

## The effect of Fiestin on the colorectal cancer: a review

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### ABSTRACT

Colorectal cancer (CRC) is a virulent tumor rising in the interior wall of the large bowel. CRC is the third deadliest cancer globally and is the 4th common in Iran. Fiestin is a flavone that is present in some fruits and vegetables and is suggested to have beneficial effects on human cancer cells. In the present study, we summarized the potential mechanisms of the effect of Fiestin on CRC. Electronic literature searches were conducted on Medline, Web of Science, and Google Scholar until March 2020. Our search was supplemented with the search of publisher databases like Elsevier and Springer. The search was conducted with “Fiestin” in combination with the following keywords: Colorectal Neoplasms, Colon, Rectum, Apoptosis, Inflammation, and “Precancerous Lesions” among humans, animal, and in-vitro studies. 14 articles during 2005 and 2018 assessed the effect of Fiestin on CRC. One was RCT, 3 of them were animal studies and 10 papers were performed on cell culture. Our Findings suggested that Fiestin may have positive effects on cancer cells due to its anti-inflammatory, apoptotic, anti-oxidative, and cell cycle modifying properties. According to the literature, it seems that Fiestin induces cell cycle arrest and suppresses cellular growth by modulating through some signaling pathways like inhibition of CDKs and Fiestin decreases protein levels of cell division cycles like CDC 2 and CDC25C. Fiestin may also induce cell apoptosis cascades such as activation of caspase 3, 7, and cleavage of procaspase 3 and inhibition of caspase 8. Fiestin also may have anti-inflammatory effects by inhibiting PGE2 production and expression of COX2. Additionally, it may have some anti-oxidant effects by reducing some tumor markers and enhancement levels of some anti-oxidants agents.

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### 1. Introduction

Colorectal cancer (CRC), is a virulent tumor rising in the interior wall of the large bowel (1). CRC represents the third deadliest cancer globally (after lung and breast cancer). The lowest prevalence of this cancer was seen in Africa and Asia and the prevalence is the highest in Australia, New Zealand, North America, and Western Europe with a death rate of roughly 30% (2). CRC is the fourth most common cancer in Iran, the incidence of this cancer has risen in the last two decades, especially in the northern provinces (3). In accordance with the World Health Organization, nearly a million people are diagnosed with CRC annually (4). The global burden of this cancer is expected to rise by 60% by 2030(5). The most common risk factors for colorectal cancer include: lifestyle, unhealthy dietary habits, low physical activity level, chronic inflammation, bowel diseases, aberrant

DNA methylation profiles, mutations in Wnt pathways, and genetic roots, these factors can be classified into two main categories: 85% sporadic factors - 15% genetic roots (6-9). Surgery (tumor resection) and chemotherapy is the first line of treatment in CRC, Radiotherapy is also used for higher stages. Despite the efficacy of these treatments in saving the lives of colorectal cancer patients, they have serious side effects, for example with using chemotherapy drugs such as 5-fluorouracil, Irinotecan, Oxaliplatin, and Capecitabine, numerous serious side effects as: nausea, vomiting, gastrointestinal discomfort has seen. Additionally, since colorectal cancer represents a broad range of radiosensitivity, radiotherapy needs to new attitudes to enhance the efficacy of its function. In addition, according to increasing health care costs, prevention is necessary (5-7, 10). Fiestin (3,3',4',7 tetrahydroxy flavone), can be found in vegetables and fruits like onion, persimmon, strawberry, and cucumber at

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concentration of 2-160 mg/g (11). Fiestin has shown diverse beneficial effects in human cancer cells including anti-oxidant effects by scavenging free radicals and increasing the intracellular levels of glutathione which is the most potent intracellular antioxidant, anti-proliferative effects by perturbing spindle checkpoint signaling, anti-angiogenic effects by preventing both the migration of endothelial cells and the formation of capillary-like structures, anti-invasive mechanism and inducing cell-cycle by inhibition of cyclin-dependent kinases (12-16). In the present review, we aimed to summarize the potential mechanisms of the effect of Fiestin on CRC.

## 2. Materials and methods

A systematic search approach was used to investigate electronic databases such as Web of Science, PubMed, Google Scholar, and Scientific Information Database (SID) to get articles that examine the effect of Fiestin on CRC from inception until March 2020. Our search was supplemented with the search of publisher databases Elsevier and Springer. The search was done with the following MeSH and non-MeSH terms like “Fiestin”, in combination with Colorectal Neoplasms, Colon, Rectum, Apoptosis, Inflammation, and “Precancerous Lesions” among human, animal, and in-vitro studies. All articles addressing the effect of Fiestin on CRC after screening the titles and abstracts were eligible to enter the study. There were no restrictions regarding the language of

publications. Also, studies that were not provided our primary outcomes were excluded. Duplicated articles were removed. Two independent authors (Kh.N, A.B) identifying articles in two phases. In the first phase, titles and abstracts were screened and in the second phase the full text of relevant studies selected in the first phase were carefully examined, and finally related articles were selected. The extracted data include general characteristics of the study (study’s first author, year of publication, study size, design, duration (in cell culture studies, the maximum duration mentioned) and supplement dosage), cell death and apoptotic indices, anti-inflammatory, anti-oxidative factors. EndNote software was used to import and classify all the articles.

## 3. Results and discussion

After excluding the studies, 14 suitable were identified for review (Fig. 1). These 14 articles were published during 2005 and 2018. One was RCT, three of them were animal studies and 10 papers were performed on cell culture. The size of the target group in the human study was 37 patients with CRC and the duration of this study was 7 consecutive weeks. In Naemi et al. study (5), the dose of Fiestin which was used was 100 mg/day. The following factors were assessed in this study: IL-8, IL-10, HS-CRP, MMP-7, MM-9, and a significant difference was seen in the plasma level of IL-8( $p<0.03$ ) compared with the placebo group.

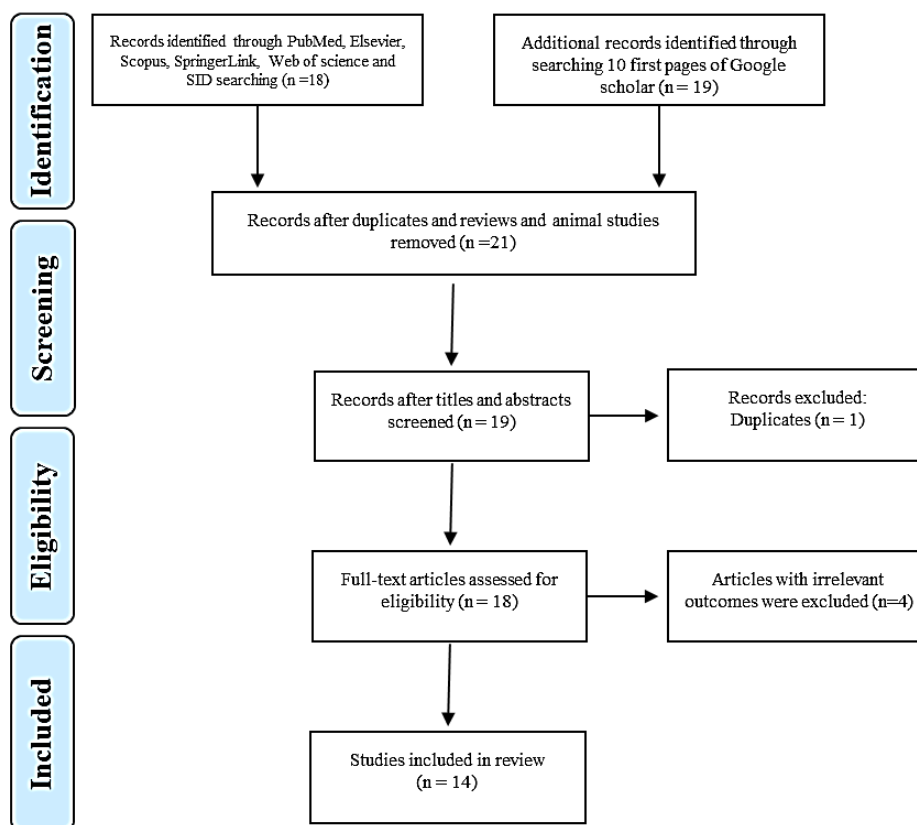


Fig. 1. Flow diagram of the included studies.

In three animal studies, the total number of the target groups was 98, the highest number of samples was 62 and the lowest was 12. The doses which were used in these studies were between 5–800 mg/kg/BW. In these animal studies the factors which were studied included: colonic length, colon weight/length ratio, spleen weight, tumor markers (5'ND, y-GT, CD), MDA, growth of tumor, and expression of its relevant factors like p53 and securin, tumor volume, mouse weight and survival rate, enzymatic and non-enzymatic antioxidants as superoxide dismutase, catalase, glutathione peroxidase, glutathione reductase, vitamin C and E, expression of apoptosis factors like Bcl-2, STAT-3, Bax and caspase-3. In the investigated factors: tumor growth decreased (6, 7), tumor volumes were decreased in one study (6) in dose-dependent manner and another study (7) it decreased significantly without dose-dependent manner ( $p < 0.05$ ), animal's weight and survival rate were notably increased ( $p < 0.05$ ) (7). In addition, treatment with Fiestin decreased the expression of Bcl2, SRAT3 (17), and securing (7) and an increment in the expression of Bax, caspase 3(17), and p53 (7) was observed. Also, Fiestin treatment protected and retained colon length and decreased spleen enlargement ( $p < 0.05$ ) (17). Moreover, the levels of tumor markers (5'ND, y-GT, CD) and MDA were reduced by Fiestin. Additionally, levels of antioxidants like GPx, GR, and GSH increased (17). Also, in the study of Leu et al. (7), HCT116 tumor growth was remarkably inhibited by combined Fiestin/radiation treatment without considerable loss of body weight. Ten other studies were considered as studies of cell culture. These studies were conducted on a variety of colorectal cancer cells such as HT-29, SNU-C4, HCT116, HCT-15, COLO-205, CaCo-2, HaCaT and etc. In these studies, the factors which were studied included: cell growth and survival rate, levels of caspase 3,7,9 and 8, levels of bcl2, MMP, PEG2, EGFR, p53, DNA synthesis and ladder formation, levels of CDKs and CDCs. In all of this studies Fiestin increased cancer cell death. In two studies (4, 11, 18) Fiestin inhibited both cell growth and DNA synthesis by Perturbing cell cycle progression and decreasing the activities of CDK, protein levels of CDC, releasing of VEGF, GM and CSF, also Fiestin induced cell death by increasing the inhibition of CYP3A4 (19). In 7 studies (4, 12, 16, 19-22) treatment with Fiestin increased apoptosis in cell lines by enhancement of its relevant factors like caspase 3,7,9, loss of MMP, decreasing bcl2 and ERK, cleavage of PARP and caspase 8, phosphorylation of p38 MAPK and decreasing p53, inhibition of EGFR and NF-kB activity, increasing DNA ladder formation, however in Chen et al. study (22) Fiestin induced more DNA fragmentation (apoptosis hallmark) in securin-null HCT116 cells but not in HT-29 and HaCaT cells and in Lee et al. study (23), treatment with Fiestin for 6 days upregulated PBR mRNA expression in SNU-C4 cells but did not affect for 3 days. On the other hand, in two studies (20), Fiestin was compared with Fiestin plus N-Acetyl-L-Cysteine (NAC) and HSP90 Inhibitors Geldanamycin, Radicolol that Fiestin-induced toxicity by encouraging apoptosis is enhanced with NAC, HSP90 Inhibitors Geldanamycin and Radicolol. In addition, in Chen et al. study (22), pretreatment with Fiestin

increased radiation-induced caspase-dependent apoptosis and cell growth arrest in HT-29 cells. In two studies (4, 19), Fiestin decreased the activity of PGF2 and expression of COX2 due to its anti-inflammatory effect. In Youns et al. study (19), treatment with Fiestin showed anti-oxidant activity by enhancement of some anti-oxidants levels (Table 1). Fiestin may have positive effects on cancer cells via several mechanisms such as: inducing cell-death by effects on cellular metabolism, inhibiting both cell growth and DNA synthesis, infusing apoptosis, and inhibiting the survival pathway, anti-inflammatory effect, and anti-oxidant effect.

### 3.1. Effects on cellular growth

We concluded that Fiestin inhibited cell growth and survival. In line with our study, Fiestin in a dose-dependent manner induced cell-death by affecting cellular metabolism like inhibition of CYP3A4 in Youns et al. study (19). Additionally, Lu et al. (11) observed that Fiestin dose-dependently may inhibit both cell growth and DNA synthesis by disturbing cell cycle progression from the G1 to S phase, in addition, G2 /M phase arrest was observed too, also they indicated that Fiestin can decrease the activities of CDK2 and CDK4; these effects were likely attributable to decreases in the levels of cyclin E and D1 and an increase in p21CIP1/WAF1 levels. Fiestin solely can target CDK4 activity in a cell-free system, indicating that CDK4 may be the direct target of it. The accumulation of E2F transcription factor activity (which are necessary for regulation of the cell cycle) done by phosphorylation of the Rb family and phosphorylation state of the retinoblastoma proteins shifted from hyperphosphorylated to hypophosphorylated in cells treated with Fiestin, the protein levels of CDC 2 and CDC25C and the activity of CDC2 were also decreased in Fiestin-treated cells (11). Moreover, Lee et al. (23) found that Fiestin inhibited the survival of the SNU-C4 adenocarcinoma cells via reducing the release of VEGF, GM-CSF from those cells in dose-dependent manner.

### 3.2. Effects on apoptosis

Our result revealed that Fiestin induced apoptosis. Apoptosis is a physiological cell death that eliminates old and damaged cells and is essential for tissue homeostasis and disruption of this process can cause growth and proliferation of cancer cells. Apoptosis occurs through two pathways: intrinsic and extrinsic. In the intrinsic pathway, cytochrome c is released from the space between the two mitochondrial membranes into the cytoplasm. Cytochrome c interacts with Apaf-1, ATP, and procaspase 9 to produce apoptosome. The apoptosome activates caspase 9 and then it activates executioners' caspases like caspase3, 6, and 7. The permeability of the mitochondrial membrane to cytochrome c is determined by the ratio of pro-apoptotic and anti-apoptotic mediators. Pro-apoptotic molecules such as Bax or Bak increase the permeability of the mitochondrial membrane. Coupling of pro-apoptotic molecules with anti-apoptotic (like Bcl-2 and Bcl-XL) factors can counteract their anti-apoptotic

**Table 1.** Characteristics of included publication.

Author	Study groups and groups size	Type of intervention	Duration	Dose	Results
<b>Naemi et al. 2018</b> (5)	n = 37 CRC patients undergoing chemotherapy	Double-blind, randomized placebo-controlled clinical trial	7 consecutive weeks	100 mg Fiestin	<ul style="list-style-type: none"> <li>IL-8: a significant difference was seen in the plasma level of IL-8 compared to placebo group (p&lt;0.03).</li> <li>IL-10, HS-CRP, MMP-7, MM-9: no significant changes were seen.</li> </ul>
<b>Jeng et al.2018</b> (6)	Nude mouse xenograft Male nude mice (8 weeks old). The animals were divided into three groups (n = 4 per group)	Animal study (+ cell culture)	4 weeks	400mg/kg/day and 800 mg/kg/day	<ul style="list-style-type: none"> <li>Fiestin prevents tumor growth in nude mice, treatment with Fiestin decreased tumor volumes in a dose-dependent manner.</li> </ul>
<b>Kuncari et al. 2016</b> (17)	Human colon cancer cell line LoVo, Oxaliplatin-resistance LoVo cell, and CPT11-resistance LoVo		24 hours	40, 60, 80, and 100 $\mu$ M	<ul style="list-style-type: none"> <li>CPT11 resistance LoVo cancer cells were more sensitive to Fiestin treatment than parental LoVo and OR- LoVo cells. In CPT11-LoVo cells, 40 <math>\mu</math>M of Fiestin dosage significantly decreased cell survival, compared to the controls (p&lt;0.05).</li> <li>Fiestin treatment increased apoptotic cells in all three cancer cells with respect to dosage. Additionally, it considerably induced TUNEL positive cells in CPT11-LoVo cancer cell than in parental or OR- LoVo cancer cells.</li> </ul>
	Male balb/c mice weighing approximately 15-20 grams The animals were divided into four groups (n = 6 per group) group 1: as control group 2: mice were injected AOM/DSS group 3: mice were induced with AOM/DSS + 20mg/kg body weight Fiestin group 4: animals received Fiestin 20mg/kg body	Animal study	62 days	20mg/kg body weight Fiestin	<ul style="list-style-type: none"> <li>Spleen weight is increased in the AOM/DSS induced group, treatment with Fiestin reduced the spleen weight when compared to the control group.</li> <li>In AOM/DSS induced group in comparison to the control group, the colon weight/length ratio was high and treatment with Fiestin significantly lowered the ration (p&lt;0.05).</li> <li>Tumor markers (5'ND, <math>\gamma</math>-GT, CD) and pathophysiological enzymes (ALP and LDH) level were reduced significantly by Fiestin in comparison to AOM/DSS induced mice.</li> <li>The level of MDA in AOM/DSS induced mice reduced by Fiestin.</li> <li>The expression of Bcl-2 and STAT-3 increased in the AOM/DSS induced group whereas treatment with Fiestin significantly decreased the anti-apoptotic Bcl-2 and STAT-3. The expression of Bax, Caspase-3 significantly reduced in the AOM/DSS induced group whereas treatment with Fiestin increased the pro-apoptotic Bax and caspase-3 protein expression.</li> <li>The reduction of enzymatic and non-enzymatic antioxidants like superoxide dismutase, catalase, glutathione peroxidase, glutathione reductase, vitamin C, and vitamin E because of AOM/DSS induction were brought near to normal by Fiestin.</li> <li>Treatment with Fiestin increased the levels of GPx, GR and GSH in comparison with the AOM/DSS induced group (p&lt;0.05).</li> </ul>

**Table 1.** Characteristics of included publication.

Author	Study groups and groups size	Type of intervention	Duration	Dose	Results
<p><b>Leu et al. 2016</b> (7)</p>	<p>A total of 62 male balb/c nude mice (weight, 20 g; age, 6 weeks). mouse tumor xenograft model generated by injection of Murine CT-26 colon cancer cells, and human HCT116<sup>WT</sup>, HCT116<sup>p53-/-</sup>, HCT116<sup>securin-/-</sup> and p53-R273H mutant HT-29 colorectal cancer cell lines</p>	Animal study	30 days	5 mg/kg twice in combination with radiation	<ul style="list-style-type: none"> <li>• Fiestin inhibits <i>in vivo</i> tumor growth in a mouse CT-26 xenograft model: Tumor volume: significantly decreased (p&lt;0.05). Survival rate: increased significantly (p&lt;0.05).</li> <li>• CT-26 tumor growth was blocked by Fiestin and radiation alone, this effect improved by combination Fiestin+radiation treatment at 16 to 29 days following treatment (p&lt;0.05).</li> <li>• HCT116 tumor growth was completely and remarkably inhibited by combined Fiestin/radiation treatment without considerable loss of body weight (p&lt;0.05).</li> <li>• Fiestin raises the p53 expression and declines the securin expression, which suppresses the growth of the tumor. Fiestin-mediated p53 and securin expression were independent of the expressive status of p53/securin (null or wild-type).</li> <li>• Fiestin downregulated the expression of the oncoprotein securin in a p53-independent manner. However, tumors of securin-null HCT116 displayed only moderate sensitivity to Fiestin treatment, and the combination of Fiestin and radiation did not noteworthy suppress the growth of the securin-null HCT116 tumor in comparison to normal HCT116 tumors.</li> <li>• The depletion of securin does not enhance the effects of Fiestin and radiation combination treatment on colorectal tumors <i>in vivo</i>.</li> </ul>
<p><b>Youns et al. 2017</b> (19)</p>	<p>The human colorectal carcinoma (CaCo-2) cells</p>	Cell culture	48 hours	0.01, 0.1, 1, 10, 100, 000, 10000 µM	<ul style="list-style-type: none"> <li>• Cellular proliferation and viability of colorectal cancer cell lines were inhibited with Fiestin treatment.</li> <li>• Activation of caspase 3/7 increase significantly by Fiestin (p&lt;0.001).</li> <li>• Significantly inhibition of PGE2 production was seen with the treatment of Fiestin (p&lt;0.05).</li> <li>• Fiestin caused a significant inhibition of CYP3A4 in a dose-dependent manner.</li> <li>• Fiestin inspires Glutathione-S-transferase enzyme inhibition in a dose-dependent manner.</li> </ul>
<p><b>Wu et al. 2014</b> (12)</p>	<p>COLO205, HCT-116, HCT-15, and HT-29 colonic carcinoma cells</p>	Cell culture	24 hours	60-120 m Fiestin	<ul style="list-style-type: none"> <li>• Fiestin-induced cytotoxicity via inducing apoptosis is enhanced with N-Acetyl-L-Cysteine treatment (p&lt;0.05).</li> <li>• Increase caspase 9 protein cleavage and loss of the MMP with decrease Bcl-2 protein enhanced by NAC in Fiestin treated COLO205 cells (p&lt;0.05).</li> <li>• Fiestin-induced apoptosis of COLO205 cells by suppression of ERK activation enhanced with NAC.NAC potentiates the cytotoxic effects of Fiestin (via characteristics of apoptotic like DNA ladders, and caspase 3 and PARP protein cleavage) against the viability of HCT-116, HT-29, and HCT-15 Colorectal carcinoma cells.</li> </ul>

**Table 1.** Characteristics of included publication.

Author	Study groups and groups size	Type of intervention	Duration	Dose	Results
<b>Wu et al. 2013</b> (20)	human COLO205 colon cancer cells	Cell culture	24 hours	30-60-120 m	<ul style="list-style-type: none"> <li>• Fiestin-induced cytotoxicity via inducing apoptosis by reducing the integrity of DNA via increasing DNA ladder formation in COLO205 cells enhanced with HSP90 Inhibitors Geldanamycin and Radicicol (<math>p &lt; 0.05</math>).</li> <li>• Fiestin-induced apoptosis (by induction of caspase-3-mediated) in COLO205 cell enhanced with GA or RAD (<math>p &lt; 0.05</math>).</li> <li>• Fiestin-induced apoptosis (by loss of the MMP with activation of caspase-9) in COLO205 cell enhanced with GA or RAD (<math>p &lt; 0.01</math>).</li> <li>• A remarkable decrease in the p53 protein was observed with Fiestin+GA or Fiestin+RAD treated in COLO205 cells in comparison to that in control (FIS-, GA-, and RAD-treated) cells.</li> </ul>
<b>YU et al. 2011</b> (21)	securin-wild-type, securin-null and p53-null human HCT116 colon cancer cells, The p53-mutant human HT-29 colorectal cancer cells	Cell culture	24 hours	The cells were treated with 25, 50, and 100 $\mu$ M Fiestin	<ul style="list-style-type: none"> <li>• Fiestin displays higher cytotoxicity in securin-null HCT116 human colon cancer cells and the reconstitution of securin expression rescues HCT116 cells from Fiestin-induced cell death.</li> <li>• Fiestin-induced cell death was enhanced with knockdown of securin expression in wild-type HCT116 cells.</li> <li>• P53-deficient human colon cells are resistant to Fiestin-induced cytotoxicity and apoptosis and the sensitivity of p53-null HCT116 cells to Fiestin enhances with securin depletion.</li> </ul>
<b>Chen et al. 2010</b> (22)	HCT116 (p53-wild type) cells, HT-29 (p53-mutant) cells, HaCaT (p53-mutant) cells	Cell culture	48 hours	0-100	<ul style="list-style-type: none"> <li>• Fiestin treatment remarkably increased the DNA fragmentation in a dose dependent manner in HCT116 cells but not in HT-29 and HaCaT cells.</li> <li>• The survival fraction was decreased dose-dependently by Fiestin pretreatment in three types of cells.</li> <li>• Fiestin pretreatment repressed radiation-induced phosphorylation of H2AX and phospho-Chk2 (Thr-68) in HT-29 cells.</li> <li>• Fiestin pretreatment prolonged radiation-induced G2/M arrest and increased radiation-induced cell growth arrest in HT-29 cells.</li> <li>• Pretreatment with Fiestin increased radiation-induced caspase-dependent apoptosis in HT-29 cells.</li> <li>• Phosphorylation of p38 MAPK contributed to radiation-induced apoptosis and radiosensitivity increment in HT29 cells pretreated with Fiestin.</li> <li>• Pretreatment with Fiestin increased radiosensitivity of irradiated HT-29 cells by prevention of AKT-ERK pathways.</li> </ul>

**Table 1.** Characteristics of included publication.

Author	Study groups and groups size	Type of intervention	Duration	Dose	Results
<b>Kim et al. 2009</b> (18)	human adenocarcinoma SNU-C4 (colorectal) cancer cell	Cell culture	6 days	10 <sup>-5</sup> M, 10 <sup>-6</sup> M, 10 <sup>-7</sup> M, 10 <sup>-8</sup> M, 10 <sup>-9</sup> M, 10 <sup>-10</sup> M	<ul style="list-style-type: none"> <li>• Fiestin indicated concentration-dependent inhibition on the survival of the SNU-C4 adenocarcinoma cells also longer duration of treatment, showed the higher repressor effect.</li> <li>• Fiestin indicated decreasing the release of VEGF, GM-CSF from SNU-C4 cells.</li> <li>• The concentration-dependent inhibitory effect on FAS activity (known as an anticancer mechanism of flavonoids) was observed in the various concentrations of the sedative-treated SNU-C4 cancer cells.</li> <li>• For the SNU-C4 adenocarcinoma cells, anticancer cytotoxicity induced by flavonoids or diazepam was not reduced by the treatment of 10<sup>6</sup> m concentration of 5-fluorouracil (5-FU), a chemotherapeutic agent. Their inhibitory action on cancer cell survival was further enhanced by the addition of PK11195(a putative chemosensitizer) (p&lt;0.05).</li> </ul>
<b>Lim et al. 2009</b> (16)	HCT-116 human colon cancer cells	Cell culture	2 days	5–20 mol/L of Fiestin	<ul style="list-style-type: none"> <li>• Fiestin induces apoptosis via increasing the cleavage of PARP, caspases 9, 3, and 7 in HCT-116 cells (p&lt;0.05).</li> <li>• Fiestin impels depolarization of the mitochondrial membrane in HCT-116 cells (p&lt;0.05).</li> <li>• Fiestin also increased the levels of Smac/Diablo and cytochrome <i>c</i> in the cytoplasm and decreased those levels in the mitochondria.</li> <li>• Fiestin mutates the levels of Bcl-2 family proteins and induces Bax translocation to mitochondria (p&lt;0.05).</li> <li>• Fiestin enhances the cleavage of caspase-8 (p&lt;0.05).</li> <li>• A caspase-8 inhibitor abates Fiestin-induced apoptosis (p&lt;0.05).</li> <li>• Inhibition of p53 expression results in reducing Fiestin-induced apoptosis and the translocation of Bax to the mitochondria. (p&lt;0.05).</li> </ul>
<b>Lee et al. 2009</b> (23)	SNU-C4 colorectal adenocarcinoma cells	Cell culture	6 days	10 <sup>-6</sup> M	<ul style="list-style-type: none"> <li>• Fiestin treatment for 3 days inhibited the survival of SNU- C4 human colorectal cancer cells.</li> <li>• Fiestin inhibited glucose consumption of SNU-C4 cells and significantly (p&lt;0.05) reduced insulin-induced enhancement of glucose consumption by SNU-C4 cells.</li> <li>• Treatment with Fiestin for 6 days upregulated PBR mRNA expression in SNU-C4 cells but did not affect for 3 days.</li> </ul>
<b>Suh et al. 2008</b> (4)	HCT116 and HT29 human colon cancer cells	Cell culture	96 hours	(30–240) M	<ul style="list-style-type: none"> <li>• Fiestin induced growth inhibition of HCT116 and HT29 colon cancer cells</li> <li>• Fiestin inhibits expression of COX2 in HT29 cells</li> <li>• Fiestin prevents COX2 promoter activity and PGE2 secretion.</li> <li>• Fiestin inhibits expression and translocation of TCF1 and TCF4 in HT29 cells</li> <li>• COX 2 expression inhibited through downregulation of TCF4 by Fiestin in HT29 cells</li> </ul>

**Table 1.** Characteristics of included publication.

Author	Study groups and groups size	Type of intervention	Duration	Dose	Results
Lu et al. 2005 (11)	HT29 human colon cancer cells	Cell culture	72 hours	20, 40, or 60 mol/L Fiestin	<ul style="list-style-type: none"> <li>• Fiestin inhibits activation of EGFR and NF-kB in HT29 cells</li> <li>• Fiestin reduces expression of Wnt target genes (cyclin D1, and MMP-7) and inhibits colony formation.</li> <li>• Fiestin dose-dependently inhibited both cell growth and DNA synthesis (<math>p &lt; 0.05</math>), which were observed with decrease in cell number. Perturbed cell cycle progression from the G1 to S phase was observed with Fiestin treatment, and G2/M phase arrest was observed too (<math>p &lt; 0.05</math>).</li> <li>• The phosphorylation state of the retinoblastoma proteins shifted from hyperphosphorylated to hypophosphorylated in cells treated Fiestin (<math>p &lt; 0.05</math>).</li> <li>• Fiestin decreased the activities of cyclin-dependent kinases CDK2 and CDK4 leading to decreased Rb phosphorylation.</li> <li>• Fiestin inhibited CDk4 activity in a cell-free system (<math>p &lt; 0.05</math>), indicating that it may directly prevent CDk4 activity.</li> <li>• The protein levels of cell division cycles CDC2 and CDC25C and the activity of CDC2 were also decreased in Fiestin-treated cells (<math>p &lt; 0.05</math>).</li> </ul>



effect. Therefore, the ratio of pro-apoptotic and anti-apoptotic mediators determines the available amount of cytochrome c for apoptosome formation. The external pathway is also via death receptors that are present in most cell membranes. When these receptors are stimulated by the respective ligands, they activate caspases and induce apoptosis (24). Consistent with our results Lim et al. (16) showed that Fiestin increased caspases 9, 3, and 7, cleaving of PARP and caspase-8, depolarization of the mitochondrial membrane, levels of Smac/Diablo and cytochrome c in the cytoplasm, rising the levels of Bcl-2 family proteins and Bax translocation to mitochondria. Also, Wu et al. (12) found that Fiestin improved the apoptotic cascades like caspase 9 activation, loss of MMP, lowering Bcl-2. Moreover, Jeng et al. (6) indicated that Fiestin induces apoptosis and inhibits survival pathway in parental and chemoresistance colon cancer cells via up-regulating caspase 8 and caspase 3 cleavage and cytochrome c expression probably via preventing inappropriate activation of IGF1R and AKT proteins in parental and in OR and CPT11-LoVo cells. As well as, Wu et al. (12) reported that Fiestin induced apoptosis by suppression of ERK activation in COLO205 cells which enhanced with NAC. In various tumors including the colon, securin is highly expressed. Knockdown of securin in HCT116 p53-null cells potentiated Fiestin-induced cytotoxicity by stimulation of apoptosis. Human colon cancer cells are sensitizing to Fiestin-induced apoptosis by securin reduction (7). Additionally, Fiestin induced more DNA fragmentation (apoptosis hallmark) in securin-null HCT116 cells that confirmed by the increases of both phosphatidylserine (PS) externalization and PI uptake but not in HT-29 and HaCaT cells (21). However, securin-null HCT116 tumors displayed only moderate sensitivity to Fiestin treatment, and the combination of Fiestin and radiation did not notably suppress securin-null HCT116 tumor growth in comparison to normal HCT116 tumors (7). Chen et al. (22) showed that Fiestin pretreatment increased radiosensitivity of irradiated HT-29 cells via inhibition of AKT-ERK pathways, causing accumulation of cells in the radiosensitive G2/M phase and suppressing cellular DNA repair capacity, thus increasing radiation-induced double-strand breaks. Additionally, phosphorylation of p38 MAPK contributed to radiosensitivity increase in HT29 cells pretreated with Fiestin also its pretreatments suppressed radiation-induced phosphorylation of H2AX and phospho-Chk2 (Thr-68) in HT-29 cells.

### 3.3. Effects on inflammation

In this review, we concluded that Fiestin has anti-inflammatory effects by decreasing serum levels of IL-8, hs-CRP, MMP-7, and inhibiting PGE2 production. The increased levels of IL-8 and higher expression of IL-8 reported in CRC and is related to tumor size and stage, depth of infiltration, and liver metastasis. Therefore, inhibiting the expression or reducing IL-8 production as one of the approaches for clinical management of CRC (5). Also, Fiestin could reduce the releasing of the cytokines directly or indirectly by downregulation in MAPKs and the suppression of NF-kB

activity (5). In the line of our result, several in vitro studies with colon cancer cells like Suh et al. study (4) have shown that treatment with Fiestin could reduce levels of MMPs levels by modulation of MAPK and NF-kB pathways but Naemi et al. (5) study showed that supplementation with Fiestin could not affect the levels of MMPs in comparison to the placebo however they showed that the levels of IL-8 significantly reduced. Moreover, the treatment of Fiestin prevents expression and activity of COX-2 by inhibiting NF-kB signaling via downregulation of TCF-4 in HT-29 cells, which are known to play a critical role in colon carcinogenesis (4).

### 3.4. Effects on oxidative stress

Our results showed that Fiestin can act as an anti-oxidant agent. By higher level of reactive oxygen species (ROS) and reactive nitrogen species in the status of oxidative stress, the cell and tissue damage, inflammation that leads to carcinogenesis, increased. Intestinal cells have enzymatic and non-enzymatic antioxidants like SOD, GPX, GR, GSH and vitamin C, E to scavenge the ROS but extreme ROS production reduces the antioxidant protection system as well as stimulates excessive production of free radicals via peroxidation of lipids. The end by-product during lipid peroxidation is MDA which is a mutagenic agent and contributes to the development of CRC (17). In the line with our result, Kuncari et al. (17) study indicated that Fiestin reduced the tumor markers (5'ND,  $\gamma$ -GT, CD), enhanced antioxidant levels, and decreased the levels of MDA.

### 4. Conclusion

Reviewing the eligible articles showed that, the use of Fiestin may have positive effects on the CRC. Fiestin induces cell cycle arrest and suppresses cellular growth by modulating through some signaling pathways and also it decreases some protein levels of cell division cycles. Additionally, Fiestin may induce cell apoptosis cascades such as activation of caspase 3, 7, and cleavage of procaspase 3 and inhibition of caspase 8. Furthermore, Fiestin may have anti-inflammatory effects by inhibiting PGE2 production and expression of COX2. In addition, it may have some anti-oxidant effects by increasing levels of some antioxidants agents and decreasing the levels of MDA and also in mice by reducing tumor markers (5'ND,  $\gamma$ -GT, CD).

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