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ANTIPROLIFERATIVE EFFECTS OF SOME MEDICINAL PLANTS ON COLORECTAL CANCER: A MINI REVIEW

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Abstract

Colorectal cancer (CRC) is one of the highly prevalent and deadly cancers worldwide. Surgery, chemotherapy and radiotherapy are the conventional treatment modalities for CRC; however, they cause adverse side effects and can be associated with cancer resistance and relapse. Currently, traditional herbal medicine is becoming increasingly studied and used in combination with conventional treatments in CRC. Various medicinal plants have shown promising anticancer effects against CRC concomitant with negligible side effects. For example, Annona muricata (soursop), Chenopodium quinoa (quinoa), Matricaria chamomilla (chamomile), and Moringa oleifera (moringa) have shown remarkable antiproliferative effects against CRC both in vitro and in vivo. Therefore, it is warranted to document the recuperative role of these potential medicinal plants in CRC. It is also important to provide scientific explanations for these effects by establishing their effects on key signaling pathways implicated in colorectal carcinogenesis. In this review, a summary of key signaling pathways that are altered in CRC is presented. The antiproliferative effects of selected medicinal plants against CRC as well as their underlying mechanisms of action are also discussed.

Keywords

Annona muricata, Chenopodium quinoa, Matricaria chamomilla, Moringa oleifera, colorectal cancer, medicinal plants

1. INTRODUCTION

Globally, colorectal cancer (CRC) is the third prevalent type of malignancy and the second cause of cancer-related mortality. In 2020, CRC was newly diagnosed among 1.9 million individuals and caused 0.9 million fatalities around the world (Xi & Xu, 2021). Based on disease incidence rates, it is predicted that the worldwide CRC burden will increase by 60% by 2030 (Arnold et al., 2017), and its cases will reach 3.2 million in 2040 (Xi & Xu, 2021). Regarding its etiology, CRC arises from a complex multi-step process that involves consecutive mutational events that sequentially drive the transformation of intestinal epithelial cells into malignancy (Mármol et al., 2017). By far, the most commonly occurring molecular events in CRC involve genes implicated in key signaling pathways that promote cellular proliferation, invasion and metastasis (Koveitypour et al., 2019). In fact, studying these pathways have paved the way for considering them as potential targets for various therapeutic approaches (Farooqi et al., 2019).

In general, CRC is conventionally treated by surgery, radiotherapy, chemotherapy, and targeted therapy (Ciombor & Goldberg, 2015). Although they show success in treating particular CRC cases, they are associated with a wide range of adverse events, limited efficacy, disease relapse and recurrence, and drug resistance (Nurgali, Jagoe, & Abalo, 2018). Meanwhile, substantial evidence suggests that the use of medicinal plants is associated with efficacy in preventing and/or treating diverse diseases, including CRC (Benarba & Pandiella, 2018). Recently, they are being extensively studied due to their minimal toxicity, bioavailability, affordability, and promising pharmacological activities (Gezici & Şekeroğlu, 2019). In this context, the current review aims at discussing the major signaling pathways implicated in CRC as well as highlighting the antiproliferative effect of selected medicinal plant extracts, namely *Annona muricata, Chenopodium quinoa, Matricaria chamomilla,* and *Moringa oleifera,* on CRC and their underlying mechanisms of action.

2. SIGNALING PATHWAYS IN CRC

Genetic and epigenetic changes lead to the progressive development of CRC (Aghagolzadeh & Radpour, 2016). In general, the genes involved in sporadic CRC belong to the Wnt, mitogen-activated protein kinase (MAPK)/ERK, phosphatidylinositol-3-kinase (PI3K)/Akt, Janus kinase (JAK)/signal transducer and activator of transcription (STAT), transforming growth factor beta (TGFβ), nuclear factor kappa B (NF-κB), and TP53 pathways (Kanthan, Senger, & Kanthan, 2012).

2.1. Wnt Pathway

The Wnt signaling pathway is a major cellular pathway that regulates embryonic morphogenesis as well as adult physiological processes (Yang, 2012). In the absence of Wnt ligand, β-catenin is sequestered in the destruction complex comprising of adenomatous polyposis coli protein (APC), scaffold protein Axin, and glycogen synthase kinase (GSK3β). The kinase in this complex phosphorylates β-catenin, causing its ubiquitinylation and subsequent degradation in the proteasome. On the other hand, Wnt binding to its frizzled receptor leads to the dissociation of the destruction complex, thereby causing the degradation of its components in the cytosol. In turn, β-catenin becomes stabilized in the cytosol, then it translocates to the nucleus, where it interacts with T-cell factor (Tcf) and lymphoid enhancer factor 1 (LEF1) to regulate the expression of cell cycle genes, such as *c-Myc* and *cyclin D1* (Ding & Ding, 2017). In CRC, the Wnt/β-catenin pathway is aberrantly activated due to significant mutations in the *ctnn1* oncogene that codes for β-catenin and in the tumor suppressor gene *APC*. Such mutations render APC incapable of binding β-catenin, which becomes stabilized in the cytoplasm (Polakis, 2012). Consequently, the signaling pathway becomes hyperactivated leading to continuous cell division and tumor growth. Therefore, the effective inhibition of this pathway is a promising way to prevent and treat CRC (Anastas & Moon, 2013).

2.2. MAPK/ERK Pathway

Receptor tyrosine kinases (RTKs) mediate intracellular signal transduction of MAPK pathways implicated in regulating differentiation, cellular proliferation, and metabolism (Cargnello & Roux, 2011). The ERK/MAPK pathway is initiated when RTKs become activated by extracellular stimuli, mainly platelet-derived growth factor (PDGF) and epidermal growth factor (EGF). Activated RTK binds to the growth factor receptor-bound protein 2 (Grb2), which in turn activates the protein son of sevenless (sos) (Meister et al., 2013). Sos facilitates Ras activation through the exchange of GDP by a GTP, and Ras GTP then activates the kinase protein Raf. Consequently, active Raf phosphorylates MAPK/ ERK kinase 1 and 2 (MEK1/2) which binds to ERK1/2, activates it, and releases it for target substrate phosphorylation in the cytosol or the nucleus, where it activates the transcription of *cMyc* and *cyclin D* (Lake et al., 2016). This pathway induces tumorigenesis in 30-50% of CRC cases (Roberts & Der, 2007). The nuclear translocation following ERK1/2 activation has been shown to promote CRC cell migration, survival, and invasion (Holck et al., 2016). Moreover, it regulates the protein and gene expression levels of FOXO3a, a crucial isoform of the fork head (FOX) transcription factors, which acts as a tumor suppressor in CRC (Grossi et al., 2019).

2.3. PI3K/Akt Pathway

PI3Ks are lipid kinases that phosphorylate different phosphatidylinositols (Engelman, Luo, & Cantley, 2006). Akt is a serine/threonine-specific protein kinase that is also known as protein kinase B (PKB) (Vivanco et al., 2002). The PI3K/Akt pathway is activated once ligands, such as PDGF or VEGF, bind to their respective tyrosine kinase receptors. This binding results in the recruitment of class IA PI3K to the cell membrane for the conversion of phosphatidylinositol-4,5-bisphosphate (PIP2) to phosphatidylinositol-3,4,5-trisphosphate (PIP3). The generated PIP3 acts as a second messenger that recruits Akt. Subsequently, Akt becomes phosphorylated by phosphoinositide dependent kinase 1 (PDK1) and mTORC2, and it phosphorylates and inactivates the complex of tuberous sclerosis 2 and 1 (TSC2-TSC1). This leads to the accumulation of GTP-bound Ras homologue enriched in brain (RHEB) and activation of mTORC1. Later, mTORC1 phosphorylates p70 S6 kinase and 4E-binding proteins leading to an increase in protein translation and ribosome biogenesis. The activation of this pathway leads to a variety of cellular functions, such as cell proliferation and survival, metabolism, and angiogenesis (Saxton & Sabatini, 2017). In CRC, the PI3K/Akt pathway is aberrantly activated inducing cell proliferation and cancer cell survival. In fact, mutations in genes that encode different components of this pathway occur in ~40% of CRCs (Johnson et al., 2010). Moreover, alterations in the PDGF signaling, especially in PDGFR-β receptor, have been observed in CRC, where the overexpression of PDGFRs results in enhanced angiogenesis, invasion, metastasis, and poor survival. Thus, the use of PDGFs/PDGFRs antagonists combined with therapeutic strategies is being increasingly used as an approach for cancers therapy (Narayanankutty, 2019).

2.4. JAK/STAT Pathway

The STAT family of proteins comprises seven members that play the same dual roles as signal transducers across the cytoplasm and as nuclear factors promoting transcription (Kiu & Nicholson, 2012). STATs were initially identified by their ability to relay signaling from interferons (IFN) α, β and γ, and IL-6 receptors upon binding to their corresponding cytokines (Aittomäki & Pesu, 2014). These cytokine receptors are associated with tyrosine kinases, especially the JAK family of kinases. Commonly, IL-6-mediated pathways lead to the activation of a JAK that activates the transcription factor STAT3 by phosphorylation (Bousoik & Aliabadi, 2018). When activated through ligand binding, JAK phosphorylates diverse tyrosine within the cytoplasmic regions of the receptor generating docking sites for STAT3. STAT3 is then phosphorylated and translocated to the nucleus to activate gene transcription (Aittomäki & Pesu, 2014). Particularly, STAT3 activation is well-documented for modulating a variety of target genes involved in cancer cells' survival, such as *Bcl-2*, *Survivin*, *Myeloid Cell Leukemia 1 (Mcl-1)*, *Bcl-xL*, *c-Myc*, *Cyclin D1*, *VEGF*, and *IL-6*

(Bousoik & Aliabadi, 2018). In CRC, IL-6/JAK/STAT3 is irregularly activated, allowing cancerous cells escape from the immune system. Moreover, elevated levels of phosphorylated STAT3 and IL-6 are correlated with tumor progression, invasion, angiogenesis, and metastasis (Wang et al., 2014).

2.5. TGF Pathway

The tumor growth factor beta (TGFβ) is a multipotent cytokine that regulates cell growth and differentiation through binding to its TGFβR receptors (Smith et al., 2012). The TGFβ superfamily comprises different cytokines, including TGFβ, activins, inhibins, bone morphogenetic proteins (BMPs), and growth differentiation factors (GDFs) (Zhang, 2017). The TGFβ1 ligand exists as a homo or heterodimer, and it is activated through proteolytic cleavage. Binding of TGFβ1 to the TGFβRII is facilitated by TGFβRIII. TGFβRI is then recruited and is phosphorylated by TGFβRII, inducing its kinase activity that phosphorylates Smad proteins (Xu et al., 2016). These proteins are divided into three classes: receptor-regulated Smads (R-Smad), the common mediator Smad (Co-Smad) and the inhibitory Smads (I-Smad). TGFβRI phosphorylates R-smads (Smad-2, Smad-4), which in turn form a complex with Smad-4 causing its translocation to the nucleus and its interaction with various DNA-binding co-activators, co-repressors and transcription factors. These interactions result in the regulation of different TGFβ responsive genes expression, mainly *p21* and *p14* that inhibit cyclin-dependent kinase causing cell cycle arrest in the G1 phase and suppression of *c-Myc* and *Bcl-2* expression (Zhang, 2017). The TGFβ signaling pathway plays a paradoxical role in the predisposition and prognosis of CRC. TGFβ is a potent inhibitor of normal colonic epithelial cells' transformation by acting as a tumor suppressor at the early stage of carcinogenesis (Cheruku et al., 2015). On the other hand, it promotes the survival, invasion and metastasis of CRC cells by acting as a tumor promoter during the late stage (Calon et al., 2012). Moreover, CRCs harbor somatic mutations in *TGFβRII* and *SMAD4* members of the TGFβ pathway (Villalba et al., 2017).

2.6. NF-κB Pathway

NF-κB is a key transcription factor that regulates the expression of many genes involved in inflammation, apoptosis and cell proliferation (Sun, 2012). Different stimuli activate the NF-κB signaling such as cytokines (TNFα, IL-6 and IL-1β), bacterial pathogens, viruses, ionizing radiation, and genotoxic agents. These stimuli initiate the signaling mostly through IκB kinase–dependent (IKK-dependent) phosphorylation and the ubiquitination and proteasomal degradation of IκB proteins (Sun, 2017). The generated NF-κB dimers translocate to the nucleus, where they regulate transcription of genes that encode chemokines, cytokines, growth factors, as well as cell adhesion molecules. The IKK complex is made up of IKKα and IKKβ subunits, and a non-enzymatic regulatory component, IKKγ/NEMO (Yu et al., 2020). In CRC, NF-κB promotes cancer progression through activating cell cycle regulators that make intestine epithelial cells proliferate rapidly. Also, NF-κB activates inflammatory cells and induces the expression of chemokines, angiogenic factors, as well as the pro-inflammatory enzymes cyclooxygenase 2 (COX-2) and inducible nitric oxide synthase (iNOS) (Soleimani et al., 2020; Vaiopoulos et al., 2013). The IKKβ-dependent NF-κB pathway provides a link between inflammation and CRC (Patel et al., 2018). Therefore, its suppression is associated with limiting the proliferation of cancer cells making it as an important therapeutic target in CRC (Soleimani et al., 2020).

2.7. P53 Pathway

P53 is a gene located on chromosome 17p; it encodes for a tumor suppressor protein that plays a crucial role in controlling the cell cycle. P53 activation triggers growth arrest at both cell cycle phases G1 and G2. P53 activation is also involved in interrupting DNA replication at S phase (Sionov et al., 2013). Moreover, p53 is a pro-apoptotic nuclear transcription factor and tumor inhibitor. It induces apoptotic cell death by activating several positive apoptosis regulators, such as Bax. Apoptosis mediated by p53 requires reactive oxygen species (ROS) generation and mitochondrial depolarization. These events

permit the release of caspase-9 and Apaf-1 which trigger apoptosis through the activation of the caspase cascade (Hafner et al., 2019). Besides, p53 regulates energy balance through activation of the AMPK pathway, cell differentiation, DNA repair, senescence, and angiogenesis. The activation and accumulation of p53 protein is triggered by a vast array of stress signals including DNA damage, hypoxia, viral infection, heat shock, and oncogenic activation (Boutelle & Attardi, 2021). In CRC, the deamination of methylated cytosine bases causes p53 mutations leading to transition mutations in C/T that are prevalent in ~60% of CRCs (Li et al., 2015). These mutations drive the transition from adenoma to adenocarcinoma and are associated with tumor development and chemoresistance (Hientz, et al., 2017). Gain of function (GOF) mutations in *p53* increase cell proliferation, migration and invasion (Michel et al., 2021).

3. MEDICINAL PLANTS AND CRC

Since the 1900s, scientists have been relying on plant sources to treat various forms of cancer. Among the anti-cancer drugs approved between 1940 and 2000, more than half of them were derived from plant-based natural products (Yuan et al., 2016). CRC has been extensively studied for being treated or prevented by medicinal plants. Despite the fact that it can be manageable by chemotherapy, this treatment option is associated with numerous side effects, thereby paving the way for alternative treatments. In this context, medicinal plants may serve as potent chemopreventive agents because they are rich reservoirs of bioactive molecules that exert cytotoxic and anti-proliferative effects (Benarba & Pandiella, 2018; Huang et al., 2019). Substantial evidence proves that extracts of medicinal plants alter distinct signaling pathways implicated in CRC (Martínez-Aledo et al., 2020). Following is a discussion of the anti-proliferative effects of selected medicinal plants of interest with particular emphasis on the targeted signaling pathways.

3.1. Annona Muricata

3.1.1 Botanical Description

Annona muricata L. (*A. muricata*), also known as soursop, guanabana, paw-paw, graviola, and sirsak, belongs to the Annonaceae family, order Magnoliales, division Magnoliophyta, and genus Annona, which includes approximately 70 species (Coria-Téllez et al., 2018). *A. muricata* is a tropical plant initially native to South and North America, but it was later introduced and cultivated in many tropical and subtropical regions worldwide including India, Malaysia, Nigeria, Cuba, Colombia, Peru, Madagascar, Cameroon, Togo, Ghana, and New Guinea (Adewole & Caxton-Martins, 2006). *A. muricata* is a green tree that is 5–10 meter tall with green colored edible fruit of white and creamy flesh. The fruit has an average weight of 0.4-1 Kg and may contain up to 170 seeds (Patel & Patel, 2016).

3.1.2 Pharmacological Activities

Extensive studies in the literature reported diverse biological activities of both aqueous and organic extracts of *A. muricata*, including anti-cancer, antioxidant, insecticidal, anti-parasitic, antibacterial, antiviral, and anti-inflammatory activities (Moghadamtousi et al., 2015; Gajalakshmi et al., 2012; Rady et al., 2018; Bitar et al, 2019). The anti-cancer effects of *A. muricata* extracts were reported against different types of cancers both *in vitro* and *in vivo.* Treatment with methanolic leaves extract (200mg/kg) for 5 weeks exerted *in vivo* cytotoxic effect against breast cancer through the downregulation of EGFR gene expression (Roham et al., 2016). In addition, treatment with aqueous leaves extract (30 mg/kg) displayed cytotoxic effect against breast, pancreatic, prostate, and skin cancers, *in vitro* and *in vivo* (Coria-Téllez et al., 2018; Rady et al., 2018).

3.1.3 Role in CRC

While the *in vitro* antiproliferative effects of *A. muricata* extracts against CRC were scarcely studied, some studies reported remarkable *in vivo* effects of this medicinal plant. In colon cancer cells (COLO-205), treatment with ethyl acetate leaves extract of *A. muricata* (10 µg/ml) induced a significant pro-apoptotic effect (Abdullah et al., 2017). As for the *in vivo* studies, treatment with ethanoic leaves extract (300 mg/kg) for 4 weeks reduced ACF formation (Eggadi et al., 2014) and induced apoptosis in AOM and DMH-induced CRC in rats (Moghadamtousi et al., 2015; Coria-Téllez et al., 2018).

Several recent studies investigated the potential preventive and therapeutic efficacy of aqueous *A. muricata* leaf extract (300 mg/kg; 5 days/week P.O.) on various signaling pathways involved in 1,2-dimethylhydrazine (DMH)-induced CRC in mice. In these studies, CRC was induced in mice using 20 mg/kg DMH for 12 and 24 weeks to study the effects of the prepared plant extract as a pre- and post-treatment. The first study was carried out by Masri (2021), who determined the effect of this extract on the Wnt/β-catenin signaling pathway and the levels of inflammatory enzymes COX-2 and iNOS. Findings revealed an increase in *Wnt, β-catenin, Tcf, Lef, c-Myc, Cyclin D1* gene expression levels, concomitant with increased COX-2 level and iNOS activity and a decrease in *APC* and *GSK3β* gene expression levels in mice with DMH-induced CRC. Treatment with *A. muricata* remarkably reversed these effects. Another study by Itani (2020) aimed at elucidating the effect of the same extract on IL-6-activated JAK/STAT3 pathway in the same model of CRC. Results demonstrated an increase in IL-6 and p-STAT3 levels in DMH-induced CRC tissues compared to the control group. *A. muricata* administration significantly reversed these levels when administered as pre-and post-treatment. Additionally, all target genes of the IL-6/JAK/STAT3 signaling pathway (*Mcl-1, Bcl-xL* and *Survivin*) which were upregulated in colorectal tissues of DMH-injected groups, were significantly minimized upon the administration of *A. muricata* extract.

Furthermore, Assaf (2020) assessed the effect of this extract on KRAS-activated ERK/MAPK pathway and its downstream regulators FOXO3a, c-fos, and cyclin-D1. Results showed that *A. muricata* extract, as a pre- and post-treatment, hindered CRC tumorigenesis represented by a reduction in polyp count. Treatments significantly decreased *KRAS* gene expressions and *p-ERK1/2* expressions in the nuclear fraction while they increased that in the cytoplasmic fraction. Also, the extract significantly decreased oncogenic *c-fos* and *cyclin-D1* gene expression with insignificant increase in the pro-apoptotic nuclear and cytoplasmic FOXO3a at the gene and the protein levels. Within the same model, Shmeas (2020) investigated the effect of *A. muricata* extract on the PI3K/Akt pathway. Results showed that *A. muricata* extract upregulated the expression of pro-apoptotic genes *PTEN* and *p53* while it downregulated that of anti-apoptotic genes *VEGF*, *Girdin* and *Bcl2*. Also, the extract significantly reduced the level of phospho-Akt. The inhibition of the PI3K/Akt pathway was more pronounced when the extract was administered as a post-treatment rather than a pre-treatment. Together, these studies suggest a preventive and therapeutic role of *A. muricata* aqueous extract against CRC, through the regulation of (1) the Wnt signaling pathway, (2) the RAS-activated ERK/MAPK pathway, (3) the IL-6-activated JAK/STAT3, and (4) the PI3K/Akt pathway as well as some of their downstream-regulated target genes (Fig. 1). Therefore, *A. muricata* aqueous extract may act as both a chemopreventive and a therapeutic natural product against CRC.

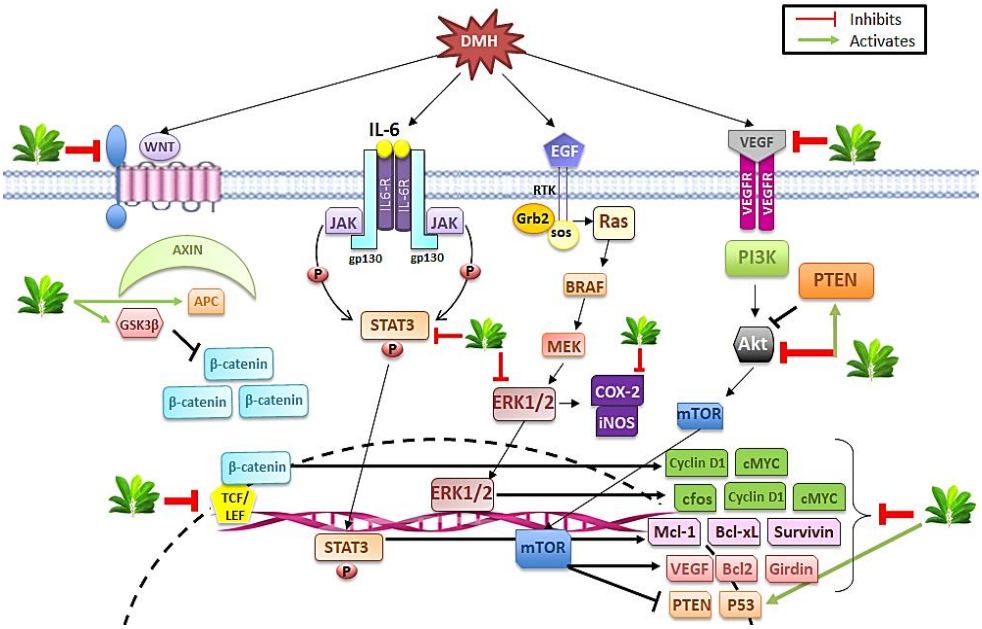

Fig.1: Effects of *A. muricata* extract on key signaling pathways involved in DMH-induced CRC.

3.2. Chenopodium Quinoa

3.2.1 Botanical description

Quinoa (*Chenopodium quinoa* Willd.) is a herbaceous annual plant that belongs to the Dicotyledoneae class, Chenopodiaceae family, Chenopodium genus, and quinoa species. It is about 1–2 meters high with hairy lobed leaves and a woody central stem that may be green, red or purple in color. The plant has flowering panicles arising from the top and having small sessile flowers colored same as the sepals. The seeds are ~ 2 mm in diameter of different colors, such as white, red or black (Maughan et al., 2007). Quinoa was indigenously cultivated in South America and was regarded as a sacred food, especially in the Lake Titicaca basin of Peru and Bolivia (Jacobsen, 2003). It can be cultivated at altitudes up 3500 m from sea level, and it shows tolerance to frost, salinity and drought. Nowadays, its cultivation has spread to more than 70 countries. Due to its increased popularity and consumption in North America, Europe, and Australia, the prices of quinoa tripled between 2006 and 2013 (Angeli et al., 2020).

3.2.2 Pharmacological Activities

Most of the studies on quinoa focus on its seeds' nutritional aspects, biodiversity and sustainability while only few address the pharmacological activities of its extract, especially those from its leaves. One study explored the antioxidant and immunomodulatory potential of the aqueous and ethanolic quinoa leaf extracts. Results revealed that ethanolic quinoa extract exerted potent antioxidant and anti-inflammatory effects. Also, it had a higher level of total phenolic content and a better ROS scavenging activity in the lipopolysaccharide (LPS)-induced murine macrophage RAW.264 compared to the aqueous extract (Chen et al., 2017). More importantly, phenolic compounds from *Chenopodium quinoa* leaves extract showed antiproliferative, antioxidant and antiangiogenic effects on prostate cancer cells (Gawlik-Dziki et al., 2013).

3.2.3 Role in CRC

Studies on the antiproliferative effect of *Chenopodium quinoa* leaves aqueous extract against CRC were done DMH model of CRC. A study by Mahmoud (2021) investigated the effect of quinoa extract (200 mg/kg, 5 days/week P.O.) on the Wnt pathway. Polyps count and the histopathological analysis revealed a significant anticancer effect of this extract when administered as a pre- and post-treatment. Quinoa extract significantly minimized alterations in Wnt, frizzled receptor (Fzd), β-

catenin, Tcf, Lef, cyclin D1, APC and GSK3β. It also caused a significant reduction in the levels of COX-2, iNOS and IL-6. Another study performed by Halawi (2021), in which the same quinoa extract and CRC model were used, aimed to unravel the effect of this extract on the PI3K/Akt/mTOR pathway. At the molecular level, the extract significantly modulated mRNA expression level of *PDGF-BB*, *PDFGR-β* and *mTOR* genes, which were altered by DMH injections. Moreover, the elevated levels of phospho-Akt upon DMH administration were reduced by quinoa pre- and post-treatment for 8 weeks. A similar pattern of increased levels of total and unphosphorylated NF-κB p65 was observed in the studied animals. More importantly, significant elevations in the level of phospho NF-κB p65 in mice receiving DMH were reduced upon pre- and 12 weeks post-treatment with quinoa extract. Taken together, these findings show that quinoa extract has the potential to prevent CRC through downregulating cell proliferation mediated by the Wnt pathway, reducing angiogenesis mediated by the PI3K/Akt pathway, and attenuating inflammation mediated by the NF-κB pathway (Fig. 2).

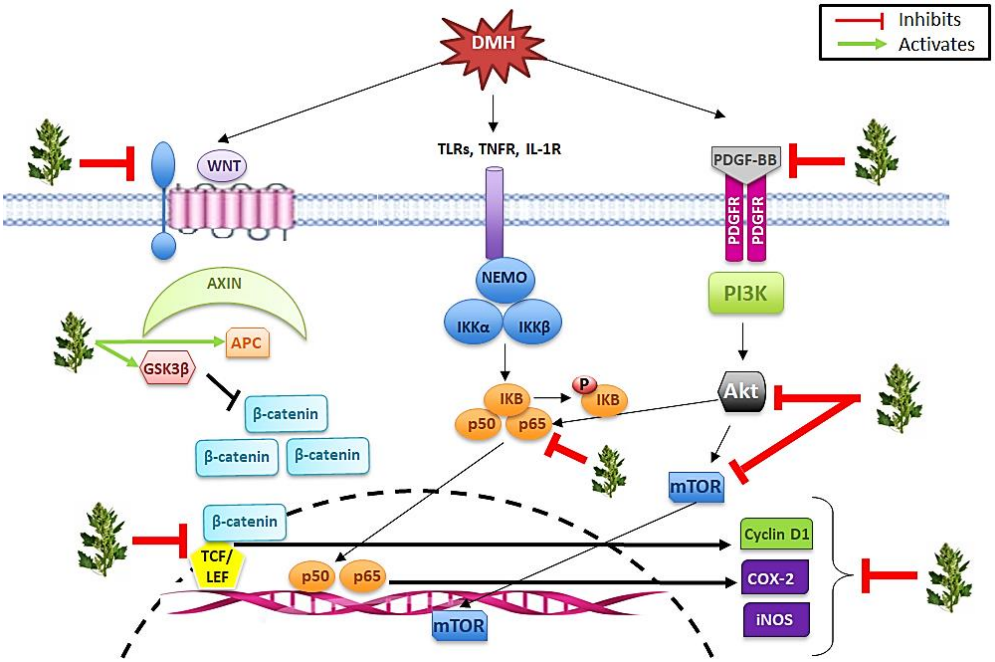

Fig.2: Effects of *C. quinoa* extract on some signaling pathways involved in DMH-induced CRC.

3.3. Matricaria Chamomilla

3.3.1 Botanical Description

Matricaria chamomilla L. (also known as German chamomile) is a valuable medicinal herb belonging to the daisy family (Asteraceae). It is traditionally used due to its profound health benefits (Singh et al., 2011). Its flower cones are hollow bright gold packed with tubular or disc florets ringed with ~ 15 white florets. It is native to Europe but is now spread in different regions throughout the world (Upadhyay et al., 2016).

3.3.2 Pharmacological Activities

The *in vitro* and *in vivo* pharmacological activities of *M. chamomilla* have been considerably studied in the literature (El Joumaa & Borjac, 2022). Several studies showed that the aqueous extracts of *M. chamomilla* exert significant antimicrobial effects against different Gram-positive and Gram-negative bacteria (Al-Ismail & Talal, 2003; Rahman & Chandra, 2015). Also, many research studies referred to the antioxidant effects of this herb through inhibiting the formation of ROS and enhancing the activity of antioxidant enzymes in different disease models (Cvetanović et al., 2015;

Jabri et al., 2016). Also, *M. chamomilla* extracts exert hypoglycemic and hypolipidemic effects in animal models of diabetes mellitus (Hajizadeh-Sharafabad et al., 2020). *M. chamomilla* extract exhibits remarkable sedative (Ross, 2013) and hepatoprotective activities (Aksoy & Sözbilir, 2012). Moreover, the anticancer activity of chamomile was pre-clinically proven against prostate, breast and ovarian cancer cell lines *in vitro* (Srivastava & Gupta, 2007).

3.3.3 Role in CRC

Studies assessing the anti-proliferative effects of *M. chamomilla* are few. A study by El Joumaa et al. (2020) aimed at deciphering the chemopreventive and anti-proliferative effects of aqueous *M. chamomilla* extract (150 mg/kg, 5 days/week P.O.) in DMH model of CRC. The extract suppressed colorectal polyps at the macroscopic and microscopic levels. At the molecular level, the administration of *M. chamomilla* extract as a pre-treatment was more effective that the post–treatment in modulating the expression levels of gene belonging to the Wnt signaling pathway, such as *Wnt, β-catenin, Tcf, Lef, APC,* and *GSK3β,* as well as its downstream genes *c-Myc* and *cyclin D1*. Using the same model and treatment strategy, Kawach (2018) assessed the effect of this extract on the TGFβ and NF-κB signaling pathways that are aberrantly altered in the DMH model of CRC. The extract down-regulated the gene expression levels of *TGFβ, Smad4,* and *p21* during the early stage of CRC. However, the gene expression levels of *TGFβ* and *TGFRII* were significantly upregulated during the late stage of carcinogenesis. As for the NF-κB signaling genes, *iNOS, COX-2* and *NF-κB* were significantly upregulated during early and late stages of CRC. In addition to being upregulated at the mRNA level, the activity of iNOS and protein level of COX-2 were found to be elevated in colonic tissues of mice receiving DMH (El Joumaa et al., 2020). In contrast, *M. chamomilla* treatment restored all these altered levels back to normal. Collectively, these studies showed that *M. chamomilla* aqueous extract exhibits anti-proliferative and anti-inflammatory effects against DMH-induced CRC through modulating the Wnt, TGFβ and NF-κB signaling pathways at early and late stages of carcinogenesis (Fig. 3).

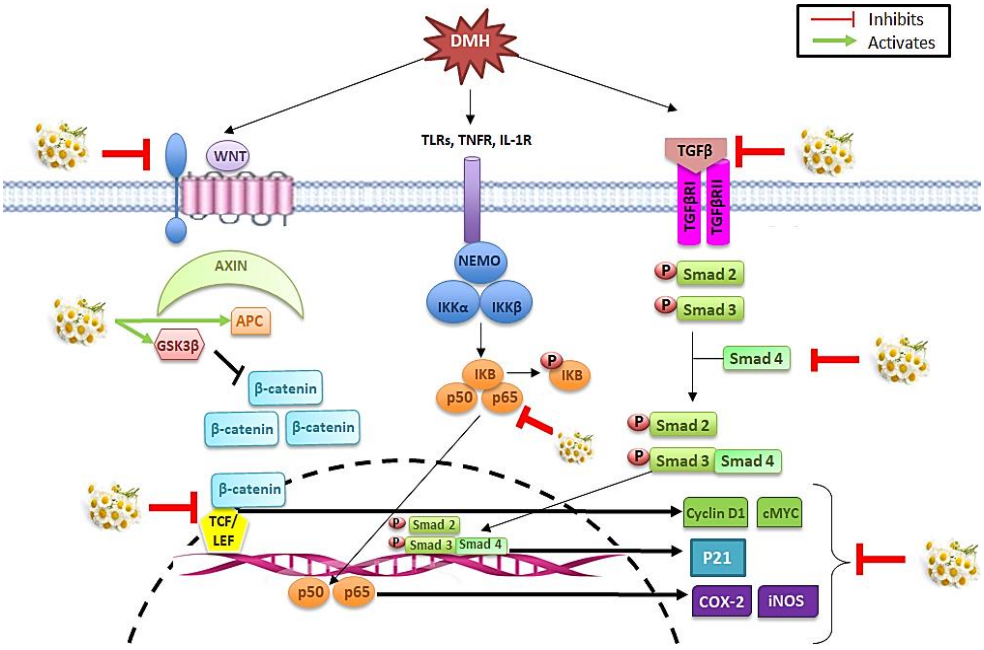

Fig.3: Effects of *M. chamomilla* extract on major signaling pathways involved in DMH-induced CRC.

3.4. Moringa Oleifera

3.4.1 Botanical Description

Moringa oleifera Lam is one of the thirteen best recognized species of Moringaceae family. The tree belongs to the Plantae kingdom, Magnoliophyta division, Magnoliopsida class, Brassicales order, Moringaceae family, Moringa genus, and oleifera species (Abdull Razis et al., 2014). *M. oleifera* tree is also known as the horseradish tree, drumstick tree, miracle tree, magic tree, and tree of life. The plant adapts to a variety of natural conditions, in which it thrives in dry to humid climates with annual rainfall ranging from 25-300 cm and temperatures between 19-28°C. *M. oleifera* is cultivated in different areas worldwide, including the western and sub-Himalayan tracts, India, Pakistan and Africa (Raja et al., 2016).

3.4.2 Pharmacological Activities

Extracts prepared from different parts of the *M. oleifera* tree have shown diverse pharmacological activities. *M. oleifera* leaves possess antioxidant properties against chemically-induced oxidative stress in mice (Verma et al., 2009). They also possess remarkable anti-inflammatory effects as they reduce the production of the cytokines TNF-α, IL-6 and IL-8 (Martínez-González et al., 2017). In addition, *M. oleifera* leaves extract exerts hepatoprotective effects against liver damage in mice (El-bakry et al., 2016). *M. oleifera* leaves extracts were shown to exhibit cardioprotective (Nandave, M., et al., 2009), nephroprotective (Akinrinde et al., 2020), hypolipidemic, and hypoglycemic effects (Chen et al., 2020) *in vivo*.

Extensive research has focused on the anticancer effects of *M. oleifera* leaves' extracts (Charoensin, 2014). Sreelatha et al. (2011) revealed that *M. oleifera* leaves extract inhibits the proliferation of human epithelial carcinoma cells (KB cell line). Other studies also confirmed the antiproliferative effects of *M. oleifera* leaves extracts on pancreatic cancer (Berkovich et al., 2013) and leukemia (Khalafalla et al., 2010) by downregulating the NF-κB signaling and inducing apoptosis, respectively. Moreover, Madi et al. (2016) reported the apoptotic effect of *M. oleifera* crude water extract on A549 lung cancer cells by inhibiting proliferation and increasing p53 and AIF levels as well as ROS production. Al-Asmari et al. (2015) reported that leaves extract of *M. oleifera* displayed antiproliferative effects in colon (HCT-8) and breast (MDA-MB-231) cancer cell lines. Similar findings were reported by Gaffar et al. (2019) in T47D breast cancer cell line.

3.4.3 Role in CRC

Reda et al. (2017) investigated the anti-cancer effect of 0.1-2.5% *M. oleifera* aqueous leaves extract (MOE) on different colorectal cancer cell lines, namely HCT116, CACO2, and HCT116P53$^{-/-}$. Results showed that MOE induced a dose and time-dependent decrease in viability (IC50 0.02-0.05%) and increase in ROS production. The extract also disrupted membrane integrity of cancer cells as evident by the release of lactate dehydrogenase upon MOE treatment. Another study that was also carried out by Reda et al. (2020) unraveled the mechanism of action of MOE extract. It showed that MOE induces the dissipation of mitochondrial membrane potential by increasing the intracellular oxidative stress, thereby causing a decrease in ATP level. In addition, an increase in preG0 and late apoptotic events were observed. In particular, the antiapoptotic effect of MOE extract was further corroborated by its ability to significantly modulating the altered expression levels of Bax, p53 and AIF in the studied CRC cell lines.

Similarly, studies by Pamok et al. (2012) reported a dose-dependent anticancer effect of aqueous and ethanol extracts (0.1-1 mg/ml) against HCT15, SW48 and SW480 CRC cell lines. Moreover, Al-Asmari and colleagues (2015) and Tragulpakseerojn et al. (2017) showed that *M. oleifera* aqueous extracts exert apoptotic effects against HCT-8 and HCT116 CRC cell lines. Several studies by Cuellar-Núñez et al. (2018, 2020, 2021) further showed that *M. oleifera* leaves

suppress CRC both *in vitro* and *in vivo* through mitigating inflammation and oxidative stress. In sum, these studies suggest that *M. oleifera* leaves extract possesses an anti-cancerous therapeutic potential against CRC (Fig. 4).

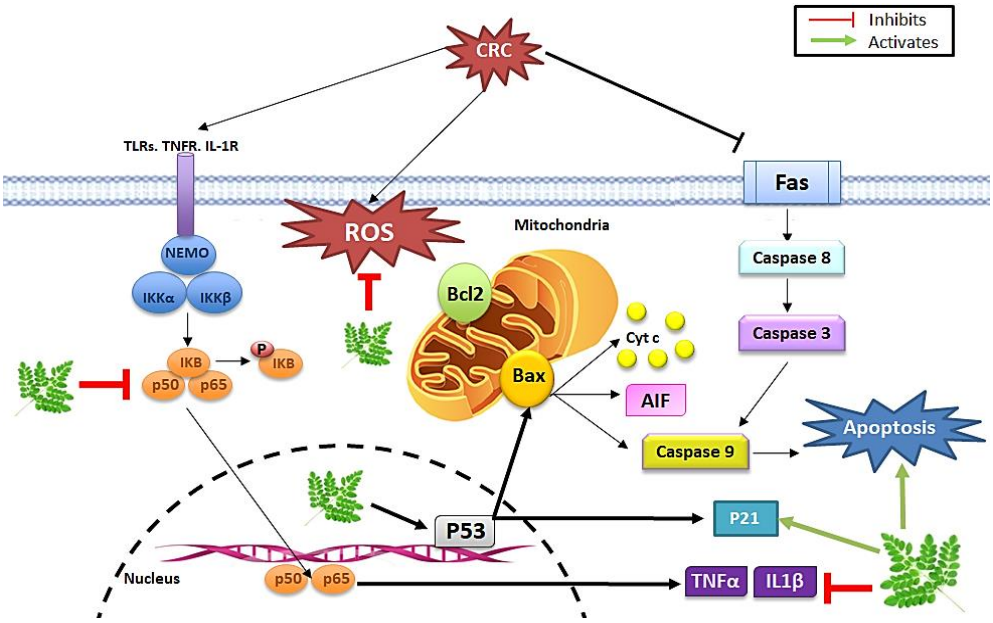

Fig.4: Effects of *M. oleifera* extract on major signaling pathways involved in CRC.

4. CONCLUSION

CRC is a prevalent disease that can be prevented and treated by some medicinal plants. Such plants possess potent antiproliferative effects, which are attributed to their richness in bioactive compounds. The presented medicinal plants herein, namely *A. muricata, C. quinoa, M. chamomilla*, and *M. oleifera*, have potent antiproliferative effects against CRC, suggesting their consideration as natural therapeutic choices for the treatment and chemoprevention of CRC. The present review suggests that these plants can be used in combination with chemotherapeutic drugs to minimize their side effects.

REFERENCES

- Abdull Razis, A. F., Ibrahim, M. D., & Kntayya, S. B. (2014). Health benefits of Moringa oleifera. *Asian pacific journal of cancer prevention*, *15*(20), 8571-8576.
- Abdullah, M., Syam, A. F., Meilany, S., Laksono, B., Prabu, O. G., Bekti, H. S., ... & Makmun, D. (2017). The value of caspase-3 after the application of Annona muricata leaf extract in COLO-205 colorectal cancer cell line. *Gastroenterology research and practice*, *2017*.
- Adewole, S. O., & Caxton-martins, E. A. (2006). Morphological changes and hypoglycemic effects of Annona muricata Linn. (Annonaceae) leaf aqueous extract on pancreatic β-cells of streptozotocin-treated diabetic rats. *African Journal of Biomedical Research 9*, 173–187.
- Aittomäki, S., & Pesu, M. (2014). Therapeutic targeting of the Jak/STAT pathway. *Basic Clin Pharmacol Toxicol*, *114*(1), 18-23.
- Akinrinde, A. S., Oduwole, O., Akinrinmade, F. J., & Bolaji-Alabi, F. B. (2020). Nephroprotective effect of methanol extract of Moringa oleifera leaves on acute kidney injury induced by ischemia-reperfusion in rats. *African Health Sciences*, *20*(3), 1382-1396.
- Aksoy, L., & Sözbilir, N. B. (2012). Effects of Matricaria chamomilla L. on lipid peroxidation, antioxidant enzyme systems, and key liver enzymes in CCl4-treated rats. *Toxicological & Environmental Chemistry*, *94*(9), 1780-1788.

- Al-Asmari, A. K., Albalawi, S. M., Athar, M. T., Khan, A. Q., Al-Shahrani, H., & Islam, M. (2015). Moringa oleifera as an anti-cancer agent against breast and colorectal cancer cell lines. *PloS one*, *10*(8), e0135814.
- Anastas, J. N., & Moon, R. T. (2013). WNT signalling pathways as therapeutic targets in cancer. *Nat Rev Cancer*, *13*(1), 11-26.
- Angeli, V., Miguel Silva, P., Crispim Massuela, D., Waleed Khan, M., Hamar, A., Khajehei, F., & Piatti, C. (2020). Quinoa (Chenopodium quinoa Willd.): An overview of the potentials of the "golden grain" and socio-economic and environmental aspects of its cultivation and marketization. *Foods*, *9*(2), 216.
- Arnold, M., Sierra, M. S., Laversanne, M., Soerjomataram, I., Jemal, A., & Bray, F. (2017). Global patterns and trends in colorectal cancer incidence and mortality. *Gut*, *66*(4), 683-691.
- Assaf, N. Y. (2020). *The effect of Annona muricata aqueous extract on EGFR signaling pathway in DMH-induced colon carcinogenesis in mice*. [Master's Thesis, Beirut Arab University, Lebanon].
- Benarba, B., & Pandiella, A. (2018). Colorectal cancer and medicinal plants: Principle findings from recent studies. *Biomedicine & Pharmacotherapy*, *107*, 408-423.
- Berkovich, L., Earon, G., Ron, I., Rimmon, A., Vexler, A., & Lev-Ari, S. (2013). Moringa Oleifera aqueous leaf extract down-regulates nuclear factor-kappaB and increases cytotoxic effect of chemotherapy in pancreatic cancer cells. *BMC complementary and alternative medicine*, *13*(1), 1-7.
-Bitar, R; Fahmi, R., Borjac, J. (2019). Annona muricata Extract Reduces Inflammation via Inactivation of NALP3 Inflammasome. *J. Natural Remedies. (19)*1, 12-23.
- Bousoik, E., & Montazeri Aliabadi, H. (2018). "Do we know jack" about JAK? A closer look at JAK/STAT signaling pathway. *Frontiers in oncology*, *8*, 287.
- Boutelle, A. M., & Attardi, L. D. (2021). p53 and tumor suppression: it takes a network. *Trends Cell Biol*, *31*(4), 298-310.
- Calon, A., Espinet, E., Palomo-Ponce, S., Tauriello, D. V., Iglesias, M., Céspedes, M. V. & Batlle, E. (2012). Dependency of colorectal cancer on a TGF-β-driven program in stromal cells for metastasis initiation. *Cancer cell*, *22*(5), 571-584.
- Cargnello, M., & Roux, P. P. (2011). Activation and Function of the MAPKs and Their Substrates, the MAPK-Activated Protein Kinases. *Microbiology and Molecular Biology Reviews*, *75*(1), 50–83.
- Charoensin, S. (2014). Antioxidant and anticancer activities of Moringa oleifera leaves. *J Med Plants Res*, *8*(7), 318-325.
- Chen, G. L., Xu, Y. B., Wu, J. L., Li, N., & Guo, M. Q. (2020). Hypoglycemic and hypolipidemic effects of Moringa oleifera leaves and their functional chemical constituents. *Food Chemistry*, *333*, 127478.
- Chen, H. L., Lan, X. Z., Wu, Y. Y., Ou, Y. W., Chen, T. C., & Wu, W. T. (2017). The antioxidant activity and nitric oxide production of extracts obtained from the leaves of Chenopodium quinoa Willd. *BioMedicine*, *7*(4).
- Cheruku, H. R., Mohamedali, A., Cantor, D. I., Tan, S. H., Nice, E. C., & Baker, M. S. (2015). Transforming growth factor-β, MAPK and Wnt signaling interactions in colorectal cancer. *EuPA Open Proteomics*, *8*, 104-115.
- Ciombor, K. K., Wu, C., & Goldberg, R. M. (2015). Recent therapeutic advances in the treatment of colorectal cancer. *Annual review of medicine*, *66*, 83-95.
- Coria-Téllez, A. V., Montalvo-Gónzalez, E., Yahia, E. M., & Obledo-Vázquez, E. N. (2018). Annona muricata: A comprehensive review on its traditional medicinal uses, phytochemicals, pharmacological activities, mechanisms of action and toxicity. *Arabian Journal of Chemistry*, *11*(5), 662-691.

- Cuellar-Núñez, M. L., de Mejia, E. G., & Loarca-Pina, G. (2021). Moringa oleifera leaves alleviated inflammation through downregulation of IL-2, IL-6, and TNF-α in a colitis-associated colorectal cancer model. *Food ResInt*, *144*, 110318.
- Cuellar-Núñez, M. L., Loarca-Pina, G., Berhow, M., & de Mejia, E. G. (2020). Glucosinolate-rich hydrolyzed extract from Moringa oleifera leaves decreased the production of TNF-α and IL-1β cytokines and induced ROS and apoptosis in human colon cancer cells. *Journal of Functional Foods*, *75*, 104270.
- Cuellar-Nuñez, M. L., Luzardo-Ocampo, I., Campos-Vega, R., Gallegos-Corona, M. A., De Mejía, E. G., & Loarca-Piña, G. (2018). Physicochemical and nutraceutical properties of moringa (Moringa oleifera) leaves and their effects in an in vivo AOM/DSS-induced colorectal carcinogenesis model. *Food Research International*, *105*, 159-168.
- Cvetanović, A., Švarc-Gajić, J., Mašković, P., Savić, S., & Nikolić, L. (2015). Antioxidant and biological activity of chamomile extracts obtained by different techniques: perspective of using superheated water for isolation of biologically active compounds. *Industrial Crops and Products*, *65*, 582-591.
- Ding, M., & Ding, M. (2017). Antagonism between Hedgehog and Wnt signaling pathways regulates tumorigenicity (Review). *Oncology Letters*, *14*, 6327-6333.
- Eggadi, V., Gundamedi, S., Sheshagiri, S. B. B., Revoori, S. K., Jupally, V. R., & Kulandaivelu, U. (2014). Evaluation of anticancer activity of Annona muricata in 1, 2-dimethyl hydrazine induced colon cancer. *World Appl Sci J*, *32*, 444–450.
- El Joumaa, M. M., & Borjac, J. M. (2022). Matricaria chamomilla: A valuable insight into recent advances in medicinal uses and pharmacological activities. *Phytochemistry Reviews*, 1-28.
- El Joumaa, M. M., Taleb, R. I., Rizk, S., & Borjac, J. M. (2020). Protective effect of Matricaria chamomilla extract against 1, 2-dimethylhydrazine-induced colorectal cancer in mice. *Journal of Complementary and Integrative Medicine*, *17*(3).
- El-bakry, K., Toson, E. S., Serag, M., & Aboser, M. (2016). Hepatoprotective effect of Moringa oleifera leaves extract against carbon tetrachloride-induced liver damage in rats. *World J Pharm Pharm Sci*, *5*(5), 76-89.
- Engelman, J. A., Luo, J., & Cantley, L. C. (2006). The evolution of phosphatidylinositol 3-kinases as regulators of growth and metabolism. *Nature Reviews Genetics*, *7*(8), 606-619.
- Farooqi, A. A., De La Roche, M., Djamgoz, M. B., & Siddik, Z. H. (2019). Overview of the oncogenic signaling pathways in colorectal cancer: Mechanistic insights. In *Seminars in cancer biology* (Vol. 58, pp. 65-79). Academic Press.
- Gaffar, S., Apriani, R., & Herlina, T. (2019). n-Hexane fraction of Moringa oleifera Lam. leaves induces apoptosis and cell cycle arrest on T47D breast cancer cell line. *Journal of Pharmacy & Pharmacognosy Research*, *7*(3), 173-183.
- Gajalakshmi, S., Vijayalakshmi, S., & Devi, R. V. (2012). Phytochemical and pharmacological properties of Annona muricata: a review. *International Journal of Pharmacy and Pharmaceutical Sciences*, *4*(2), 3-6.
- Gawlik-Dziki, U., Świeca, M., Sułkowski, M., Dziki, D., Baraniak, B., & Czyż, J. (2013). Antioxidant and anticancer activities of Chenopodium quinoa leaves extracts–in vitro study. *Food and Chemical Toxicology*, *57*, 154-160.
- Gezici, S., & Şekeroğlu, N. (2019). Current perspectives in the application of medicinal plants against cancer: novel therapeutic agents. *Anti-Cancer Agents in Medicinal Chemistry*, *19*(1), 101-111.
- Grossi, V., Fasano, C., Celestini, V., Lepore Signorile, M., Sanese, P., & Simone, C. (2019). Chasing the FOXO3: Insights into its new mitochondrial lair in colorectal cancer landscape. *Cancers*, *11*(3), 414.

- Hafner, A., Bulyk, M. L., Jambhekar, A., & Lahav, G. (2019). The multiple mechanisms that regulate p53 activity and cell fate. *Nature reviews Molecular cell biology*, *20*(4), 199-210.
- Hajizadeh-Sharafabad, F., Varshosaz, P., Jafari-Vayghan, H., Alizadeh, M., & Maleki, V. (2020). Chamomile (Matricaria recutita L.) and diabetes mellitus, current knowledge and the way forward: A systematic review. *Complement Ther Med*, *48*, 102284.
- Halawi, R. H. (2022). *The effect of quinoa on Pi3K/AKT/mTOR pathway in DMH induced colorectal cancer.* [Master's Thesis, Beirut Arab University, Lebanon].
- Hientz, K., Mohr, A., Bhakta-Guha, D., & Efferth, T. (2017). The role of p53 in cancer drug resistance and targeted chemotherapy. *Oncotarget*, *8*(5), 8921.
- Holck, S., Bonde, J., Pedersen, H., Petersen, A. A., Chaube, A., Nielsen, H. J., & Larsson, L.-I. (2016). Localization of active, dually phosphorylated extracellular signal-regulated kinase 1 and 2 in colorectal cancer with or without activating - BRAF and KRAS mutations. *Human Pathology*, *54*, 37–46.
- Huang, X. M., Yang, Z. J., Xie, Q., Zhang, Z. K., Zhang, H., & Ma, J. Y. (2019). Natural products for treating colorectal cancer: A mechanistic review. *Biomedicine & Pharmacotherapy*, *117*, 109142.
- Itani, Y. T. (2020). *Biochemical effect of Annona muricata extract on the JAK/STAT3 signaling pathway in colon carcinogenesis in mice.* [Master's Thesis, Beirut Arab University, Lebanon].
- Jabri, M. A., Sani, M., Rtibi, K., Marzouki, L., El-Benna, J., Sakly, M., & Sebai, H. (2016). Chamomile decoction extract inhibits human neutrophils ROS production and attenuates alcohol-induced haematological parameters changes and erythrocytes oxidative stress in rat. *Lipids in health and disease*, *15*(1), 1-10.
- Jacobsen, S. E. (2003). The worldwide potential for quinoa (Chenopodium quinoa Willd.). *Food reviews international*, 19(1-2), 167-177.
- Johnson, S. M., Gulhati, P., Rampy, B. A., Han, Y., Rychahou, P. G., Doan, H. Q., ... & Evers, B. M. (2010). Novel expression patterns of PI3K/Akt/mTOR signaling pathway components in colorectal cancer. *Journal of the American College of Surgeons*, *210*(5), 767-776.
-Kanthan, R., Senger, J.L. and Kanthan, S.C. (2012). Molecular Events in Primary and Metastatic Colorectal Carcinoma: A Review. Pathology Research International, 2012, Article ID: 597497.
- Kawach, R. M. (2018). *Effect of Matricaria chamomilla on a tumor suppressor gene and hepatic inflammation in mice with colon cancer.* [Master's Thesis, Beirut Arab University, Lebanon].
- Khalafalla, M. M., Abdellatef, E., Dafalla, H. M., Nassrallah, A. A., Aboul-Enein, K. M., Lightfoot, D. A., ... & El-Shemy, H. A. (2010). Active principle from Moringa oleifera Lam leaves effective against two leukemias and a hepatocarcinoma. *African Journal of Biotechnology*, *9*(49), 8467-8471.
- Kiu, H., & Nicholson, S. (2012). Biology and significance of the JAK/STAT signalling pathways. *Growth factors*, *30*(2), 88-106.
- Koveitypour, Z., Panahi, F., Vakilian, M., Peymani, M., Seyed Forootan, F., Nasr Esfahani, M. H., & Ghaedi, K. (2019). Signaling pathways involved in colorectal cancer progression. *Cell & bioscience*, *9*(1), 1-14.
- Lake, D., Corrêa, S. A. L., & Müller, J. (2016). Negative feedback regulation of the ERK1/2 MAPK pathway. *Cellular and Molecular Life Sciences*, *73*(23), 4397–4413.
- Li, X. L., Zhou, J., Chen, Z. R., & Chng, W. J. (2015). P53 mutations in colorectal cancer-molecular pathogenesis and pharmacological reactivation. *World journal of gastroenterology: WJG*, *21*(1), 84.
- Madi, N., Dany, M., Abdoun, S., & Usta, J. (2016). Moringa oleifera's nutritious aqueous leaf extract has anticancerous effects by compromising mitochondrial viability in an ROS-dependent manner. *J Am Coll Nutr*, *35*(7), 604-613.

- Mahmoud, N. K. (2021). *The effect of Chenopodium quinoa extract on 1, 2-DimethyIhydrazine induced colon cancer in rodents.* [Master's Thesis, Beirut Arab University, Lebanon].
- Mármol, I., Sánchez-de-Diego, C., Pradilla Dieste, A., Cerrada, E., & Rodriguez Yoldi, M. J. (2017). Colorectal carcinoma: a general overview and future perspectives in colorectal cancer. *International journal of molecular sciences*, *18*(1), 197.
- Martínez-Aledo, N., Navas-Carrillo, D., & Orenes-Piñero, E. (2020). Medicinal plants: active compounds, properties and antiproliferative effects in colorectal cancer. *Phytochemistry Reviews*, *19*(1), 123-137.
- Martínez-González, C. L., Martínez, L., Martínez-Ortiz, E. J., González-Trujano, M. E., Déciga-Campos, M., Ventura-Martínez, R., & Díaz-Reval, I. (2017). Moringa oleifera, a species with potential analgesic and anti-inflammatory activities. *Biomedicine & Pharmacotherapy*, *87*, 482-488.
- Masri, L. N. (2021). *The effect of Annona muricata leaves extract extraxt on Wnt signaling pathway in DMH induced colorectal cancer in mice*. [Master's Thesis, Beirut Arab University, Lebanon].
- Maughan, P. J., Bonifacio, A., Coleman, C. E., Jellen, E. N., Stevens, M. R., & Fairbanks, D. J. (2007). Quinoa (Chenopodium quinoa). In *Pulses, Sugar and Tuber Crops* (pp. 147-158). Springer, Berlin, Heidelberg.
- Meister, M., Tomasovic, A., Banning, A., & Tikkanen, R. (2013). Mitogen-Activated Protein (MAP) Kinase Scaffolding Proteins: A Recount. *International Journal of Molecular Sciences, 14*(3), 4854–4884.
- Michel, M., Kaps, L., Maderer, A., Galle, P. R., & Moehler, M. (2021). The role of p53 dysfunction in colorectal cancer and its implication for therapy. *Cancers*, *13*(10), 2296.
- Moghadamtousi, S. Z., Fadaeinasab, M., Nikzad, S., Mohan, G., Ali, H. M., & Kadir, H. A. (2015). Annona muricata (Annonaceae): A review of its traditional uses, isolated Acetogenins and biological activities. *Int J Mol Sci*, *16*(7), 15625-58.
- Nandave, M., Ojha, S. K., Joshi, S., Kumari, S., & Arya, D. S. (2009). Moringa oleifera leaf extract prevents isoproterenol-induced myocardial damage in rats: evidence for an antioxidant, antiperoxidative, and cardioprotective intervention. *Journal of medicinal food*, *12*(1), 47-55.
- Narayanankutty, A. (2019). PI3K/Akt/mTOR pathway as a therapeutic target for colorectal cancer: a review of preclinical and clinical evidence. *Current drug targets*, *20*(12), 1217-1226.
- Nurgali, K., Jagoe, R. T., & Abalo, R. (2018). Adverse effects of cancer chemotherapy: Anything new to improve tolerance and reduce sequelae?. *Frontiers in pharmacology*, *9*, 245.
- Pamok, S., Vinitketkumnuen, S. S. U., & Saenphet, K. (2012). Antiproliferative effect of Moringa oleifera Lam. and Pseuderanthemum palatiferum (Nees) Radlk extracts on the colon cancer cells. *J Med Plant Res*, *6*(1), 139-145.
- Patel, M. S., & Patel, J. K. (2016). A review on a miracle fruits of Annona muricata. *J Pharmacogn Phytochem*, *5*(1), 137.
- Patel, M., Horgan, P. G., McMillan, D. C., & Edwards, J. (2018). NF-κB pathways in the development and progression of colorectal cancer. *Translational Research*, *197*, 43-56.
- Plotnik, J. P., Budka, J. A., Ferris, M. W., & Hollenhorst, P. C. (2014). ETS1 is a genome-wide effector of RAS/ERK signaling in epithelial cells. *Nucleic Acids Research*, *42*(19), 11928–11940.
- Polakis, P. (2012). Wnt signaling in cancer. *Cold Spring Harb Perspect Biol*, *4*(5).
- Rady, I., Bloch, M. B., Chamcheu, R. C. N., Banang Mbeumi, S., Anwar, M. R., Mohamed, H., & Chamcheu, J. C. (2018). Anticancer properties of Graviola (Annona muricata): a comprehensive mechanistic review. *Oxid Med Cel. Longev*, *2018*.
- Raja, R. R., Sreenivasulu, M., Vaishnavi, S., Navyasri, D. M., Samatha, G., & Geethalakshmi, S. (2016). Moringa oleifera-An overview. *RA J Appl Res*, *2*(9), 620-4.

- Reda, F., Borjac, J., & Usta, J. (2020). Moringa oleifera leaves aqueous extract induce apoptosis in HT29 cell line. *BAU Journal-Science and Technology*, *2*(1), 7.
- Reda, F., Borjac, J., Fakhouri, R., & Usta, J. (2017). Cytotoxic effect of Moringa oleifera on colon cancer cell lines. *Acta horticulturae*, *1158*, 45-54.
- Roberts, P. J., & Der, C. J. (2007). Targeting the Raf-MEK-ERK mitogen-activated protein kinase cascade for the treatment of cancer. *Oncogene*, *26*(22), 3291–3310.
- Roham, P. H., Kharat, K. R., Mungde, P., Jadhav, M. A., & Makhija, S. J. (2016). Induction of mitochondria mediated apoptosis in human breast cancer cells (T-47D) by Annona reticulata L. leaves methanolic extracts. *Nutrition and cancer*, *68*(2), 305-311.
- Ross, S. M. (2013). Generalized anxiety disorder (GAD): efficacy of standardized Matricaria recutita (German chamomile) extract in the treatment of generalized anxiety disorder. *Holistic Nursing Practice*, *27*(6), 366-368.
- Saxton, R. A., & Sabatini, D. M. (2017). mTOR signaling in growth, metabolism, and disease. *Cell*, *168*(6), 960-976.
- Shmeas, S. (2020). *Potential role of Annona muricata extract on PI3K/Akt signaling pathway in cancer of the digestive system.* [Master's Thesis, Beirut Arab University, Lebanon].
- Singh, O., Khanam, Z., Misra, N., & Srivastava, M. K. (2011). Chamomile (Matricaria chamomilla L.): an overview. *Pharmacognosy reviews*, *5*(9), 82.
- Sionov, R. V., Hayon, I. L., & Haupt, Y. (2013). The regulation of p53 growth suppression. In *Madame Curie Bioscience Database [Internet]*. Landes Bioscience.
- Smith, A. L., Robin, T. P., & Ford, H. L. (2012). Molecular pathways: targeting the TGF-β pathway for cancer therapy. *Clinical Cancer Research*, *18*(17), 4514-4521.
- Soleimani, A., Rahmani, F., Ferns, G. A., Ryzhikov, M., Avan, A., & Hassanian, S. M. (2020). Role of the NF-κB signaling pathway in the pathogenesis of colorectal cancer. *Gene*, *726*, 144132.
- Sreelatha, S., Jeyachitra, A., & Padma, P. R. (2011). Antiproliferation and induction of apoptosis by Moringa oleifera leaf extract on human cancer cells. *Food and Chemical Toxicology*, *49*(6), 1270-1275.
- Srivastava, J. K., & Gupta, S. (2007). Antiproliferative and apoptotic effects of chamomile extract in various human cancer cells. *Journal of agricultural and food chemistry*, *55*(23), 9470-9478.
- Sun, S. C. (2012). The noncanonical NF-κB pathway. *Immunological reviews*, *246*(1), 125-140.
- Sun, S. C. (2017). The non-canonical NF-κB pathway in immunity and inflammation. *Na Rev Immunol*, *17*(9), 545-558.
- Tragulpakseerojn, J., Yamaguchi, N., Pamonsinlapatham, P., Wetwitayaklung, P., Yoneyama, T., Ishikawa, N., ... & Apirakaramwong, A. (2017). Anti-proliferative effect of Moringa oleifera Lam (Moringaceae) leaf extract on human colon cancer HCT116 cell line. *Tropical Journal of Pharmaceutical Research*, *16*(2), 371-378.
- Upadhyay, R. K., Singh, V. R., & Tewari, S. K. (2016). New agro-technology to increase productivity of chamomile (Matricaria chamomilla L.). *Industrial Crops and Products*, *89*, 10-13.
- Vaiopoulos, A. G., Athanasoula, K. C., & Papavassiliou, A. (2013). NF-κB in colorectal cancer. *J Mol Med*, *91*(9), 1029-37.
- Verma, A. R., Vijayakumar, M., Mathela, C. S., & Rao, C. V. (2009). In vitro and in vivo antioxidant properties of different fractions of Moringa oleifera leaves. *Food and Chemical Toxicology*, *47*(9), 2196-2201.
- Villalba, M., Evans, S. R., Vidal-Vanaclocha, F., & Calvo, A. (2017). Role of TGF-β in metastatic colon cancer: it is finally time for targeted therapy. *Cell and tissue research*, *370*(1), 29-39.
- Vivanco, I., & Sawyers, C. L. (2002). The phosphatidylinositol 3-kinase–AKT pathway in human cancer. *Nature Reviews Cancer*, *2*(7), 489-501.

- Wang, S. W., & Sun, Y. M. (2014). The IL-6/JAK/STAT3 pathway: potential therapeutic strategies in treating colorectal cancer. *International journal of oncology*, *44*(4), 1032-1040.
- Wang, S., & Zhang, W. (2016). Genetic variants in IL-6/JAK/STAT3 pathway and the risk of CRC. *Tumor Biology*, *37*(5), 6561-6569.
- Xi, Y., & Xu, P. (2021). Global colorectal cancer burden in 2020 and projections to 2040. *Transl Oncol*, *14*(10), 101174.
- Xu, F., Liu, C., Zhou, D., & Zhang, L. (2016). TGF-β/SMAD pathway and its regulation in hepatic fibrosis. *Journal of Histochemistry & Cytochemistry*, *64*(3), 157-167.
- Yang, Y. (2012). Wnt signaling in development and disease. *Cell & bioscience*, *2*(1), 14-14.
- Yu, H., Lin, L., Zhang, Z., Zhang, H., & Hu, H. (2020). Targeting NF-κB pathway for the therapy of diseases: mechanism and clinical study. *Signal transduction and targeted therapy*, *5*(1), 1-23.
- Yuan, H., Ma, Q., Ye, L., & Piao, G. (2016). The traditional medicine and modern medicine from natural products. *Molecules*, *21*(5), 559.
- Zhang, Y. E. (2017). Non-Smad signaling pathways of the TGF-β family. *Cold Spring Harb Perspect Biol*, *9*(2), a022129.