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The Role of Hesperidin in Cancer Therapy

Nafla M Nizar, Deepa Manohar R, Shaiju S Dharan
Department of Pharmaceutics, Ezhuthachan College of Pharmaceutical Sciences, Neyyattinkara-695124, Thiruvananthapuram, Kerala, India

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ABSTRACT: Cancer refers to a group of diseases involving uncontrolled growth of abnormal cells with a potential to spread or invade to other parts of the body. Factors which can causes cancer in human body are Tobacco, excessive exposure to ultraviolet light, smoking, Alcohol consumption, diet, infections etc.Oxidative stress plays an important role in the pathophysiology of different types of cancer. Therefore the development of antioxidants as an effective therapeutic strategy against cancer. Flavonoids are potent antioxidants and which possess a wide variety of anticancer effects. An in vitro and in vivo study shows that many types of flavonoids have strong anticancer activities. Hesperidin, a flavanone glycoside obtained from citrus fruits with potent antioxidant and anticancer effects. The anticancer effects of hesperidin are associated with its anti-oxidant and anti-inflammatory activities. The current review emphasizes onto the anticancer effects of hesperidin and its molecular mechanisms.

KEYWORDS: Hesperidin, Anti-Cancer, Flavonoids.

I. INTRODUCTION

Cancer is one of the leading causes of death worldwide,which annually occurred in more than 12.7 million people in the world. In the most basic terms, cancer refers to a group of diseases involving uncontrolled growth of abnormal cells with a potential to spread or invade to other parts of the body. Prostate, lung, and colorectal cancers are the most common cancers among the men. In Women, the most common types of cancers are breast, colorectal, and lung cancers (1). In Children, the most common types are Leukemia, lymphoma, and brain tumors.Factors that influence the causes of cancer are Alcohol, Smoking and tobacco use, diet, Infections, excessive exposure to sunlight, obesity, etc. Till now, Cancer can be treated by surgery, radiation therapy, targeted therapy, chemo therapy and hormonal therapy (2).

Current research mainly focused on the promising effects of antioxidants for the treatment of various diseases (3). Anti-oxidants play a key role in prevention of cancer. Today, great attention has been paidto natural antioxidants due to their low adverse effects and high efficacy in comparison with synthetic antioxidants (4). The antioxidant activity of naturalproducts especially plants and their polyphenolic compounds havedifferent beneficial effects on human health (5, 6).

Flavonoids are potent antioxidants and which possess a wide variety of anticancer effects. Hesperidin, a flavanone glycoside obtained from citrus fruits with potent antioxidant and anticancer effects. Hesperidin hasproven therapeutic activities for cancer. In addition, evidence has suggested its promising role in inhibiting cancer cell proliferation (7). The main aim of this paper is to review the available scientific reports on hesperidin anticancer activities.

II. FLAVONOIDS

Flavonoids are polyphenolic low weight secondary metabolites formed from plants and extractedfrom almost all parts of plants.For the time being, greater than 8000 distinct flavonoids were identified, and that wide variety continuously will increase (8). Beginning from 1936, with the primary article about flavonoids bioactivity, several literature data has been published about their structure and characteristics in general related to anti oxidative activity (9).

Flavonoids commonly consumed in diets and have an essential role in human dietarynutrition with daily dietary supplements attaining as much as 500 mg. It offers health benefits via cell signaling pathways and antioxidant pathways (10).

Several researcheshave supported the antioxidant, anti-inflammatory, anti-diabetic, cardioprotective, anticancer, and anti-ageing effects of flavonoids families. Most currently nutritional phytochemicals had been studied and reported as preventive and may be usable towards most cancers through their antioxidant activities.

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Chemically, flavonoids have a common fundamental structure this is in a glycosylated or esterified form, includes carbon atoms assembled in two aromatic C6 rings which are linked through a C3 bridge, C6-C3-C6, accordingly forming a biphenyl propane structure with the central unit being a benzo-γ-pyrone. Multiple hydroxyl groups, sugar, oxygen, or methyl groups are connected to center structure. A group of flavonoids is differentiated in several classes according to the levels of unsaturation or oxidation of heterocyclicring which includes specific radical groups and the position of the link results in the formation of various flavonoid classes. The main flavonoid classes are: catechins, dihydroxychalcones, chalcones, flavanones, flavanols, flavonols, flavones, isoflavones, anthocyanidols, and aurones. Depending on the oxidation state of the heterocyclic ring, flavonoid classes areobtained primarily based on the structural properties of ring C. This wide range in flavonoids comes fromhydroxylation, glycosylation, methoxylation, or prenylation of the fundamental skeleton of flavonoids or additional radicalgroups (11).

A large variety of medicinal plants includes flavonoids,which have been mentioned by many authors as having antibacterial, anti-inflammatory, antiallergic, antimutagenic, antiviral, antineoplastic, antithrombotic and coronary heart disease actions (12). Overwhelmingly, pharmacological effects are associated with antioxidant activity of flavonoids, arising via their capacity to scavenge free radicals. While generated in excess, free radicals can harm biomolecules, and are consequently implicatedin the etiology of several diseases and ageing (13). Radicalscavenging through flavonoids takesplace via electron donation fromthe free hydroxyls at the flavonoid nucleus, with the formationof a much less reactive flavonoid aroxyl radical stabilized throughresonance and consequently playing a role in thepropagation of the radical-triggered damages in biologicalsystems. Antioxidant activity of flavonoids correlates properly with their physiological feature in vivo, because oxidative stress is understood to participate in the initial process of differentpatho-physiological events (14). Literature data report that flavonoids can prevent damage due to free radicals.

III. HESPERIDIN

Hesperidin was first isolated from the internal part of orange peels in 1828. Hesperidin is also known as Vitamin P (15). Hesperidin is a flavanone glycosides majorly found in epicarp, mesocarp, and endocarpof different Citrus species (16). In addition,Citrus juice is another supply of hesperidin (17). It's been isolated at large scale from the rinds of paradisi Macfad, Citrus sinensis and Citrus unshiu Marcovitch (18). It has beenreported from other Citrus species such as Citrus aurantium L. var.dulcis and Citrus reticulata Blanco (19). It's been demonstrated that the stages of hesperidin in albedo, membranes and pith of Citrus species is greater than in its juice vesicles and seeds (20). It has also been reported that levels of maturity of green fruits influences the stages of hesperidin (21). There may be a close correlation among hesperidin concentrations and seed germination that is confirmed that light exposure stimulates this compound production. Similarly, it's been recognized in different species of Fabaceae, Betulaceae, Lamiaceaeand Papilionaceae families (22,23). It has additionally been stated that hesperidin is determined withinside the bark of Zanthoxylum avicennae and Zanthoxylumcuspidatum (24). In Cynara species, hesperidin is observed as neohesperidin form. Hesperidin, a chemical constituent ofthe roots of Acanthopanax setchuenensis(25).

The content material of hesperidin in citrus fruits varies substantially with species, a part ofthe fruit itself, geographic sites of cultivation, and processing procedures (26).Generally, hesperidin content material is higher in citrus peel than in the different components of the citrus fruits. However lemon seeds include extrahesperidin than peel with the aid of using methanol extraction (27). Juice from pigmented citrus includes extra hesperidinthan that from non-pigmented citrus (28). It's far possibly thatimmature citrus can also additionally comprise greater hesperidin than ripen citrus does (29). Pasteurization with heat did not lower hesperidin content material in citrus juice at least stored at 4∘C forup to 12 days. Instead, hesperidin content material will increase followingpasteurization of citrus juice at 90∘C for 20 seconds (30).Hesperidin content ranges from 555 to 761 mg/L in single-strength juice and from 470 to 614 mg/L in concentrated juice, suggesting that processing process impacts hesperidin content in citrus juice (31). Further to citrus fruits, peppermint additionally comprises hesperidin, whose content material increases following UVB irradiation. Methanol extract of Porphyra dentata, edible seaweed, comprises 5% hesperidin (32).

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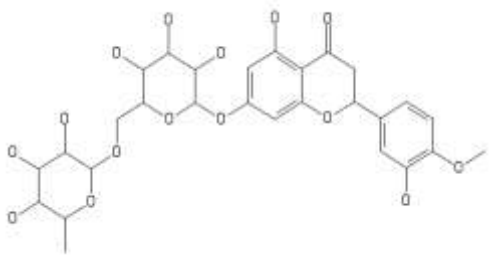
The conventional extraction method, with a totally low recovery, treats triturated peel with alkaline compounds to dissolve the flavonoids, which, after separation of solid residue, are precipitated through acidification. A modern technique to recover hesperidin from orange peel and waste water to make use of styrene-divinyl-benzene as the absorbing phase and eliminating the flavonoid with alcoholic NaOH. The effects can be a beneficial starting point for industrial applications (33).

IV. CHEMISTRY

On the chemical perspective, hesperidin structure harbors an aglycone referred to as alkyl eriodictyol (hesperetin) bonded to rutinose. Hesperidin contains a glycoside moiety that could be a disaccharide (glucose and rhamnose) that is present in two isomeric forms, i.e. neohesperidose and rutinose (34). Neohesperidose is chemically referred to as 2-O-alphal-Rhamnopyranosyl-d-glucopyranose. It's found to be present in citrus fruits within the form of hesperetin 7-O-neohesperidoside (35). Rutinose on the opposite hand, a disaccharide, typically obtained from plant sources is with chemicals 6-O-(-l-Rhamnosyl)-d-glucose. These oligosaccharide moieties are responsible for the bitterness of bioflavonoids. Of the disaccharides, the taste of citrus fruits is tasteless due to the presence of rutinosides moiety, whereas the neohesperidosides moiety is accountable for the bitter taste. Hesperidin is often found in neohesperidosides within the type of grapefruit (bitter) and in rutinoside within the type of orange (no bitter) (36). Within the chemical Hesperidin skeleton, hesperetin (aglycone part) is linked to aldohexose and rhamnose is further linked through aldohexose moiety.

Hesperidin is obtained from the alkaline reaction of hesperetenic acid and phloroglucinol, which throughout acid hydrolysis gets converted into hesperetin, l-rhamnose, and d glucose. The biological activity of Hesperidin is because of the presence of hydroxyl group in both the heterocyclic and aromatic rings (37). Further, there is a detailed relationship between the presence and variety of hydroxyl group moieties with the potential inhibitor capability of hesperidin. In addition, Hesperidin conjointly harbors neuro-protective actions and has the potential of penetrating the blood–brain barrier (38).



Figure(1): Structure of Hesperidin

V. BIOLOGICAL EFFECTS OF HESPERIDIN

Even though hesperidin has no normal structural elements to signify it as a great free radicals capturer and chelator, the capacity for chelating metallic ions was confirmed in our experiments (39, 40). Several other investigators have tested hesperidin antioxidant property and radical scavenging properties using a lot of assay systems (41, 42). Hesperidin reduces superoxide ions in electron switch plus concerted proton switch reaction in vitro. Further, Hesperidin turned into determined to be effective in protecting liposomes from UV irradiation brought about peroxidation, likely through scavenging the oxygen free radicals generated through UV irradiation (43). Several researches showed a potent bioactivity of hesperidin, such as effects on the vascular system, anti-inflammatory effects, antioxidant property, anti-carcinogenic activity, cell aggregation inhibition, anti-allergic effects, UV protecting activity, etc. (44).

Sl No	Mechanism	Activities
1	In comparison to the enalapril-hesperidin combination, Hesperidin and enalapril alone treatment exhibited similar effects, attenuating pathological changes, suppressing oxidative stress together with increasing PPAR-γ expression.	Cardioprotective

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2	Hesperidin mitigated the biomarker levels such as AST, ALT, and MDA, 88GSH, GPx, CAT, SOD which are opposite in nano zinc oxide induced hepatotoxicity.	Hepatoprotective
3	Patients have been observed with a significant difference in diastolic, mean arterial blood pressure, along with a reduction in TAC serum and anti-inflammatory markers suggesting anti-hypertensive and anti-inflammatory action	Anti inflammatory and antihypertensive
4	Depression cause increases inflammatory cytokines and oxidative markers and decreases BNDF levels in the hippocampus region. Hesperidin reversed these levels	Anti depressant
5	Enhancement in skin lesions of imiquimod-induced mice and hesperidin also prohibited lipopolysaccharide-induced human keratinocytes cell proliferation. In addition, it also reduced and normalized insulin and glucose levels, further formed to modulate the levels of leptin, adiponectin, and resistin. Inhibiting the initiation of the IRS-1/ERK1/2 signaling pathway	Anti dermatitis
6	Hesperidin moderately reversed the reduced phosphorylation of GSK-3β, Akt, lessened Nrf2 and increased expression of HO-1, phosphorylation of IκBα along with caused inhibition of RAGE expression and NF κB/p65	Neuroprotective
7	It considerably reduced the iron levels in the brain responsible for Parkinson disease, along with scavenging of reactive species, refining motor function, and cholinergic activity in the flies	Anti parkinsonism

VI. TOXICITY

Hesperidin is a bioflavonoid and there are few clinical reviews about adverse effects of hesperidin even with inside the pregnant women. It's also referred to as non-accumulative compound (45). According to animal study, phosphorylated hesperidin may be used as an antifertility compound and trauma, contamination and systemic illness can't suppress its antifertility activity (46).

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In some other scientific study, Daflon 500 mg (diosmin + hesperidin, twice per day for six weeks to one year) administration causes minor antagonistic impacts of patients compared with placebo group (47). Be that as it may, hesperidin can associated with common drugs that increase the uptake across the blood brain barrier. Hence, this consider recommended that maintaining a strategy from bioflavonoid containing foods (Hesperidin) during intake of P-glycoprotein substrates. Animal studies confirmed that phosphorylated hesperidin (4%) is secure and nontoxic and it is effortlessly acclimatized and caused no unfavorably susceptible responses. It has too been reported that oral administration of methyl hesperidin (5%) causes no mutagenic carcinogenic activities in mice. In expansion, hesperidin (0.3–5%) ingestion did not have unfavorable impacts on food intake and bodyweight. Another study confirmed that 13 weeks administration of methyl hesperidin (0.3–5%) did not cause mortality, weight loss or variations from the norm within the typical capacities of rats (48).

Hesperidin has great antioxidant activity and it indicates decreased lipid profile. Management of hesperidin averts oxidative pressure in liver and kidney tissues. Some of researchers have tested the antioxidant property and radical scavenging activity of hesperidin usage of quite a few assay systems. Remedy with hesperidin in iron-intoxicated rats protects the depletion of non-enzymatic antioxidants through its metallic chelating property and antioxidant property and might reduce the use of those antioxidants, hence restoring their levels. Administration of hesperidin decreased the histological changes triggered by iron. It may be attributed to the antioxidant and chelating capacity of hesperidin, which considerably decreased the oxidative threat leading to reduction of pathological changes and recovery of normal physiological functions (49).

VII. HESPERIDIN IN CANCER THERAPY

i. Hesperidin in cancer apoptotic cell death

A sort of cell death in which a series of molecular steps in a cell lead to its passing. This is often one strategy the body uses to get rid of abnormal cells. The method of apoptosis may be blocked in tumor cells. Moreover called modified cell death. Cancer is one of the scenarios where as well small apoptosis happens, coming about in malignant cells that will not pass on. The mechanism of apoptosis is complex and includes numerous pathways.

Hesperidin can actuate both apoptotic and autophagic cell death in colon carcinogenesis. Hesperidin actuates human colon cancer SNU-C4 cell apoptosis as determined by a reduction in messenger RNA expression of bcl-2 and an increment of bax mRNA levels with an increase of caspase-3 expression and movement. Cell death is a fundamental component to maintain tissue homeostasis and advancement, failure of which leads to different humandiseases like cancer (50). Apoptosis may be predominant sort of modified cell death and may be a major therapeutic strategy in chemotherapy and radiotherapy (51). Besides, apoptosis machinery comprises of two distinct pathways which incorporates intrinsic and extrinsic. Mitochondria zone control center of intrinsic pathway which has both pro-apoptotic(Bax/Bak) and anti-apoptotic (Bcl2/BclXL) individuals of regulatoryproteins. These two proteins work together and keep up mitochondrial membrane potential. Upon apoptotic incitement,MMP gets diminished and releases a few pro-apoptotic memberslike cytochrome c, smac/dialbo, endonuclease G, and apoptosisinducing factor to the cytosol and core. At last, caspase cascade gets actuated by caspase 3 which is dependable for cleavage ofcellular proteins that lead to cell death (52). On the opposite hand, extrinsic pathways happen when various death signals and ligands tie on the passing receptors of cellmembrane. It comes about in actuation of caspase cascades dependent(particularly caspase 8 & 3) apoptosis which is free of mitochondria.

ii. Hesperidin initiated apoptosis in Colon cancer

5-fluorouracil (5-FU) is the primary choice of cytotoxic chemotherapy for actuating apoptosis in patients with colon cancer. Though, defects in apoptosis are observed as chemo resistance of 5-FU treated colon cancer (53). Hesperidin was treated together with 5-FU on colon cancer WiDr cells, to assess whether it improves the viability of 5-FU in the treatment of colon cancer. Combination of hesperidin (200µm)and 5-FU (500µm) changed the morphology of WiDr cells and initiated apoptosis through chromatin condensation, formation of apoptoticbodies and cell cycle arrest. The possible mechanism in increasing the activity of 5-FA, may be that hesperidin expanded the transportof 5-FU into cells by changing membrane fluidity. The

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results advisedthe usage of hesperidin as a combination drug, so that the dosageand also the toxic effects of 5-FU can be reduced in colon cancerchemotherapy. The need of p53 in hesperidin interceded apoptosis in colon cancer cells. Anti-apoptotic impact ofhesperidin was evaluated in both p53 expressing (HCT116 p53+/+)and p53 Knockout (HCT116 p53−/−) cells. The results showed thatHsd altogether inhibited the development and actuate G1 cell cycle inp53 positive cells when compared to the p53 negative cell. Advancethe proapoptotic (Bax) and cyclin dependent kinase inhibitor(p21) was actuated by hesperidin as it were in p53 positive cells whichdemonstrated that Hsd requires useful p53 for their apoptotic effect. The chemo-preventive potential of hesperidin against Azoxymethane (AOM)initiated colon carcinogenesis in mice model (54).

Oral administrationof hesperidin (25 mg/kg body weight) in mice hindered the NF-κB (nuclear factor-kappa B) dependent inflammatory reactions comprising iNOS and COX-2 actuation.Further studies on explanation of molecular mechanism uncovered that hesperidin actuated apoptosis and cell cycle arrest in AOMinduced mouse. colon carcinogenesis through hindering P13K/Akt (Phosphoinositide 3-kinase/Protein kinase B) pathway and suppressingArurora-A and Akt interceded GSK-3β/β-catenin cascade. β cateninis an oncoprotein which play an essential part in AOM mediatedcolon tumor development in mice. Hesperidinessentially changed AOM mediated anti-apoptotic process by balancing the Bax: Bcl2 proportion as well as causing an increment incytochrome-c level and enactment of caspase 3 and 9. In expansion to that, hesperidin moreover expanded the expression level ofp53 & p21, in this manner, suppressing tumor. Moreover, hesperidinblocked the phosphoinositide-3-kinase (PI3K)/Akt signaling cascade by up-regulating the expression of PTEN- Phosphatase andtensin homolog (tumor suppressor protein) and repressing Aurora-A (upstream regulator of PI3K/Akt pathway). Hindrance of P13/Aktpathway actuates apoptosis conjointly represses tumor progression in tumor cells. Assist, hesperidin administration invigorated autophagic markers like Beclin-1 and LC3-II by restraining mTORprotein (mammalian target of rapamycin), which could be a key playerin autophagy mediated cell death, Also hesperidin anticipate the aggregation of β-catenin in cytoplasm by advancing the action of glycogen synthase kinase-3 beta (GSK-3β). Hesperidin plays major part in AOM mediated coloncarcinogenicity by blocking the Aurora-A mediated PI3K/Akt/GSK-3β and mTOR signaling pathways by fortifying apoptosis andautophagy mediated cell death in mice (55).

iii. Hesperidin initiated apoptosis in Gastric cancer

Hesperidin has been detailed to initiate apoptotic cell deathin different cancer cells by actuating both Extrinsic and intrinsic pathways (56). Hesperidin actuates particular intracellular death-receptor pathways in SNU-C4colon cancer cells by actuating DNA fragmentation and perinuclear apoptotic bodies' arrangement. Apoptosis was actuated typically through up regulation of Bax and Caspase-3 mRNA at the concentration of 100µM. Treatment of SNU-668HumanGastric cells with hesperidin (100µM) appeared apoptotic changeslike chromatin condensation, apoptotic morphology of cellularbodies, modulation of Bcl-2 and enactment of Caspase3, which recommended the possible utilization of hesperidin in gastric cancer patients (57).

iv. Hesperidin initiated apoptosis in Breast cancer

Evaluation of apoptotic activity of hesperidin in human mammary carcinoma cell line (Michigan cancer foundation-7 MCF-7) revealed that hesperidin (80µM) substantially triggered the shrinking of cells, vacuolation, and development of plasma membrane bleb sand cell detachment. Anti-cancer effect was also indicated by the other apoptotic characteristics like increase in LDH (lactated hydrogenase) level, depletion of GSH (Glutathione) level, DNA fragmentation, and accumulation of p53 protein and stimulation of caspase 3 protein (58).

v. Hesperidin initiated apoptosis in Lung cancer

Novel molecular design for hesperidin mediated apoptosis in MSTO-211H cells, which was applied as a model for malignant pleural mesothelioma, an uncommon type of cancer which affects the pleura of the lungs. Overexpression of the transcription factor Sp1 has been observed in many types of cancer, hence targeting of Sp1 may be a significant target for cancer therapy. The study showed that treatment of hesperidin at the concentration of 160µM at 48 h, significantly down-regulate the expression of Sp1 Transcription Factor in both mRNA and protein level and also modulate several Sp1 regulatory protein such

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asp27, p21, cyclin D1, Mcl-1 and surviving in MSTO-211H cells. Hesperidin also managed the expression of Bcl-2, Bax, caspase-3 and PARP, in MSTO-211H cells, which established the novel molecular mechanism of hesperidin i.e., Sp1 targeting in human malignant pleural mesothelioma (59).

vi. Hesperidin initiated apoptotic cell death in liver cancer

Though induction of cell death by proteolytic enzyme has been determined because the classical event in neoplastic cell death, proteolytic enzyme independentnon-apoptotic mechanisms like autophagy, mitotic catastrophe paraptosis etc., may mediate programmed necrobiosis in cancercells. Paraptosis, that could be a distinct mode of cell death, is elicited by hesperidin in HepG2 cells. Cytoplasmic vacuolization, mitochondria and endoplasmic reticulum swelling and uncondensedchromatin granule were determined in hesperidin treated cells within the concentration of one millimeter at 24 h, which suggests that hesperidin elicited paraptosis and not apoptosis in Hep G2. Hesperidin failed to have a bearing on the opposite characters of apoptosis additionally, together with chromatincondensation, DNA fragmentation, proteolytic enzyme activation, PARP cleavage and cathepsin D release. Since ERK1/2 protein kinase, whichis concerned in paraptosis is elicited by hesperidin in HepG2 cells; it should be probable that hesperidin may act as a promising therapeutic agent against liver disease by inducement non-apoptotic celldeath (60).

VIII. MECHANISM OF ACTION

i. Apoptotic and cell cycle arrest

Apoptosis induction and cell cycle arrest are among the foremost necessary mechanisms of Hesperidin action against cancer cells. Studies discovered that the pro-apoptotic action of Hesperidin is claimed to totally different enzyme pathways. For instance, suppression of AKT Serine/threonine kinase/ inhibitor of kappa light peptide factor in B-cells (PI3K/Akt/IKK) signal pathway in NALM-6 human pre-B cells by hesperidin were reportable to induce apoptosis. In malignant neoplastic disease cell line, MCF-7, hesperidin induced apoptotic events like phosphatidyl-serine externalization, DNAfragmentation, caspase-7 activation, and PARP (Poly (ADP-ribose) polymerase) cleavage, that are related to the activation of caspase-9, loss of mitochondrial membrane potential, release of hemoglobin, and enhance Bax:Bcl-2 ratio. More experiments discovered that hesperidin induced necrobiosis by accumulating reactive oxygen species (ROS) and activation of peptidase mediated apoptosis signal regulating kinase 1/ Jun N-terminal kinase (ASK1/JNK) pathway. Besides these, the most necessary mechanism of apoptotic impact of hesperidin through generation of ROS (61).

At high ROS levels, in conjunction with adenosine triphosphate (ATP) and calcium are liable for the induction of peptidase mediated necrobiosis by hesperidin in hepatocarcinoma cells through the activation of mitochondrial pathway. Similarly, in gastric cancer cells and esophageal cancer cells, hesperidin induces necrobiosis by increasing ROS and activating mitochondrial pathway. Caspase-dependent apoptosis induction by hesperidin in NCI-H358 and A549 non-small cell lung cancer (NSCLC) cells was additionally ascertained, whereas hesperidin was additionally found to be concerned in arrest cell cycle at G0/G1 phase via the down regulation of cyclinD1, and enhancing the expression of p21 and p53. Additionally, the endoplasmic reticulum stress pathway was additionally found to be concerned in a caspase-mediated cell death induction in HeLa (immortal cervical cancer cells) by hesperidin, along with arresting cell cycle at G0/G1 phase via the down regulation of cyclin D1, cyclin E1, and cyclin-dependent kinase 2 (Cdk2) at macromolecular level. The ROS-mediated caspase cell death by hesperidin in human gall bladder cancer was additionally reported however but they found cell cycle arrest at G2/M phase. The proof showed that hesperidin could be a potent inhibitor of calcium/calmodulin-dependent protein kinase IV (CAMKIV), and by inhibiting CAMKIV alongsidewith activating the caspase-3-dependent intrinsic pathway through the upregulation of pro-apoptotic macromolecule, Bax (BCL2 associated X, caspase-mediated cell death regulator), hesperidin exerts its anti-apoptotic and antitumor activities (62).

The in vitro activity of hesperidin in inducementcaspase mediated cell death andsensational cell cycle is in addition proved in vivo. For example,in azoxymethane-induced mouse model of carcinoma, hesperidin was found to vary the anti-apoptotic state of affairs by modulating Bax/Bcl-2 ratio, in conjunction with enhanced release of cytochrome-c and induction of caspase-3/9.Experimental studies discovered that hesperidin

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initiates necrobiosisby inhibiting the action of activated Aurora-A-mediated PI3K/Akt/GSK-3b pathway, and mTOR (mammaliantarget of rapamycin) pathway as well as the stimulationof autophagy throughout this malignant neoplastic disease model. Hesperidin along with fistein has been reportable to inhibit cellular proliferationthrough triggering programmed necrobiosis in humanK562 chronic myelocytic leukemia (CML) cells via activationof caspase-3 and JAK/STAT (Janus Kinase/Signaltransducer and activator of transcription pathway andgenes of JAK/STAT pathway have additionally known as candidates of CML therapy. Inelement cyanide cellulose ester (Fe-NTA)-induced urinary organ cancer model ofWistar rats, Hesperidin wasfound to initiate apoptosis-related proteins caspase-3, caspase-9, Bax expression and downregulation of Bcl-2 (BCL2necrosis regulator), NF-jB (nuclear factor kappa B subunit),iNOS (inducible nitric oxide synthase), TNF-a(tumor death factor-alpha), PCNA (proliferating cell nuclear antigen) expression, that were related to hesperidin anti-tumor activities in vivo. In transplant model of colon cancerous mice, Hesperidin also showed mitochondrial pathway-mediated necrobiosis induction and cell cycle arrest at G2/M phase (63).

ii. Anti-oxidant effect and anti inflammatory effects

Inflammation could be a complex physiological and biological process during which body fights toward the harmful stimuli like undigested particles, chemical irritants, damaged cells, moreover microorganisms and parasitic infections by overexpression of many cytokines, chemokines, and proinflammatory mediators, as well as TNF-a, COX-2, interleukin-1b, nitric oxide, prostaglandins, and eicosanoids. Except for the vigorous dependence of the expression level of pro-inflammatory mediators on the activation of numerous signaling pathways that may be regulated by various factors like mitogen-activated protein kinases, NF-jB, intercellular adhesion molecule-1), and VCAM-1, there's a good relationship between inflammation and production of ROS and reactive nitrogen species. Moreover, the mounting evidences indicate that inflammation and oxidative stress drive carcinogenesis by deactivation of growth suppressor genes, activation of oncogenes, and disruption of assorted cellular signal pathways. On the opposite hand, it's a large thought that hesperidin and its derivatives is thought of because the substantial and effective ancient flavonoids on oxidative stress and inflammation together with proliferation, apoptosis, DNA damage, free radicals, carcinogenesis, CV diseases, and hyperglycemia. The studies on anti-inflammatory mechanisms of hesperidin said that hesperidin can decrease the extent of inflammatory factors such as COX-2, PGE2 (prostaglandin E2), IL-4, IL-6, iNOS, and NO2 (64).

The anti-oxidative, chelating, and robust reducing properties together with the hydroxyl radical, peroxide, superoxide, and free radical scavenging activities of hesperidin occurred looking on the concentration and originated from the chemical structureby acting as a hydrogen donor to the radical molecules, and vie a radical target role to make new complexes between the inhibitor radicals and also the lipid radicals due to the presence of 300-hydroxy, 400-o-methoxy system within the B ring. The Hesperidin and hesperetin stimulate increase within the level and activity of the anti-oxidant enzymes may be achieved through nuclear factor erythroid 2-related factor 2 pathway, that is though because the major regulator of oxidative and electrophilic stress.Briefly, Hesperidin and hesperetin enhanced the expression of Nrf2, separate Keap1-Nrf2 complicated, and enhanced the nuclear translocation of Nrf2, and also the production of anti-oxidant enzymes is enlarged by the activation of gene transcription due to the binding of Nrf2 to the antioxidant response element at intervals within gene promoter region.Further, Hesperetin derivative-14 (HD-14) has additionally been reported to possess anti-inflammatory potential and has been shown to inhibit p-JAK1/p-STAT2 through PPAR-c upregulation in LPS-treated RAW264.7 cells.Same results were reported using another Hesperetin derivative-12 (HDND-12) in RAW264.7 cells and were reportable to down-regulate p-JAK2/p-STAT3 expression. Consequently, the derived perception from the revealed studies regarding hesperidin and its derivatives indicates that the antitumor activity of Hesperidin appears to be for the most part originatedfrom its anti-oxidant and anti inflammatory property (65).

iii. Anti-angiogenesis and anti-metastasis

In view of the constraints of presently marketed monoclonal antibodies and artificial compounds as anti-angiogenic agents regarding to toxicity and high cost sustained, natural compounds are being widely explored as potential anti-angiogenic agents. These natural products like flavonoids (e.g. hesperidin) modulate tumor

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angiogenesis through targeting vascular endothelial growth factor, matrix metalloproteinases (MMPs), basic fibroblast growth factor, epithelial cell proliferation, migration, and metastasis attributable to their anti-proliferative potential. The primary excerpts of the anti-metastatic and anti-proliferative result of hesperetin in B16-F10 pathological process murine melanoma cells in vitro and C57BL6/N mice in vivo. Then, anti-metastatic potential of Hesperidin in vitro in HepG2 human carcinoma cells wherever hesperidin suppressed emitted cytosolic MMP-9 expression through inhibition of activator protein-1, JNK signaling pathway and NF-jB signaling pathway. Hesperidin suppressed 12-Otetradecanoylphorbol-13-acetate (TPA)-induced cytosolic MMP-2 and MMP-9 and COX-2 expression through modulating NF-jB and AP-1 elicited growth cell invasion and metastasis in lung carcinoma and hepatocellular carcinoma. Hesperetin was achieved to inhibit transforming growth factor-b signaling pathway-induced tumor migration and metastasis through phosphorylation of Smad3 (66).

Inhibitory impact of hesperidin on tube shaped structure formation in human umbilical vascular endothelial cells (HUVECS) and mouse embryonic vegetative cell (mES)-derived endothelial cells through obstruction AKT and mTOR signal pathway that results in inhibition of cell migration, suppression of micro-vessel ontogeny, and tube formation in HUVECS. The anti-angiogenic impact of hesperidin in MCF-7 and HUVECs cells wherever hesperidin reserved migration and tube formation in the human carcinoma cells through down regulation of nuclear factor of activated T cells expression. In feminine BALB/c nude mice, a xenograft tumor model in vivo hesperidin suppressed tumor growth, decreased vascular density, reserved VEGF, and down regulated NFATc3, VEGF, and VEGFR2 expression through NFAT signaling pathway. Any analysis within the domain of anti-metastatic potential of Hesperidin in carcinoma has shown to focus MKK3/6 and p38 intracellular signaling pathways. In non-small cell carcinoma, Hesperidin was represented to considerably inhibit neoplasmmigration capability through targeting SDF-1a (stromal cell derived factor 1) resulting in down regulation of C-X-C chemokine receptor type 4, p-Akt, p-IjB (phosphorylated- I kappa B), and p-p65 expression (SDF-1/CXCR-4 signaling cascade).Hesperidin was conjointly documented to inhibit cell migration and invasion in the human sarcoma MG-63 cells via wound healing and matrigel assay and in vivo in male BALB/c transplant mice model (67).

Hesperetin administered together with platinum medication in vitro in A549 lung adenocarcinoma cells and in invivo in C57BL/6 mice inhibited growth proliferation and migration through targeting UDP-glucuronosyltransferse (UGT) family one member A3 (UGT1A3) a lot of considerably as compared to single drug treatment regime. In another study, combined administration of naringenin with hesperetin was shown to increase anti-metastatic result in Panc-1 human carcinoma cells through down regulation of FAK (focal adhesion kinase) and p38 signal pathway. The potential anti-angiogenic and anti-metastatic potential of hesperidin which can be used as a promising anti-cancer strategy for the management of human cancers while not eliciting harmful effects on surrounding normal cells (68).

IX. SYNERGISTIC IMPACT OF HESPERIDIN WITH DIFFERENT ANTI-CANCER AGENTS

Hesperidin, a flavanone, is present in numerous citrus fruits and possesses variety of biological activities. Hesperidin is known to possess potent anti-inflammatory, anti-carcinogenic, and anti-oxidant activities in numerous studies. The chemical structure of Hesperidin consists of hesperetin (methyl eriodictyol) certain to rutinose. The glycoside entity of hesperidin could be an oligosaccharide that contains rhamnose and aldohexose. The analysis in presenttimes on metastatic tumor activities of natural compounds has been targeted on induction of neoplastic cell death. These natural compound induces neoplastic cell death initiated by necrobiosis (type I programmed cell death), autophagic cell death (type II programmed cell death), and necroptosis (programmed necrosis).Hesperidin causes cell proliferation delay in numerous cancer models. Cell cycle arrest associated with cytostatic effects has been reportable in cells that have elevated p53 and cyclin dependent enzyme inhibitor levels, alongside lowered levels of cyclins and cyclin-dependent kinases (69).

X. SYNERGISTIC INTERACTION OF HESPERIDIN WITH VARIOUS COMPOUNDS

i. Doxorubicin

Doxorubicin is wide utilized employed in anti-tumour therapies. Inspite of the systematic

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therapy with doxorubicin, it provides solely peripheral enhancements in a very survival of the malignant hepatoma patients. The most mode of action of doxorubicin includes embolism inside DNA base pairs, therefore leading to resulting in DNA strand breakage and an inhibition of DNA and RNA synthesis by inhibition of topoisomerase II, leading to DNA damage and caspase mediated cell death induction. The doxorubicin additionally initiates the ROS generation therefore inflicting cell death. The application of apigenin and Hesperidin abroad doxorubin discovered the impact on doxorubicin-induced toxicity. These compounds altered the expression levels of glycolytic pathway genes – HK2 (hexokinase 2) and LDHA (lactate dehydrogenase A), that possess a serious role within the Warburg impact. The simultaneous administration of doxorubicin and apigenin or Hesperidin eradicated the injury with the simultaneous increase within the doxorubicin toxicity. In another study, impact of Hesperidin has been investigated together with doxorubicin to visualize its impact on doxorubicin-resistant MCF7 carcinoma cell lines. The cytotoxic effects were studied mistreat using MTT assay. Hesperidin combined with doxorubicin was unable to extend the apoptotic initiation however repressed the PgP (P-glycoprotein) expression that is especially responsible for developing the multidrug resistance once administration of doxorubicin (70).

ii. Tamoxifen

Tamoxifen could be a unremarkably used antineoplastic drug for estrogen receptor (ER)-positive breast carcinoma treatment. In numerous studies, it absolutely was shown that the some natural compounds, like Hesperidin, piperine (Pip) and bee venom (BV) have repressive impact effect on the breast cancer cells growth used on an individual basis. The combined impact of those natural compounds and Tam was investigated in a very study with a hypothesis that these compounds will increase the potential efficacy of growth repressive activity of Tam. The cytotoxic activity of Hesperidin, BV, and piperine was examined on MCF7 and T47D carcinoma cell lines via MTT assay and achieved equitable IC50 comparable to Tam results. The impact of various combinations was investigated and increased anti-proliferative result was obtained on MCF7 and T47D cell lines due to the synergistic impact. These natural compounds will synergistically increase the potential antineoplastic property of Tam against MCF7 and T47D cells most likely by causing the cell death, cell cycle size and EGFR, and Era down regulation (71).

iii. Quercetin

Etoposide, a by-product of natural product, podophyllotoxin. Etoposide acts as therapeutic agent in several types of cancer because of its ability to inhibit the topoisomerase II catalyst and induction of DNA breaks. In a study, the synergistic result of Hesperidin and quercetin was studied to boost oxidative damage caused by Etoposide on reproductive system in male rats. During this study, it absolutely was observed that administration of quercetin at concentration levels of twenty mg/kg body weight and Hesperidin at 25 mg/kg body weight for 2 months considerably increased sperm motility and count in experimental groups as compared to etoposide-treated group. This improvement in spermatozoan and count is attributed to the H2O2 scavenging by quercetin and Hesperidin, leading to inhibition of cellular DNA damage (72).

XI. NANO-TECHNOLOGY IN HESPERIDIN DELIVERY

Natural anti-oxidants like hesperidin have shown promising effect for the treatment of malignant growth and other different diseases because of high efficacy and lower side effects as compared to synthetic drugs. But hesperidin clinical use was extraordinarily restricted because of lower aqueous solubility and poor bioavailability. Thus there's need to overcome these problems for best use of this compound (73).

Nanotechnology is an interdisciplinary area of analysis having broad applications like molecular imaging, molecular diagnosis, and significantly targeted drug delivery. Further, this technology also overcomes the solubility and bioavailability problems with drugs as these parameters have tremendous impact on treatment of cancer. Therefore, many studies were initiated on developing hesperidin-based nanoparticles to increase the bioavailability, absorption, and bio-distribution of these flavonoids. Two Nanoformulations, i.e. hesperetin-TPGS (D-a-tocopheryl polyethylene glycol 1000 succinate) micelles and hesperetin-phosphatidylcholine complexes to increase the aqueous solubility, antioxidant activity, and oral absorption of hesperetin and led to enhancement of 16.2- and 18.0-fold in in-vitro antioxidant activity and in vivo oral absorption of hesperetin (74).

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XII. CONCLUSION

Hesperidin, a flavanone glycoside. It is vital Citrus bioflavonoids that have wide range of pharmacological activity. In present paper, we tend todemonstrate the anti-tumor effects of Hesperidin in several cancers with special emphasis on its molecular mechanism of action. The impact of hesperidin on various forms of cancer development has revealed thathesperidin modulates the various hallmarks of cancer specifically apoptosis, inflammation and oxidative stress mechanism. In addition,the chemo preventive mechanism of hesperidin has established that it promotes programmed cell death in cancer cells by numerous mechanisms including NF-κB, p53, PPARɣ, PI3K/AKT, mTOR, β-catenin and Aurora Akinase. Additionally, anti-inflammatory mechanism of hesperidin exhibit that it down-regulates several pro-inflammatory mediators and enzymes that are involved in carcinogenesis process likeTNF-α, IL-1β, IL-6, COX-2 and iNOS. Likewise, hesperidin also enhances the anti-oxidant defense mechanism by enhancing thelevel of enzymatic and non-enzymatic antioxidants. However, the valuablechemo preventive effect of hesperidin has been going through only in pre-clinical in vitro and in vivo cancer models. Combining data revealed that hesperidin plays multiple roles in itsanticancer effects. Nevertheless, respect to insignificant clinical trials on the useful role of hesperidin, it is very tough to make a cleardecision about most effective doses for its anticancer effects inhuman body. Finally we advised that future studies should emphasizeon:

i) Enhancing the bioavailability and absorption of hesperidin.
ii) Detecting the accurate molecular mechanisms of anticancer effects of hesperidin.
iii) Finding of simplest doses for future clinical trials on hesperidin.
iv) Assessing of anti-tumor effects of hesperidin in patients whosuffered from cancer.

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