilli. *Food and Chemical Toxicology*. 2012;**50**(12):4302-4309
- [66] Guo G, Wang HX, Ng TB. Pomegranin, an antifungal peptide from pomegranate peels. *Protein and Peptide Letters*. 2009;**16**(1):82-85
- [67] Haidari M, Ali M, Casscells SW, Madjid M. Pomegranate (*Punica granatum*) purified polyphenol extract inhibits influenza virus and has a synergistic effect with oseltamivir. *Phytomedicine*. 2009;**16**(12):1127-1136
- [68] Neurath AR, Strick N, Li Y-Y, Debnath AK. *Punica granatum* (pomegranate) juice provides an HIV-1 entry inhibitor and candidate topical microbicide. *BMC Infectious Diseases*. 2004;**4**(1):41
- [69] Kotwal GJ. Genetic diversity-independent neutralization of pandemic viruses (eg HIV), potentially pandemic (eg H5N1 strain of influenza) and carcinogenic (eg HBV and HCV) viruses and possible agents of bioterrorism (variola) by enveloped virus neutralizing compounds (EVNCs). *Vaccine*. 2008;**26**(24):3055-3058
- [70] Moradi M-T, Karimi A, Alidadi S, Saedi-Marghmaleki M. In vitro anti-adenovirus activity of pomegranate (*Punica granatum* L.) peel extract. *Advanced Herbal Medicine*. 2015;**1**(4):1-8
- [71] Fukuchi K, Sakagami H, Okuda T, Hatano T, S-i T, Kitajima K, et al. Inhibition of herpes simplex virus infection by tannins and related compounds. *Antiviral Research*. 1989;**11**(5-6):285-297
- [72] Reddy BU, Mullick R, Kumar A, Sudha G, Srinivasan N, Das S. Small molecule inhibitors of HCV replication from pomegranate. *Scientific Reports*. 2014;**4**:5411
- [73] Breman JG. The ears of the hippopotamus: Manifestations, determinants, and estimates of the malaria burden. *The American Journal of Tropical Medicine and Hygiene*. 2001;**64**(1_suppl):1-11

- [74] Reddy MK, Gupta SK, Jacob MR, Khan SI, Ferreira D. Antioxidant, antimalarial and antimicrobial activities of tannin-rich fractions, ellagitannins and phenolic acids from *Punica granatum* L. *Planta Medica*. 2007;**53**(05):461-467
- [75] Steinmann P, Keiser J, Bos R, Tanner M, Utzinger J. Schistosomiasis and water resources development: Systematic review, meta-analysis, and estimates of people at risk. *The Lancet Infectious Diseases*. 2006;**6**(7):411-425
- [76] Gryseels B, Polman K, Clerinx J, Kestens L. Human schistosomiasis. *The Lancet*. 2006;**368**(9541):1106-1118
- [77] Fahmy Z, El-Shennawy A, El-Komy W, Ali E, Hamid SA. Potential antiparasitic activity of pomegranate extracts against schistosomules and mature worms of *Schistosoma mansoni*: In vitro and in vivo study. *Australian Journal of Basic and Applied Sciences*. 2009;**3**(4):4634-4643
- [78] Mansouri MT, Farbood Y, Naghizadeh B, Shabani S, Mirshekar MA, Sarkaki A. Beneficial effects of ellagic acid against animal models of scopolamine- and diazepam-induced cognitive impairments. *Pharmaceutical Biology*. 2016;**54**(10):1947-1953
- [79] Naidoo R, Knapp ML. Studies of lipid peroxidation products in cerebrospinal fluid and serum in multiple sclerosis and other conditions. *Clinical Chemistry*. 1992;**38**(12):2449-2454
- [80] Kim YE, Hwang CJ, Lee HP, Kim CS, Son DJ, Ham YW, et al. Inhibitory effect of punicalagin on lipopolysaccharide-induced neuroinflammation, oxidative stress and memory impairment via inhibition of nuclear factor-kappaB. *Neuropharmacology*. 2017;**117**:21-32
- [81] Velagapudi R, Baco G, Khela S, Okorji U, Olajide O. Pomegranate inhibits neuroinflammation and amyloidogenesis in IL-1 β -stimulated SK-N-SH cells. *European Journal of Nutrition*. 2016;**55**(4):1653-1660
- [82] Braidy N, Essa MM, Poljak A, Selvaraju S, Al-Adawi S, Manivasagam T, et al. Consumption of pomegranates improves synaptic function in a transgenic mice model of Alzheimer's disease. *Oncotarget*. 2016;**7**(40):64589
- [83] Essa MM, Subash S, Akbar M, Al-Adawi S, Guillemin GJ. Long-term dietary supplementation of pomegranates, figs and dates alleviate neuroinflammation in a transgenic mouse model of Alzheimer's disease. *PLoS One*. 2015;**10**(3):e0120964
- [84] Forouzanfar F, Afkhami Goli A, Asadpour E, Ghorbani A, Sadeghnia HR. Protective effect of *Punica granatum* L. against serum/glucose deprivation-induced PC12 cells injury. *Evidence-Based Complementary and Alternative Medicine*. 2013;**2013**
- [85] Binyamin O, Larush L, Frid K, Keller G, Friedman-Levi Y, Ovadia H, et al. Treatment of a multiple sclerosis animal model by a novel nanodrop formulation of a natural antioxidant. *International Journal of Nanomedicine*. 2015;**10**:7165
- [86] Loren DJ, Seeram NP, Schulman RN, Holtzman DM. Maternal dietary supplementation with pomegranate juice is neuroprotective in an animal model of neonatal hypoxic-ischemic brain injury. *Pediatric Research*. 2005;**57**(6):858-864

- [87] West T, Atzeva M, Holtzman DM. Pomegranate polyphenols and resveratrol protect the neonatal brain against hypoxic-ischemic injury. *Developmental Neuroscience*. 2007;**29**(4-5):363-372
- [88] Afaq F, Malik A, Syed D, Maes D, Matsui MS, Mukhtar H. Pomegranate fruit extract modulates UV-B-mediated phosphorylation of mitogen-activated protein kinases and activation of nuclear factor kappa B in normal human epidermal keratinocytes. *Photochemistry and Photobiology*. 2005;**81**(1):38-45
- [89] Syed DN, Malik A, Hadi N, Sarfaraz S, Afaq F, Mukhtar H. Photochemopreventive effect of pomegranate fruit extract on UVA-mediated activation of cellular pathways in normal human epidermal keratinocytes. *Photochemistry and Photobiology*. 2006;**82**(2):398-405
- [90] Kasai K, Yoshimura M, Koga T, Arai M, Kawasaki S. Effects of oral administration of ellagic acid-rich pomegranate extract on ultraviolet-induced pigmentation in the human skin. *Journal of Nutritional Science and Vitaminology*. 2006;**52**(5):383-388
- [91] Türk G, Sönmez M, Aydın M, Yüce A, Gür S, Yüksel M, et al. Effects of pomegranate juice consumption on sperm quality, spermatogenic cell density, antioxidant activity and testosterone level in male rats. *Clinical Nutrition*. 2008;**27**(2):289-296
- [92] Azadzi KM, Schulman RN, Aviram M, Siroky MB. Oxidative stress in arteriogenic erectile dysfunction: Prophylactic role of antioxidants. *The Journal of Urology*. 2005;**174**(1):386-393
- [93] Forest C, Padma-Nathan H, Liker H. Efficacy and safety of pomegranate juice on improvement of erectile dysfunction in male patients with mild to moderate erectile dysfunction: A randomized, placebo-controlled, double-blind, crossover study. *International Journal of Impotence Research*. 2007;**19**(6):564-567
- [94] Vasconcelos LCS, Sampaio FC, Sampaio MCC, Pereira MSV, Higino JS, Peixoto MHP. Minimum inhibitory concentration of adherence of *Punica granatum* Linn (pomegranate) gel against *S. Mutans*, *S. Mitis* and *C. Albicans*. *Brazilian Dental Journal*. 2006;**17**(3):223-227
- [95] Badria FA, Zidan OA. Natural products for dental caries prevention. *Journal of Medicinal Food*. 2004;**7**(3):381-384
- [96] Menezes SM, Cordeiro LN, Viana GS. *Punica granatum* (pomegranate) extract is active against dental plaque. *Journal of Herbal Pharmacotherapy*. 2006;**6**(2):79-92
- [97] Sastravaha G, Gassmann G, Sangtherapitikul P, Grimm W-D. Adjunctive periodontal treatment with *Centella asiatica* and *Punica granatum* extracts in supportive periodontal therapy. *Journal of the International Academy of Periodontology*. 2005;**7**(3):70-79
- [98] César de Souza Vasconcelos L, MCC S, Sampaio FC, Higino JS. Use of *Punica granatum* as an antifungal agent against candidosis associated with denture stomatitis. *Mycoses*. 2003;**46**(5-6):192-196
- [99] Seeram NP, Lee R, Heber D. Bioavailability of ellagic acid in human plasma after consumption of ellagitannins from pomegranate (*Punica granatum* L.) juice. *Clinica Chimica Acta*. 2004;**348**(1):63-68

- [100] Pérez-Vicente A, Gil-Izquierdo A, García-Viguera C. In vitro gastrointestinal digestion study of pomegranate juice phenolic compounds, anthocyanins, and vitamin C. *Journal of Agricultural and Food Chemistry*. 2002;**50**(8):2308-2312
- [101] McDougall G, Fyffe S, Dobson P, Stewart D. Anthocyanins from red wine—their stability under simulated gastrointestinal digestion. *Phytochemistry*. 2005;**66**(21):2540-2548
- [102] McGhie TK, Walton MC. The bioavailability and absorption of anthocyanins: Towards a better understanding. *Molecular Nutrition and Food Research*. 2007;**51**(6):702-713
- [103] Mertens-Talcott SU, Jilma-Stohlawetz P, Rios J, Hingorani L, Derendorf H. Absorption, metabolism, and antioxidant effects of pomegranate (*Punica granatum* L.) polyphenols after ingestion of a standardized extract in healthy human volunteers. *Journal of Agricultural and Food Chemistry*. 2006;**54**(23):8956-8961
- [104] Seeram NP, Henning SM, Zhang Y, Suchard M, Li Z, Heber D. Pomegranate juice ellagitannin metabolites are present in human plasma and some persist in urine for up to 48 hours. *The Journal of Nutrition*. 2006;**136**(10):2481-2485
- [105] Cerdá B, Espín JC, Parra S, Martínez P, Tomás-Barberán FA. The potent in vitro antioxidant ellagitannins from pomegranate juice are metabolised into bioavailable but poor antioxidant hydroxy-6H-dibenzopyran-6-one derivatives by the colonic microflora of healthy humans. *European Journal of Nutrition*. 2004;**43**(4):205-220
- [106] González-Sarriás A, García-Villalba R, Núñez-Sánchez MÁ, Tomé-Carneiro J, Zafrilla P, Mulero J, et al. Identifying the limits for ellagic acid bioavailability: A crossover pharmacokinetic study in healthy volunteers after consumption of pomegranate extracts. *Journal of Functional Foods*. 2015;**19**:225-235
- [107] Wang R, Ding Y, Liu R, Xiang L, Du L. Pomegranate: Constituents, bioactivities and pharmacokinetics. *Fruit, Vegetable and Cereal Science and Biotechnology*. 2010;**4**(2):77-87
- [108] Vidal A, Fallarero A, Peña BR, Medina ME, Gra B, Rivera F, et al. Studies on the toxicity of *Punica granatum* L.(Punicaceae) whole fruit extracts. *Journal of Ethnopharmacology*. 2003;**89**(2):295-300
- [109] Patel C, Dadhaniya P, Hingorani L, Soni M. Safety assessment of pomegranate fruit extract: Acute and subchronic toxicity studies. *Food and Chemical Toxicology*. 2008;**46**(8):2728-2735
- [110] Heber D, Seeram NP, Wyatt H, Henning SM, Zhang Y, Ogden LG, et al. Safety and antioxidant activity of a pomegranate ellagitannin-enriched polyphenol dietary supplement in overweight individuals with increased waist size. *Journal of Agricultural and Food Chemistry*. 2007;**55**(24):10050-10054