(REVIEW ARTICLE)

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Natural products targeting the p53 tumor suppressor-A novel approach for future cancer therapy- A review

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Abstract

Mutations of p53, a tumor suppressor gene, are known to be involved in multiplication and metastasis of tumors. A number of natural products targeted the p53-MDM2 pathway. This review is an attempt to highlight the medicinal plants that can modulate the expression and activity ofp53tumor suppression, for cancer prevention and treatment.

Keywords: Tumor multiplication; Tumor metastasis; Natural; P53;Medicinal plants

1. Introduction

Human cancer is associated with alterations in a number of oncogenes and tumor suppressor genes that occur at various stages, from carcinogenesis to tumor growth, progression and metastasis.Proto-oncogenes normally control cell division and growth; molecular alterations, including gain-of-function mutations, amplification, and overexpression, can trigger the activation of oncogenes, leading to uncontrolled cell division, finally causing cancer [1-3].

Thep53 protein was identified in 1979. The human p53 gene is located on the seventeenth chromosome. The p53 tumor suppressor gene is the most frequently mutated gene in human cancer. The p53 protein is a phosphoprotein made of 393 amino acids. which grouped into six domains: a- the N-terminal region consists of the Transcription Activation Domain (TAD), which subdivided into two regions (TD_1 and TD_2), b- the Proline-Rich Region (PRR), which is constant in themajority of p53s, c- the central core domain (p53C), where the DNA-binding part is located and more than 90% of human mutations take place, d- the nuclear localization signal domain, e- the tetramerization (TET) domain, and e- the C-terminal domain (CT), which is the nonspecific DNA-binding domain [4-7].

Several signals can generate p53 within minutes, included ultraviolet radiation, and hypoxia, blockage of transcription, oncogene signaling, ionizing radiation, and lack of nucleotides. Then, p53 will induce cell cycle arrest, and apoptosis and block angiogenesis and DNA repair [8]. Ithas a number of other functions that recent data strongly implicate in tumor suppression, particularly with regard to the control of metabolism and ferroptosis (iron- and lipid-peroxide-mediated cell death) by p53. Wild-type (WT) p53 negatively regulates lipid synthesis and glycolysis in normal and tumor cells, and positively regulates oxidative phosphorylation and lipid catabolism. Mutant p53 in tumor cells does the converse, positively regulating lipid synthesis and glycolysis. The role of p53 in ferroptosis is even more complex: in normal tissues, WT p53 appears to positively regulate ferroptosis, and this pathway appears to play a role in the ability of basal, unstressed p53 to suppress tumor initiation and development [9-12]. In-vitro introduction of p53 in to p53-deficient cells has been shown to cause rapid death of cancer cells or prevention of further division. Various strategies have been proposed to restore p53 function in cancer cells [13]. The p53-MDM2 interactions have become the cornerstone of intensive cancer based research due to their effective anti-cancerproperties [14].This mini review is an attempt to highlight the medicinal plants that can modulate the expression and activity ofp53tumor suppressor.

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2. Medicinal plants modulated the expression and activity ofp53tumor suppressor

2.1. Allium sativum

The antiproliferative effects of diallyl sulfide (DAS), diallyl disulfide (DADS) and garlic extract were investigated on p53-wild type H460 and p53-null type H1299 non-small cell lung cancer cells (NSCLC). The DAS and DADS treatment of both H460 and H1299 cells resulted in the highest numbers of cells in apoptotic state as measured by acridine orange staining. DADS was found to be more effective in inducing apoptosis on NSCLC. The level of p53 protein in H460 cell was increased following DADS treatment. DAS and garlic extract treatment of H460 cells induced a rise in the level of Bax and a fall of Bcl-2 level [15-16].

2.2. Calendula Species

The hepatoprotective effect of calendula flowers and/or thyme leave extracts on aflatoxins (AFs)-induced oxidative stress, genotoxicity and alteration of p53 bax and bcl2 gene expressions were evaluated. Animals treated with the extracts 1 week before AFs treatment showed a significant decrease in oxidative damage markers, micro nucleated cells, DNA fragmentation and modulation of the expression of pro-apoptotic genes [17-18].

2.3. Capsicum Species

The effects of capsaicin on leukemic cells *in vitro* and *in vivo* and the molecular mechanisms of capsaicin-induced apoptosis were investigated in myeloid leukemic cells. Capsaicin suppressed the growth of leukemic cells, but not normal bone marrow mononuclear cells, via induction of G0-G1 phase cell cycle arrest and apoptosis. Capsaicin-induced apoptosis was in association with the elevation of intracellular reactive oxygen species production. Interestingly, capsaicin-sensitive leukemic cells were possessed of wild-type p53, resulting in the phosphorylation of p53 at the Ser-15 residue by the treatment of capsaicin. Abrogation of p53 expression by the antisense oligonucleotides significantly attenuated capsaicin-induced cell cycle arrest and apoptosis [19-20].

2.4. Carthamus tinctorius

The mechanism of regulating HIF-1alpha expression by hydroxysafflor yellow A (HSYA) in Eahy 926 cell line under 1% O2 hypoxia was studied. Eahy 926 cells were incubated with HSYA (100, 10 and 1 micromol x l-1) under hypoxia for the indicated time after treatment. HSYA at 100 micromol x l-1 increased Eahy 926 cells proliferation rate under hypoxia. HIF-1alpha mRNA and protein expression were up-regulated in the presence of HSYA. VHL, p53 mRNA and protein expression decreased significantly after 8 hours of treatment under hypoxia. Accordingly, HSYA protected Eahy 926 cells from hypoxia, and up-regulated HIF-1alpha expression partially via its inhibition of VHL and p53 expression [21-22].

2.5. Catha edulis

Catha edulis possessedp53 induction and cell cycle arrest, decreased mitochondrial function and activation of receptor- and mitochondria-mediated cell death pathways. Experiments using a p53 knock-down cell line and murine p53 knock-out bone marrow cells indicated that p53 was redundant in *Catha edulis*-mediated cell death *in vitro* [23].

2.6. Citrus aurantifolia

The bioactive compounds isolated from the seeds of *Citrus aurantifolia* were found to possess the potential of inhibiting human pancreatic cancer cells. While, the compounds purified from peel had the potential of suppressing the colon cancer cells. The purified compounds from seeds exhibited significant inhibition of Panc-28 cells with IC_{50} values in the range of 18.1-100 μM, which was confirmed by viable cell count. DNA fragmentation and expression of proteins in cells treated with compounds showed the induction of apoptosis through p53 and caspase-3 mediated pathway. The volatile oil showed 78 per cent inhibition of human colon cancer cells (SW-480) with 100 μg/ml concentration at 48 h. Lime volatile oil showed DNA fragmentation and induction of caspase-3 up to 1.8 and two folds after 24 and 48 h, respectively [24-25].

2.7. Corchorus Species

The effects of Saikosaponin-A on human breast cancer cell lines (MDA-MB-231 and MCF-7) were investigated. Results demonstrated that Saikosaponin-A inhibited the proliferation or viability of the MDA-MB-231 and MCF-7 cells in a dose-dependent manner. Saikosaponin-A treatment of MDA-MB-231 for 3 hours and of MCF-7 cells for 2 hours, respectively caused an obvious increase in the sub-G1 population of cell cycles. Apoptosis in MDA-MB-231 cells was independent of the P53/p21 pathway mechanism and was accompanied by an increased ratio of Bax to Bcl-2 and c-myc levels and

activation of caspase-3. In contrast, apoptosis of MCF-7 cells was initiated by the Bcl-2 family of proteins and involved p53/p21 dependent pathway mechanism, and was accompanied by an increased level of c-myc protein [26-27].

2.8. Crocus sativus

In order to examine saffron's anti-proliferative and pro-apoptotic effects in colorectal cancer cells, two p53 isogenic HCT116 cell lines (HCT wildtype and HCT p53-/-)were treated with different doses of the drug and analyzed cell proliferation and apoptosis in a time-dependent manner. Saffron extract induced a p53-dependent pattern of cell cycle distribution with a full G2/M stop in HCT116 p53 wildtype cells. However, it induced a remarkable delay in S/G2 phase transit with entry into mitosis in HCT116 p53 -/- cells. The apoptotic Pre-G1 cell fraction as well as Annexin V staining and caspase 3 cleavage showed a more pronounced apoptosis induction in HCT116 p53 wildtype cells. Obviously, the significantly higher DNA-damage, reflected by γH2AX protein levels in cells lacking p53, was coped by up-regulation of autophagy. The saffron-induced LC3-II protein level was a remarkable indication of the accumulation of autophagosomes, a response to the cellular stress condition of drug treatment [28-29].

MTT assay was performed to detect the inhibitory action of crocin on the proliferation of ovarian cancer HO-8910 cells. Flow cytometry was used to test the cell cycle distribution and apoptosis rate of ovarian cancer HO-8910 cells. Western blot analysis was utilized to measure the levels of apoptotic proteins such as p53, Fas/APO-1, and Caspase-3. MTT analysis revealed that crocin significantly inhibited the growth of HO-8910 cells. Additionally, flow cytometry illustrated that crocin raised the proportion of HO-8910 cells in the G0/G1 phase and increased their apoptosis rate. Furthermore, Western blot analysis revealed that crocin up-regulated the expression of p53, Fas/APO-1, and Caspase-3.Accordingly, crocin significantly inhibited the growth of HO-8910 cells and arrest them in the G0/G1 phase. Crocin also promoted ovarian cancer HO-8910 cell apoptosis, most likely by increasing p53 and Fas/APO-1 expression, and activating the apoptotic pathway regulated by Caspase-3 [30].

2.9. Curcuma longa

Curcumin50 μg/kg and 200 μg/kg(ip) every other day for 28 days, and diet containing curcumin at 0.6%; possessed antiproliferative effects and increased apoptosis and p53 expression in mice bearing malignant breast tumors [31-32].

2.10. Daucus carota

The anticancer activity of the pentane fraction (F1) and the 1:1 pentane: diethyl ether fraction (F2) of the *Daucus carota* oil extract was evaluated against human colon adenocarcinoma cell lines (HT-29 and Caco-2). Treatment of cells with various concentrations of F1 or F2 fractions produced a dose-dependent inhibition of cell proliferation. Flow cytometric analysis indicated that both fractions induced sub-G1 phase accumulation and increased apoptotic cell death. Western blot revealed the activation of caspase-3, PARP cleavage, and a considerable increase in Bax and p53 levels, and a decrease in Bcl-2 level. Treatment of HT-29 cells with either fraction markedly decreased the levels of both phosphorylated Erk and Akt [33-34].

2.11. Digitalis Species

Regarding the mechanisms of anticancer effects of cardiac glycosides, it appeared that digitoxin induced cell cycle arrest in G2/M phase via down-regulation of cyclin B1, cdc2 and surviving and increased the intracellular Ca^{2+} concentration. Digoxin increased intracellular Ca^{2+} concentration and induced DNA topoisomerases I and II and induced cell cycle arrest via the up-regulation of HIF-1α. OuabaindepletedNa$^+$/K$^+$- ATPase and up-regulated p21, increased intracellular Ca^{2+} concentration and inhibited DNA topoisomerases I and II. Oleandrin attenuated NF-kB, JNK and AP-1activation. Bufalin induced cell cycle arrest in G2/M phase via up-regulation of p21 WAF1 and p53 and the down-regulation of cyclin D, and inhibitedDNA topoisomerases I and II. Proscillaridin A, inhibited DNA topoisomerases I and II andincreasedintracellular Ca^{2+}[35-36].

2.12. Eucalyptus Species

Anticancer activities of *p*-menth-1-ene-4,7-diol (EC-1) isolated from *Eucalyptus camaldulensis* were studied on Ehrlich ascites carcinoma (EAC) cells. Anticancer activities also analyzed in EAC-bearing mice by assessment of cancer growth inhibition, changes in cancer volume, changes in life span, and hematological parameters. Apoptosis was analyzed by fluorescence microscope, DNA fragmentation assay, and flow cytometry. The expression of apoptosis-related genes, *Bcl-2*, Bcl-X, PARP-1, p53, and Bax, were analyzed using polymerase chain reaction (PCR).P-menth-1-ene-4, 7-diol (EC-1)significantly inhibited proliferation of EAC cells *in vivo* and restored the altered hematological parameters of EAC-bearing mice. Cytological observation by fluorescence microscope showed apoptosis of EAC cells upon treatment with EC-1. Also, DNA fragmentation assay revealed EAC cells' apoptosis following EC-1 treatment. Increased mRNA

expressions of p53 and Bax genes and negative expressions of Bcl-2 and Bcl-Xwere observed in cells treated with EC-1.MTT assay showed dose-dependent anticancer activity of EC-1 against EAC cell. Cell cycle analysis revealed that EC-1 treatment caused suppression of EAC cells at S phase [37-38].

2.13. Ficus religiosa

The anti-neoplastic potential of aqueous extract of *Ficus religiosa* bark was studiedin human cervical cancer cell lines, SiHa and HeLa. The aqueous extract of*Ficus religiosa* altered the growth kinetics of SiHa (HPV-16 positive) and HeLa (HPV-18 positive) cells in a dose-dependent manner. It blocked the cell cycle progression at G1/S phase in SiHa that was characterized by an increase in the expression of p53, p21 and pRb proteins with a simultaneous decrease in the expression of phospho Rb (ppRb) protein. In HeLa, aqueous extract of *Ficus religiosa* induced apoptosis through an increase in intracellular Ca^{2+} leading to loss of mitochondrial membrane potential, release of cytochrome-c and increase in the expression of caspase-3. The aqueous extract of*Ficus religiosa* also reduced the migration as well as invasion capability of both cervical cancer cell lines accompanied with down-regulation of MMP-2 and Her-2 expression. In addition, italso reduced the expression of viral oncoproteins E6 and E7 in both cervical cancer cell lines [39-40].

2.14. Gleditsia sinensis

*Gleditsia sinensis*thorn extract decreased cell growth and increased cell cycle arrest during the G2/M-phase of colon cancer cells. The arrest was correlated with increased p53 levels and down-regulation of cyclinB1 [41].

2.15. Glossostemon bruguieri

The antiproliferative effects of moghat root extract and its apoptotic mechanism were investigated in hepatocellular carcinoma (HCC) cells, HepG2 and Hep3B. MTT assay, morphological changes, apoptosis enzyme linked immunosorbent assay, caspase and apoptotic activation, flow cytometry, and immunoblot analysis were employed. The IC_{50} of moghat root extract for HepG2 was 910 ± 6 μg/ml and for Hep3B was 1510 ± 5 μg/ml, it induced significant growth-inhibitory effects against

HCC cells, with no cytotoxic effect on normal hepatocytes. Moghat root extract treatment induced apoptotic effects to HepG2 cells in a caspase dependent manner and via up-regulating p53/p21 and PCNA. The upregulation of p21 was controlled by p53 expression in HepG2 but not in Hep3B despite up-regulation of Bax protein in both cell lines. P21 may be a remarkable switch to G1 arrest in HepG2 cells, but not in Hep3B cells. Furthermore, Fas- and mitochondria-mediated pathways were found to be involved in moghat root extract -induced apoptosis in Hep3B cells [42-43].

2.16. Hamelia patens

Alkaloids: Rumberine, Alakaloid A, Isopteropodine, and Maruquine isolated from *Hamelia patens* were successfully dock inside the same active binding site of MDM2 protein where p53 peptide binds with a binding energy in a range of -7.42 to -6.79 Kcal/mol. Among the five tested compounds Palmirine has shown to be the best MDM2 inhibitor with a binding energy -7.42 Kcal/mol, whereas Maruquine compound showed the least binding affinity towards MDM2 with a binding energy -6.79 kcal /mol[44].

2.17. Hibiscus sabdariffa

Human gastric carcinoma (AGS) cells were susceptible to Hibiscus polyphenol-rich extract (0.95 mg/ml HPE inhibited its growth by 50%). AGS cells underwent DNA fragmentation, and had an increase in the distribution of hypo diploid phase (apoptotic peak, 52.36%) after a 24-h treatment with HPE (2.0 mg/ml). The effect of HPE on AGS cells might be mediated via p53 signaling and p38 MAPK/FasL cascade pathway, as demonstrated by an increase in the phosphorylation of p53 and the usage of a specific p38 inhibitor [45].

Hibiscus anthocyanins (Has) inhibited the serum-stimulated proliferation of smooth muscle cell (SMC) and resulted in cell apoptosis. The inducing cell apoptosis was dose dependent. HAs induced apoptosis via activating p38 MAP kinase that subsequently phosphorylates target protein c-Jun and transduces the signal to further activate the apoptotic protein cascades that contain Fas-mediated signaling (Fas/caspase-8 signaling module) and activating p53 and inducing bax expression [46-47].

2.18. Juglans regia

The effect of walnut green husk extracts on cell proliferation was evaluated on PC-3 human prostate cancer cells. Green husk extracts suppressed proliferation and induced apoptosis in a dose- and time dependent manner by modulating expression of apoptosis-related genes. This involved DNA fragmentation and significant changes in levels of mRNA and

the expression of corresponding proteins. An increase in expressions of Bax, caspase-3, and tp53 genes and their corresponding proteins was detected using real-time PCR and western blot analysis in PC-3 cells treated with the green husk organic extracts. In contrast, Bcl2 expression was down regulated after exposure to the extracts [48].

The antiproliferative effect of root bark of *Juglans regia* (RBJR) organic extracts was studied in cell proliferation on MDA-MB-231 human breast cancer cells. The results demonstrate that walnut root bark suppressed proliferation and induced apoptosis in a dose and time dependent manner by modulating expression of key genes. This involved characteristic changes in cytoplasmic and nuclear morphology, DNA fragmentation, levels of mRNA and expression of corresponding proteins. The expression of Bax, caspases, tp53, and TNF-alpha was markedly increased in MBA-MB-231 cells treated with the root bark extract. In contrast Bcl2 and mdm-2 expression was down regulated after exposure [49-50].

2.19. Juniperus communis

Imbricatolic acid isolated from the methanolic extract of the fresh ripe berries of *Juniperus communis* was evaluated for its ability to prevent cell cycle progression in p53-null CaLu-6 cells. It induced up-regulation of cyclin-dependent kinase inhibitors and their accumulation in the G1 phase of the cell cycle, as well as the degradation of cyclins A, D1, and E1[51].

The effects of juniper berry extract was evaluated on p53 protein, gene expression and DNA fragmentation in human neuroblastoma SH-SY5Y cells. The juniper berry extract activated cellular relocalization of p53 and DNA fragmentation-dependent cell death. Differentially expressed genes between treated and non-treated cells were evaluated with the cDNA-RDA method at the early time point of apoptotic process when p53 started to be activated. Twenty one over-expressed genes related to cellular stress, protein synthesis, cell survival and death were detected. They included Endoplasmic Reticulum (ER) stress inducer and sensor HSPA5 and other ER stress-related genes CALM2 and YKT6 indicating that ER stress response was involved in juniper berry extract mediated cell death. The authors suggested that juniper berry extract induced the p53-associated apoptosis through the potentiation and synergism by several phenolic compounds [52-53].

2.20. Lycium barbarum

A preparation of a carotenoid nanoemulsion of *Lycium barbarum* was evaluated to know the mechanism of inhibition on HT-29 colon cancer cells. Both the carotenoid nanoemulsion and the extract were effective at inhibiting growth of HT-29 colon cancer cells, with IC_{50} of 4.5 and 4.9 µg/ml, respectively. Also, both treatments up-regulated p53 and p21 expression and down-regulated CDK2, CDK1, cyclin A and cyclin B expression and arrest the cell cycle at G2/M [54].

Crude hot water extract of *Lycium barbarum* (2-5 g/l) dose dependently inhibited proliferation of H-4-II-E cells and HA22T/VGH cells ($p < 0.01$) after 24 h. The apoptosis was significantly increased in H-4-II-E cells after 24 h treatment with higher doses of crude hot extract (2-5 g/l) ($p < 0.01$). The expression of p53 protein in H-4-II-E cells was 119% and 143% for 2 and 5 g/l of hot extract of *Lycium barbarum* respectively, after 24 h, compared with control group [55].

The anti-proliferative effect of *Lycium barbarum* polysaccharide (LBP, 0, 10, 30, 100, and 300 µg/ml for 24 h.) was studied against MCF-7 cells. LBP arrested MCF-7 cell cycle in S phase. It also dose-dependently activated ERK, which may be associated with p53 pathway [56].

The hepatoprotective effects and mechanisms of LBP was studied in non-alcoholic steato-hepatitis (NASH)-induced hepatic injury in rat. Female rats were fed a high-fat diet to induce NASH with or without an oral 1 mg/kg LBP daily for 8 weeks compared with control rats, NASH rats showed increase in liver injury, lipid content, fibrosis, oxidative stress, inflammation and apoptosis. In contrast, NASH+LBP-co-treated rats showed improving of histology and free fatty acid levels, re-balance of lipid metabolism, reduction in profibrogenic factors through the TGF-β/SMAD pathway, improving of oxidative stress through cytochrome P450 2E1-dependent pathway, reduction in hepatic pro-inflammatory mediators and chemokines production and amelioration of hepatic apoptosis through the p53-dependent intrinsic and extrinsic pathways[57].

2.21. Marrubium vulgare

Marrubium vulgare ethanolic extract dose-dependently reduced viability of melanoma (B16) and glioma (U251) cells, but not peripheral blood mononuclear cells. It arrested cell cycle in S + G2/M phase, associated with activation of MAP kinase p38 and up-regulation of antiproliferative genes p53, p21 and p27. *Marrubium vulgare* ethanolic extract also induced mitochondrial depolarization, activation of caspase-9 and -3, Parp cleavage, phosphatidylserine exposure and DNA fragmentation. The mitochondrial apoptotic pathway was associated with the up-regulation of proapoptotic genes Pten, Bak1, Apaf1, and Puma and down-regulation of antiapoptotic genes survivin and Xiap. It also stimulated the

expression of autophagy-related genes Atg5, Atg7, Atg12, Beclin-1, Gabarab and Sqstm1, as well as LC3-I conversion to the autophagosome associated LC3-II. The most abundant phenolic components of *Marrubium vulgare* ethanolic extract (ferulic, phydroxybenzoic, caffeic and chlorogenic acids), did not exert a profound effect on viability of tumor cells, suggesting that other components were responsible for its cytotoxicity [58-59].

2.22. Momordica charantia

The effects of *Momordica charantia* leaf extract and a purified component, Kuguacin J (KuJ), were investigated on androgen-dependent LNCaP human prostate cancer cells. Both treatments exerted growth inhibition through G1 arrest and induction of apoptosis. KuJ markedly decreased the levels of cyclins (D1 and E), cyclin-dependent kinases (Cdk2 and Cdk4) and proliferating cell nuclear antigen, and caused an increase in p21 and p27 levels. Its induction of apoptosis was accompanied by an increase in cleavage of caspase-3 and poly (ADPribose) polymerase, attributed to augment of Bax/Bcl-2 and Bad/Bcl-xL and reduction of survivin levels. The extract and KuJ also reduced the expression of androgen receptor, prostate-specific antigen while induced P53 protein level. The extract and KuJ inhibited cell growth partly through p53-dependent cell cycle arrest and apoptotic pathways [60-61].

2.23. Myrtus communis

The ameliorate effect of *Myrtus communis* extract on the toxic effect of arsenic chloride (AsCl$_3$) on the production of the tumor suppressor protein (P53) was studied in rat. The results showed that AsCl3 induced negative effects on the P53-based gene expression levels and probably the protein activity of P53 gene resulting in low gene expression, while, *Myrtus communis* improved the status by increasing the levels of P53- based gene expression if used alone or mixed with AsCl$_3$ [62].

2.24. Nigella sativa

Thymoquinone inhibited doxorubicin-resistant human breast cancer MCF-7/DOX cell proliferation. It arrested MCF-7/DOX cells at G2/M phase and increased cellular levels of p53 and p21 proteins. A significant increase in Sub-G1 cell population and appearance of DNA ladders were recorded with thymoquinone treatment, indicating cellular apoptosis. Thymoquinone - induced apoptosis was associated with disrupted mitochondrial membrane potential and activation of caspases and PARP cleavage in MCF-7/DOX cells. Furthermore, it increased Bax/Bcl2 ratio via up-regulating Bax and down-regulating Bcl2 proteins [63].

Human breast cancer cells (MCF-7) were treated with methanolic extract of *Nigella sativa* seeds, the IC$_{50}$ of the extract was 62.8 µl/ml. When MCF-7 cells were exposed to 50 and 100 µl/ml extract for 24 h, 48 h and 72 h, the investigation revealed a dose- and time dependent increase in apoptosis. In addition, the expression of the caspase-3, -8, -9 and p53 genes was increased significantly according to the dose and time [64].

The anti-tumor effects of thymoquinone have also been investigated in tumor xenograft mice models for colon, prostate, pancreatic and lung cancer. The combination of thymoquinone and conventional chemotherapeutic drugs produced greater therapeutic effect and reduced the toxicity of the chemotherapy. The anticancer effects of thymoquinone were mediated through different modes of action, including antiproliferation, apoptosis induction, cell cycle arrest, ROS generation and anti-metastasis/anti-angiogenesis. It also exhibited anticancer activity through the modulation of multiple molecular targets, including p53, p73, PTEN, STAT3, PPAR-γ, activation of caspases and generation of ROS [65].

The growth-suppression potentiality of a crude saponin extract of *Nigella sativa* was investigated against human colon cancer cells, HCT116. The extract inhibited proliferation and induced apoptosis (the extract-treated cells exhibited morphological hallmarks of apoptosis including cell shrinkage, irregularity in cellular shape, cellular detachment and chromatin condensation). The pro-apoptotic effect of the extract was caspase-3-independent and associated with increase of the Bax/Bcl-2 ratio. It down-regulated transcriptional and DNA-binding activities of NF-κB and AP-1 proteins, associated with down-regulation of their target oncogenes, c-Myc, cyclin D1 and survivin. In addition, the extract up-regulated transcriptional and DNA-binding activities of Nrf2 and expression of cytoprotective genes. It also modulated the expression levels of ERK1/2 MAPK, p53 and p21 [66].

The anticancer effects of *Nigella sativa* extract was tested against human cervical cancer cells. The extract showed an 88.3% inhibition of proliferation of SiHa human cervical cancer cells at a concentration of 125 microl/ml methanolic extract at 24 h, (IC$_{50}$ value was 93.2 microl/ml). Extract exposure induced apoptosis in SiHa cell through both p53 and caspases activation [67].

2.25. Olea europaea

Oleuropein induced HeLa cells apoptosis as demonstrated by induction of a sub-G1 peak in flow cytometry and apoptosis-related morphological changes observed by fluorescence microscopy after being stained by Hoechst 33324. The results also showed that 150 - 200 µM oleuropein treated HeLa cells were arrested at the G2/M phase. The phosphorylated ATF-2, c-Jun NH2-terminal kinase (JNK) protein, p53, p21, Bax, and cytochrome c protein in the cytoplasm significantly increased in a dose dependent manner after treatment with oleuropein for 24 h[68].

3. Conclusion

Targeting the p53-MDM2 pathway can be a promising approach to develop compounds for cancer treatment and prevention. A number of natural products have been developed to target the p53-MDM2 pathway. The p53-MDM2 interactions have become the cornerstone of intensive cancer based research due to their effective anti-cancer properties. This review is an attempt to highlight the medicinal plants that can modulate the expression and activity ofp53tumor suppression, for cancer prevention and treatment.

Compliance with ethical standards

Acknowledgments

Disclosure of conflict of interest

The authors confirm that this paper's content has no conflict of interests.

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