黄酮类衍生物抗肿瘤作用研究进展

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摘要: 恶性肿瘤是严重危害人类生命健康的疾病, 一直是科研工作者的研究重点之一。天然黄酮及其衍生物具有多种生理活性, 尤其在抗肿瘤生长方面具有独特生物活性, 既可以通过参与干扰肿瘤细胞周期, 改变肿瘤细胞线粒体膜电位, 促进肿瘤细胞的凋亡, 也可以通过提高人体免疫力, 减小肿瘤细胞的免疫逃逸, 阻止肿瘤转移。在人体内, 它们调整生物信号转导, 导致促凋亡蛋白表达上调, 通过调控血管上皮细胞的生长, 阻断肿瘤组织中血管的生成, 达到抑制实体肿瘤生长的目的。此类化合物在对多种肿瘤的研究过程中均表现药理活性, 有望开发为新型抗肿瘤药物。本文总结了近年来黄酮类化合物在抗肿瘤作用机制及药效学方面的进展, 旨在为相关科研工作者提供一定的参考与帮助。

关键词: 黄酮类化合物; 癌症; 抗肿瘤; 天然植物; 作用机制

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Research progress on the antitumor effect of flavonoid derivatives

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Abstract: Malignant tumors seriously endanger human life and health, and their treatment has always been a research focus of scientists all over the world. Natural flavonoids and their derivatives have a variety of biological activities, especially regarding antitumor growth, with unique biological activities. They can interfere with the growth cycle of tumor cells, change the mitochondrial membrane potential, promote apoptosis, and can reduce the immune escape of tumor cells and prevent tumor metastasis by improving human immunity. In the human body, they regulate the biological signal transduction, leading to the up-regulation of pro-apoptotic protein expression. They inhibit the growth of solid tumors by regulating the growth of vascular epithelial cells and blocking the formation of blood vessels in tumor tissue. Recent studies have shown that these compounds can play an important role in the treatment of various human tumors and are expected to be developed into new antitumor drugs. This review summarizes the recent research results on the antitumor mechanism of flavonoids and their ability to inhibit tumor growth.

Key words: flavonoid; cancer; antitumor; plant; mechanism

恶性肿瘤是危害人类生命健康的主要疾病[1,2]。2015年世界卫生组织调查报告显示, 在172个国家中, 癌症是91个国家70岁以下人群的第一或第二大死因, 并预计将成为全球在21世纪的首位死因[3]。天然产物作为天然药库, 是推动我国医药事业发展的重要组成部分[4], 为我国医药学研究奠定了基础。黄酮类化合物在自然界中并不少见, 是一类天然多酚化合物, 基本

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化学结构如图1所示, 这类化合物广泛存在于各类中草药中, 发挥着重要作用[5,6]。因其具有独特的化学结构, 黄酮类化合物显现了广泛的生理和药理作用[7], 包括抗氧化[8-10]、抗菌[11]、抗炎[12,13]、抗病毒[14]、抗肿瘤[15-17]以及防治心血管疾病[18]等多方面应用[19-22]。部分天然黄酮类化合物以及其衍生物展现出了开发成新型抗肿瘤药物的良好前景 (表1)。作者就天然黄酮类化合物的抗肿瘤特性以及抗肿瘤作用机制进行归纳总结, 希望对相关科研工作者有所帮助。

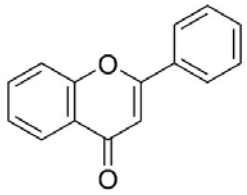

Figure 1 The parent nucleus structure of flavonoids

1 黄酮类化合物抗肿瘤作用机制研究

黄酮类化合物具有多种抗肿瘤作用, 例如: 抑制肿瘤细胞增殖和促进肿瘤细胞凋亡; 调节肿瘤组织中的血管生成; 增强人体免疫功能; 干预信号通路等 (图2)。

1.1 抑制肿瘤增殖和促进细胞凋亡

抑制肿瘤细胞的增殖或者促进肿瘤细胞的凋亡是黄酮类化合物发挥其抗肿瘤作用的主要机制。例如, 5-羟基川陈皮素 (5-hydroxy-6,7,8,3',4'-pentamethoxy-flavones, 5HPMF), 它是从中药陈皮中分离得到的一种结构新颖的黄酮类化合物, 对转运蛋白P-糖蛋白 (P-glycoproteins, P-gp) 表现出显著抑制作用, 用于治疗和预防由于P-gp引发的相关疾病。Cui等[23]以肿瘤细胞的糖胺聚糖 (glycosaminoglycans, GAGs) 为切入点, 以5HPMF为研究对象, 研究了它对肺癌细胞 (A549)、结肠癌细胞 (HT29和HCT116) 中GAGs的影响, 实验发现, 来源于HT29和A549癌细胞中的GAGs可促进成纤维细胞生长因子1 (fibroblast growth factor 1, FGF1) 介导的BaF3细胞 (小鼠原B细胞株) 的生长, 而5HPMF可显著抑制BaF3细胞的生长。并且, 5HPMF显著降低FGF2或FGF8介导的GAGs·FGF·FGFR三元复合物在HCT116和A549细胞表面的蛋白表达, 却对GAGs中葡萄糖胺和半乳糖胺的含量无明显影响。此研究表明, 5HPMF抑制结肠癌和肺癌细胞增殖的作用主要是通过影响多种重要蛋白如p21、cyclin D1、caspase-3、PARP (poly ADP-ribose polymerase) 和Bax等来调控细胞周期, 从而促进凋亡, 达到抑制肺癌和结肠癌细胞增殖的目的。Zhu等[24]通过检测半胱氨酸−天冬氨酸蛋白酶的活性, 发现黄酮类化合物白花蛇舌草水提物发挥其抗肿瘤作用是以凋亡蛋白酶为作用靶点从而引起肿瘤细胞凋亡的, 研究发现, 随着给药浓度的增加, 凋亡蛋白酶caspase-3和caspase-8的活性及蛋白水平随之增强, 所以白花蛇舌草水提物能通过诱导细胞的凋亡达到抑制肿瘤细胞增殖的目的。Duan[25]以主要成分为黄酮类的蒙药材叉分蓼为主要研究对象, 发现其可抑

Table 1 Flavonoids used in clinical trials. CDKs: Cyclin-dependent kinases; TOP2: Topoisomerase II

Drug	Mechanism	Registration mark (testing institution)	Development phase (status)	Treatment
Flavopiridol	CDKs inhibitor	NCT0023894 (Novartis)	Phase II completed	Endometrial cancer
		NCT00016939 (Southwest Oncology Group)	Phase II completed	Kidney cancer
		NCT00003256 (National Cancer Institute, NCI)	Phase II completed	Prostate cancer
		NCT00006245 (Memorial Sloan Kettering Cancer Center)	Phase II completed	Esophageal cancer
P276-00	CDKs inhibitor	NCT01903018 (Piramal Enterprises Limited, PEL)	Phase II completed	Radiation induced mucositis in head and neck cancers
		NCT00898287 (PEL)	Phase I/II completed	Pancreatic cancer
Wogonin	CDK4 and cyclin D1 inhibitor	CTR20171665 (Yiyang Central Hospital)	Phase I completed	Liver cancer and gastric cancer
Genistein	Estrogen receptor	NCT00290758 (NCI)	Phase II completed	Breast cancer
		NCT00099008 (UNC Lineberger Comprehensive Cancer Center)	Phase I completed	Breast cancer and endometrial cancer
		NCT01325311 (NCI)	Phase II completed	Prostate cancer
Silibin-Phytosome	Unidentified	NCT00487721 (University of Colorado, Denver)	Phase II completed	Prostate cancer
Silymarin	Unidentified	NCT01829178 (Tehran University of Medical Sciences)	Phase II/III completed	Upper G1 cancer
Polyphenon E	Unidentified	NCT00363805 (Sherry Chow)	Phase II completed	Lung cancer prevention
		NCT00516243 (NCI)	Phase I completed	Breast cancer
		NCT00459407 (NCI)	Phase I completed	Prostate cancer
		NCT00666562 (NCI)	Phase II completed	Bladder cancer
Soy isoflavone	Unidentified	NCT02075112 (Emory University)	Phase I completed	Head and neck cancers

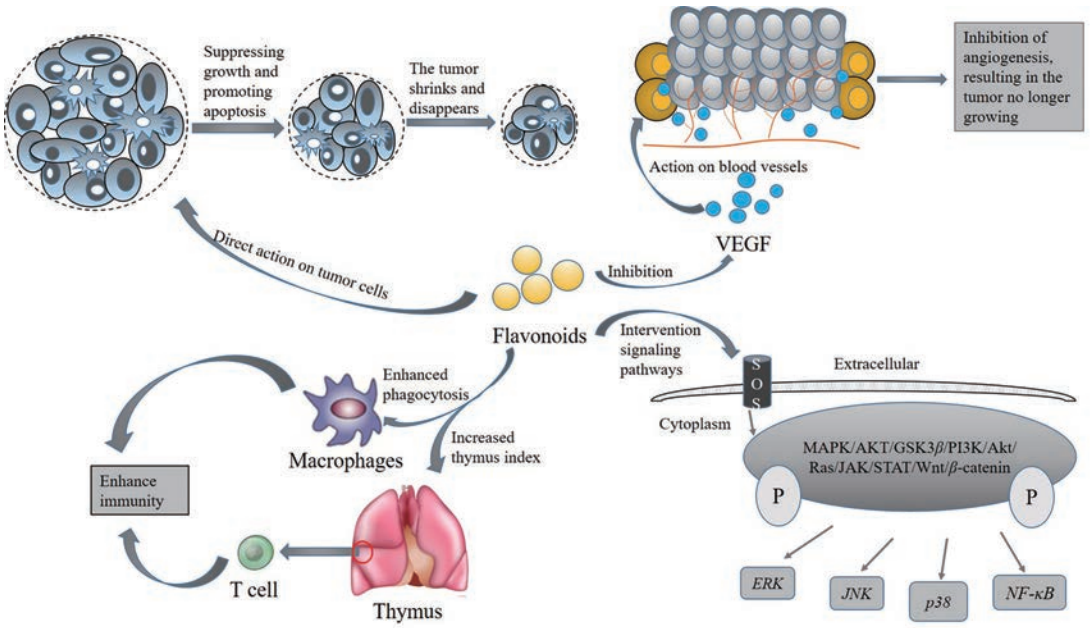

Figure 2 Summary of antitumor mechanism of flavonoids. Flavonoids play an antitumor role through the above four mechanisms of action, including directly acting on tumor cells, inhibiting cell proliferation, and promoting cell apoptosis; acting on vascular endothelial growth factor (VEGF), regulating angiogenesis in tumor tissue, and thus inhibiting tumor growth; acting on immune cells or organs to improve the body's immunity; acting on the signaling pathway, regulating tumor cell growth, and interfering with protein expression in tumor cells. ERK: Extracellular signal-related kinase; JNK: c-Jun N-terminal kinase; NF-κB: Nuclear factor kappa-light-chain-enhancer of activated B cells

制人乳腺癌MDA-MB-231细胞的增殖, 并可诱导B淋巴细胞瘤-2基因 (B-cell lymphoma-2, *Bcl-2*)、*Bcl-xl*、细胞周期蛋白依赖性激酶 (cyclin-dependent kinase, *CDK*)、磷酸烯醇式丙酮酸羧激酶 (phosphoenolpyruvate carboxykinase, *PEPCK*) 和基质金属蛋白酶2 (matrix metalloproteinase 2, *MMP2*) 基因水平的下调。增殖细胞核抗原 (proliferating cell nuclear antigen, PCNA) 为DNA聚合酶的一种辅助蛋白, 参与调节DNA的合成, 它在体内的含量与细胞增殖密切相关, 是评价癌细胞增殖状态的重要指标之一。Hailiwu[26]的研究发现, 高良姜总黄酮在体内能够降低血清中PCNA、MMP-9、血管内皮生长因子 (vascular endothelial growth factor, VEGF)、白细胞介素-6 (interleukin-6, IL-6)、IL-1β和IL-17的水平, 起到抑制胃癌MFC移植瘤生长的作用。Wang[27]对代代花化合物的抗肿瘤作用的研究结果表明, 黄酮类化合物柚皮素对肝癌细胞增殖的抑制作用较强, 在最大安全浓度时的抑制效果优于5-氟尿嘧啶, 而对于乳腺癌细胞, 柚皮素的抑制效果更好。综上, 黄酮类化合物能通过抑制肿瘤细胞增殖或促进细胞凋亡的途径起到抗肿瘤细胞增殖的作用。

1.2 调节血管生成

实体肿瘤血管生成在肿瘤的发生发展中起到至关重要的作用, 当恶性肿瘤大小超过1～2 mm^{3}时, 其继续生长与转移必须依靠新生血管提供足够的营养才能实现。近年来, 抗肿瘤血管生成已成为肿瘤治疗的方案之一, 成为最有希望的肿瘤治疗靶标。研究表明, 许多黄酮类化合物的抗肿瘤作用与其抑制肿瘤血管生成的作用有关[28]。例如, 常用于治疗乳腺癌的清毒颗粒由多味中药组成, 其中黄芪和甘草等均含有天然黄酮类化合物。Zhao[29]研究发现, 清毒颗粒能抑制裸鼠人乳腺癌血管的新生, 通过建立大鼠眼前房荷人乳腺癌模型, 将人乳腺癌MCF-7单细胞悬液减压接种于大鼠眼前房, 结果发现, 前期逐渐变红并伴有云雾状血管形成的大鼠眼睛在后期其红色逐步消退, 进一步说明清毒颗粒能阻止肿瘤新生血管的生成, 起到抗乳腺癌的作用。另外, 研究发现清毒颗粒是通过下调促癌炎症信号来抗乳腺癌血管新生的。通过分离培养人脐静脉血管内皮细胞 (human umbilical vein endothelial cells, HUVECs), 并测定回归细胞的VEGF刺激内皮增殖的量效关系, 然后以培养细胞接触性抑制的时效与量效, 建立癌细胞-内皮共培养肿瘤血管新生离体模型, 发现清毒颗粒可通过抑制内皮活化T细胞核因子 (nuclear factor activation T, NFAT) 通路起到抗乳腺癌血管新生的作用。Huang[30]同样发现在体外HUVEC和体内鸡胚尿囊膜模型中, 杨梅素和高良姜素能够有效抑制血管生成, 通过研究这两种化合物抑制血管生成的分子

机制, 发现杨梅素和高良姜素抑制了卵巢癌细胞A2780/CP70和OVCAR-3分泌血管生成的关键调控因子VEGF, 并且减少了p-Akt、p-p70S6K和缺氧诱导因子-1α (hypoxia inducible factor-1α, HIF-1α) 的蛋白表达水平, 通过瞬时转染实验显示, 杨梅素和高良姜素可通过Akt/p70S6K/HIF-1α抑制VEGF分泌, 这些结果表明天然黄酮类化合物是具有抗肿瘤血管生成活性的物质。

1.3 增强免疫功能

免疫能力的增强不仅可以抑制由于免疫逃逸引发的肿瘤生长, 还能降低患肿瘤的概率。研究表明, 富含黄酮类化合物的白花蛇舌草提取物可通过增强免疫功能达到抗肿瘤作用[31]。Li等[32]探讨了白花蛇舌草总黄酮对宫颈癌U14荷瘤小鼠免疫功能的影响, 通过对给药组与模型对照组小鼠的胸腺指数进行比较, 发现白花蛇舌草总黄酮组小鼠的胸腺指数显著升高, 表明白花蛇舌草总黄酮可以提高动物免疫力, 起到抗肿瘤增殖作用。Che等[33]对白花蛇舌草不同部位的抗肿瘤作用进行了整理总结, 发现白花蛇舌草的水提部分能调节机体免疫, 并且发现白花蛇舌草注射液 (*Hedyotis diffusa* injection, HDI) 对于肺癌A549、肝癌HepG2等肿瘤细胞都具有抑制生长的作用, 不仅可极大提升荷瘤小鼠脾自行消除瘤细胞的能力, 还可增强荷瘤小鼠腹腔巨噬细胞的吞噬能力, 以及增加体液中免疫血清中的抗氧化物质的量[34], 即HDI具有增强荷瘤小鼠自身免疫能力的作用。石斛属植物中所含的黄酮类化合物也同样具有重要的增强免疫力的药理作用[35]。

1.4 干预信号通路

黄酮类化合物还可通过调控癌细胞内相关基因的表达而干预信号通路, 以抑制肿瘤生长。信号转导途径的异常是引起肿瘤细胞生长速度变缓的原因之一。介导凋亡的信号转导途径有外部途径和内部途径, 外部途径包括死亡受体介导途径, 内部途径包括线粒体介导的途径[36]。Luo等[37]使用实时荧光定量PCR法测定相关基因mRNA的表达, 结果显示, 新西兰牡荆苷II对肝癌HepG2细胞凋亡相关基因的mRNA表达有影响, 表现为通过调控丝裂原活化蛋白激酶 (mitogen-activated protein kinase, MAPK)、Bax/Bcl-2和caspase信号通路等靶点, 诱导HepG2细胞发生凋亡, 主要提高了MAPK凋亡信号通路中关键基因细胞外信号调节激酶 (extracellular signal-related kinase, *ERK*)、c-Jun氨基末端激酶 (c-Jun N-terminal kinase, *JNK*)、蛋白38 (protein 38, *p38*) 和核因子κB (nuclear factor kappa-light-chain-enhancer of activated B cells, *NF-κB*) 的mRNA表达水平。异泽兰黄素来源于艾属植物种, 它的一种有效成分是黄酮类化合物, Wang[38]利用蛋白质印迹法检测了异泽兰黄素作用于食管癌细胞TE1的蛋白激酶B-糖原合成酶激酶-3β (protein kinase B-glycogen synthase kinase-3β, Akt-GSK3β) 和MAPK/ERK信号通路的变化, 结果发现, 异泽兰黄素可以通过降低信号通路Akt-GSK3β中Akt及其下游GSK3β的磷酸化水平, 达到抑制Akt-GSK3β信号通路的作用, 并随着异泽兰黄素浓度的增高, Akt及其下游GSK3β的磷酸化水平依次呈现下降趋势。异泽兰黄素也可以通过降低MAPK/ERK信号通路中磷酸化ERK水平, 起到抑制肿瘤生长的作用。随着科学家们对中草药抗肿瘤作用研究的深入, 人们把研究方向逐渐转向针对肿瘤细胞特异性信号靶点药物开发。Sun等[39]综合叙述了近年来天然黄酮类化合物在肿瘤细胞生长增殖、血管生成与侵袭转移、细胞凋亡、DNA与染色体调节和多药耐药性产生等过程中的主要作用信号靶点, 总结出天然黄酮类化合物主要通过作用于磷脂酰肌醇3激酶 (phosphatidylinositol 3 kinase, PI3K)/Akt、癌蛋白 (Ras protein, Ras)/MAPK、JAK (Janus kinase)/信号传导及转录激活因子 (signal transducer and activator of transcription, STAT)、分泌型糖蛋白Wnt/β-链蛋白 (β-catenin) 等靶点, 达到抑制肿瘤细胞生长的作用。Hong等[40]对中药抗肿瘤的相关实验进行归纳, 总结出具有抗肿瘤作用的中药主要涉及了PI3K/Akt/哺乳动物雷帕霉素靶蛋白 (mammalian target of rapamycin, mTOR)、Hedgehog、ERK、JNK、p38 MAPK、活化B细胞的NF-κB、Notch、p53、STAT3、转化生长因子-β (transforming growth factor β, TGF-β) 和Wnt等11种信号通路, 中药中的黄酮类化合物作用于这些信号通路, 从而达到抑制肿瘤生长的作用。

2 代表性天然黄酮类化合物的抗肿瘤作用

我国天然产物资源丰富, 其中具有抗肿瘤活性的天然产物资源更是种类繁多,我国科研人员对于天然抗肿瘤药物的开发也在不断的探索中[41]。其中, 白杨素、甘青虎耳草、乌腺金丝桃、黄芪和芒柄花素这几种具有代表性的天然黄酮类化合物具有一定的抗肿瘤活性。表2中归纳总结了这5种天然黄酮类化合物的体内外抗肿瘤作用的临床前研究进展。

2.1 黄酮类天然黄酮化合物

白杨素 (chrysin, 图3) 又名白杨黄素 (5,7-二羟基黄酮), 是一种天然黄酮类化合物, 因其广泛的药理活性备受青睐, 可以从木蝴蝶 (一种紫薇科植物) 的种子和茎皮中提取, 具有抗肿瘤活性。Li等[42]指出作用于激活蛋白-1 (activator protein-1, AP-1) 信号通路的抗肿瘤天然产物中包括白杨素, AP-1参与细胞存活、增殖

Table 2 Advances in preclinical studies of antitumor effects of compounds chrysin, quercetin, hypericin, formononetin, and calycosin-7-*O*-glucoside *in vitro* and *in vivo*. ALP: Alkaline phosphatase; GGT: γ-Glutamyl transpeptadase; AFU: α-*L*-Fucosidase; Bcl-2: B-cell lymphoma-2

Compound	*In vitro* study	*In vivo* study
Chrysin	Human gastric cancer MGC-803 cells: cell cycle arrest in G0/G1 phase; down-regulated expression of Bcl-2; up-regulated expression of Bax. Breast cancer cell T47D: reduce local recurrence	S180 tumor-bearing mice: a positive correlation between tumor inhibition and drug concentration; no damage to mouse immunity; promote hematopoietic function
Quercetin	Human liver cancer cells H22: scavenging of reactive oxygen; inhibition of cell damage caused by peroxidation	Mice inoculated with H22 liver cancer cells: tumor mass reduction; decrease ALP, GGT, and AFU expression; protect liver function; has antioxidant ability; longer survival time
Hypericin	Human liver cancer cell line SMMC7721 and osteosarcoma cell line U2OS; the inhibitory activity is strong. Human lung cancer cell line H460: up-regulated expression of Bax; down-regulated expression of Bcl-2; activated caspase-3	Myocardial ischemia-reperfusion injury in rats: improve the pathological morphology of myocardium; reduce apoptosis of cardiomyocytes
Formononetin	Prostate cancer cell PC-3: cell cycle arrest in G1 phase; act on the MAPK signaling pathway. Osteogenic sarcoma: inhibit U2OS cell proliferation	Rat model of *in situ* brain glioma: inhibit tumor growth; human gastric cancer cell MKN-45 tumor-bearing nude mouse mode: activate the MAPK signaling pathway; human prostate cancer cell PC-3 tumor-bearing nude mouse mode: down-regulated protein levels of cyclin D1 and CDK4
Calycosin-7-*O*-glucoside	Hepatocellular carcinoma cell line BEL-7402: cell cycle arrest in G0/G1 phase. Human cervical cancer HeLa cells: increase the activity of caspase-3; down-regulated expression of Bcl-2; up-regulated expression of Bax	C57BL/6 Lewis lung cancer model in mice: inhibit tumor volume growth; increase thymic and splenic index. Zebrafish: inhibition of vascular growth

和分化等重要生物学过程, 在恶性肿瘤浸润和转移中发挥关键作用, 白杨素通过下调AP-1信号通路因子表达发挥抗肿瘤转移作用[43]。对白杨素的结构进行修饰和功能性改善, 以及对其抗肿瘤作用进行探索, 仍是当前黄酮类化合物抗肿瘤药物研究的热点。

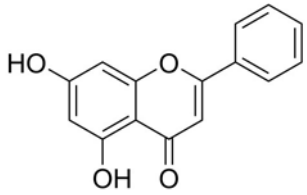

Figure 3 Structure of chrysin

白杨素有许多生物活性, 但溶解度和生物利用度较差。为了改善这一情况, Sathishkumar等[44]使用白杨素作为直接生物还原剂和封端剂, 配制出了具有高稳定性的纳米药物, 相比于白杨素具有良好的血液生物相容性和更高的体外抗癌活性。Zheng等[45]也设计合成了PEG-白杨素偶联的新型纳米粒, 为制备抗肿瘤药物递送系统提供了新策略。在白杨素母核上加入氨基酸并引入氟元素等其他基团合成白杨素衍生物, 以增强和改善其活性, 增加水溶性, 提高生物利用度[46-51]。Liu等[52]合成了白杨素的氨基酸衍生物, 发现该类化合物可介导MGC-803胃癌细胞的凋亡, 并阻滞细胞周期于G2/M期, 同时下调抗凋亡蛋白Bcl-2表达, 上调促凋亡蛋白Bax表达, 呈浓度依赖性关系, 表明该白杨素衍生物有可能成为有效的化疗药物。为增强白杨素的抗肿瘤活性, Wang等[53]合成了结构多样的2-氨基-3-氰基-苯并吡喃白杨素, 表现出了比母体化合物更强的抑制肿瘤增殖活性, 其不仅以浓度依赖性方式诱导髓性白血病K562细胞凋亡, 还可以依赖于线粒体介导的caspase激活, 及通过下调Bcl-2, 并以剂量依赖性的方式上调Bax蛋白的表达, 来促进细胞色素c和其他促凋亡因子从线粒体中的释放, 从而促进细胞凋亡DNA的形成, 导致癌细胞凋亡。Pan等[54]利用超临界反溶剂工艺辅助制备白杨素-聚乙烯-吡咯烷酮亚微粒子来提高白杨素的抗肿瘤作用。Chen等[55]用白杨素和酰氯通过酯化合成新化合物, 生成一个具有灵活结构的长链白杨素衍生物, 具有更好的溶解性, 其抗肝癌细胞增殖半数抑制浓度 (half maximal inhibitory concentration, IC_{50}) 为14.8 μmol·L^{-1}, 相比白杨素 (IC_{50}为75.0 μmol·L^{-1}) 有所增强。Sara等[56]为了减少乳腺癌的局部复发, 评估了姜黄素和白杨素共载的静电纺丝纳米纤维作为抗癌方式对T47D乳腺癌细胞的体外抗肿瘤活性, 结果表明, 多载药的纳米纤维在体外对肿瘤细胞生长的抑制具有更好效果。

2.2 黄酮醇类天然黄酮化合物

2.2.1 甘青虎耳草 甘青虎耳草为虎耳草科虎耳草属植物, 是一种多年生草本植物, 生长于海拔 (3～5)×10^3 m的林下、灌丛和高山草甸等地, 它富含槲皮素 (quercetin, 图4) 及其衍生物等黄酮类物质, 研究表明, 甘青虎耳草具有抑菌、抗病毒、抗炎和抗肿瘤等作用[57]。为探究甘青虎耳草的抗肝癌作用, Cui等[58]对甘青虎耳草进

行乙醇粗提后用乙酸乙酯萃取制备其总黄酮, 将小鼠接种H22肝癌细胞获得原位移植模型, 通过口服灌药检测该总黄酮对抑制小鼠肝癌指标变化、血清生化指标和对肝组织抗氧化性能的影响, 结果表明甘青虎耳草黄酮可显著降低*α*-*L*-岩藻糖苷酶 (*α*-*L*-fucosidase, AFU)、碱性磷酸酶 (alkaline phosphatase, ALP) 和谷氨酰转肽酶 (*γ*-glutamyl transpeptadase, GGT) 等肝癌诊断指示酶的表达, 具有较强抗氧化能力, 并可延长小鼠生存时间, 起到肝保护作用。

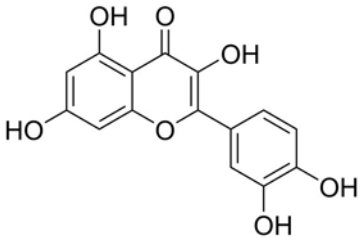

Figure 4　Structure of quercetin in *Saxifraga tangutica* Engl.

肿瘤的发生和发展与自由基在体内的富集有关, 自由基能引起细胞的脂质过氧化, 使细胞DNA发生交联或断裂, 引发癌变发生。生物活性物质可以提高机体内源性抗氧化酶的活性并清除活性氧来降低活性氧浓度, 抑制脂质过氧化所致的细胞破坏, 达到抗肿瘤细胞生长作用。综上所述, 甘青虎耳草中黄酮衍生物具有体内抗肿瘤活性, 其作用机制与提高机体抗氧化能力有关。

2.2.2　乌腺金丝桃　乌腺金丝桃属于金丝桃属植物, 主要化学成分为黄酮类、挥发油和间苯三酚衍生物类, 其中黄酮类成分包含的槲皮素、山柰酚以及金丝桃苷 (hypericin, 图5) 均具有一定的抗肿瘤活性[59]。含有金丝桃属植物成分的产品具有许多药用价值, 例如具有抗菌消炎、抗病毒、抗肿瘤细胞增殖、镇痛和收敛等生理活性, 还可用于治疗抑郁症、肝炎和痢疾等疾病[60]。

乌腺金丝桃中总黄酮的提取和抗肿瘤活性的研究越来越受到广大科学工作者的关注。Liu等[61]以乌腺金丝桃中总黄酮的提取率为评价指标, 在优选提取工艺方面做了有意义的工作。Jin等[62]利用超声方法对乌腺金丝桃内的总黄酮进行提取, 并对提取液的抗氧化活性进行了测定, 发现其对不同种类自由基表现出不同程度的自由基清除能力。研究发现, 乌腺金丝桃地上部分提取物L (新型多聚酰化间苯三酚类化合物) 对肺腺癌细胞A549和乳腺癌细胞MCF-7具有抑制作用, IC_{50}值分别为3.9和4.3 μmol·L^{-1}[63]。Zhou等[64]从乌腺金丝桃植物中分离得到6个新的结构复杂的多环聚戊二烯酰化酰基间苯三酚A～F及12个类似物, 发现其对人肝癌细胞系SMMC7721和人骨肉瘤细胞系U2OS有抑制作用, IC_{50}值分别为10.1和10.6 μmol·L^{-1}。Li[65]研究发现, 乌腺金丝桃对人肺癌H460细胞的增殖具有显著抑制, 其可通过提高Bax和降低Bcl-2的表达激活caspase-3以诱导H460细胞的凋亡。

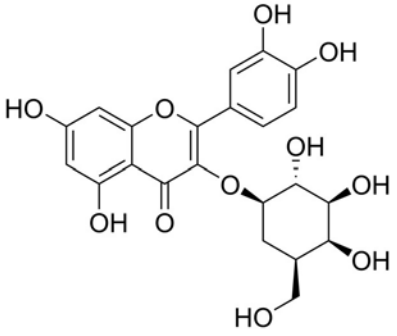

Figure 5　Structure of hypericin

对乌腺金丝桃的研究不单局限于其总黄酮提取物, 将其与其他药物配伍的混合物研究也受到关注。例如, Li等[66]发现乌腺金丝桃与当归配伍对大鼠心肌缺血再灌注损伤 (myocardial ischemia reperfusion injury, MIRI) 具有保护作用, 可通过上调模型大鼠心肌Bcl-2蛋白表达和下调Bax蛋白表达明显改善模型大鼠心肌病理形态, 减少心肌细胞凋亡。Wang[67]发现尖叶假龙胆与乌腺金丝桃配伍后的提取物可抑制A549细胞增殖, 且尖叶假龙胆与乌腺金丝桃2:1配伍组抑制作用最强, 可明显上调Fas、FasL、caspase-8、caspase-3、Bax、Cyt-c及caspase-9蛋白表达并下调Bcl-2蛋白表达, 从而诱导A549细胞的凋亡。

2.3　异黄酮类天然黄酮化合物

2.3.1　芒柄花素　芒柄花素 (formononetin, FN, 图6) 又称芒柄花黄素和刺芒柄花素 (7-羟基-4'-甲氧基异黄酮), 是一种异黄酮类植物雌激素, 可从豆科植物红车轴草和刺芒柄花全草中提取, 是豆科植物红三叶草的主要成分, 具有良好的抗肿瘤作用, 可以预防乳腺癌、前列腺癌和结肠癌, 可通过浸提、超声波、微波、超临界、亚临界水和加压法等方法从天然植物中提取, 也可以通过人工方法进行制备[68]。

刺芒柄花素的药理作用广泛, 但其生物利用度较低。为了寻找合适的载药系统, 提高芒柄花素生物利用度, Liu等[69]选用水溶性较好的羟丙基-*β*-环糊精共

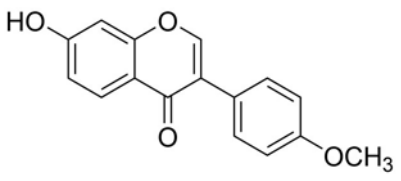

Figure 6　Structure of formononetin

价修饰已羧基化的单壁碳纳米管和多壁碳纳米管, 对FN进行载药实验, 结果显示, 经载药后的FN对肿瘤细胞具有更好抑制作用[70]。Lei[71]揭示了USP5蛋白对Slug的去泛素化及其在肝癌恶性演化中的作用, 发现芒柄花黄素能靶向USP5并抑制其去泛素化Slug而抑制肝癌细胞EMT。Ren等[72]发现芒柄花素氮芥衍生物可诱导肿瘤细胞凋亡。Fu等[73]发现芒柄花素二硫代氨基甲酸酯衍生物可抑制PC-3前列腺癌细胞生长, 通过作用于MAPK信号通路阻滞细胞在G1期而抑制其生长。

Liu等[74]对芒柄花素诱导肿瘤细胞凋亡、阻滞细胞分裂周期、抑制肿瘤侵袭迁移, 以及逆转肿瘤多药耐药性等方面做了总结。芒柄花素在抗乳腺癌, 以及女性生殖系统、泌尿系统和消化系统恶性肿瘤方面, 显示出了优良的生物活性[75]。Liu[76]发现具有抗脑神经胶质瘤作用的九味通窍汤的有效成分芒柄花素可抑制大鼠原位脑神经胶质瘤肿瘤细胞的生长。Dong等[77]发现芒柄花黄素可抑制人胃癌细胞MKN-45增殖, 其机制与激活细胞内MAPK信号转导通路而诱导细胞凋亡有关。Zhao[78]也发现芒柄花黄素对裸鼠体内移植瘤的增殖具有显著抑制, 其作用可能与细胞内cyclin D1和CDK4的mRNA和蛋白水平下调有关。Hu[79]发现芒柄花黄素可显著抑制骨肉瘤U2OS细胞增殖, 且可诱导其凋亡, 可能是通过上调Bax及下调Bcl-2和miR-375的表达实现的。

2.3.2 黄芪 黄芪中的皂苷、黄酮和多糖等化合物具有广泛药用价值, 毛蕊异黄酮葡萄糖苷 (calycosin-7-*O*-glucoside, 图7) 是从中药黄芪的干燥根中提取的, 是具有广泛药理活性的单体成分, 属于异黄酮类化合物。它不仅能直接抑制肿瘤细胞增殖, 诱导肿瘤细胞凋亡, 还能通过增强机体免疫力来抵抗放化疗的毒副作用, 提高患者生存率[80]。Yang等[81]发现, 黄芪中含有叶酸、胡萝卜素、多糖、黄酮、异黄酮、氨基酸、维生素D、皂苷、三萜类化合物以及钙铁硒微量元素等多种有效成分, 能有效增强机体细胞免疫和体液免疫功能, 加强对肿瘤细胞的杀伤和生长抑制作用, 从而导致肿瘤细胞凋亡。

黄芪和黄芪类化合物与化疗药物联合使用时, 可以提高抗肿瘤药物的疗效, 同时减少化疗并发症。研究发现, 由人参、黄芪和苦参制成的康艾注射液具有抑制肿瘤细胞增殖、促进肿瘤细胞凋亡、调节肿瘤血管生成, 以及提高人体免疫能力的生物效应, 这种注射液也可以通过影响肿瘤生长的微环境来抑制肿瘤生长, 体现了中药抗肿瘤药物多成分、多靶点、多通路的特点[82]。人们发现黄芪能够通过对肿瘤细胞的直接抗增殖或促凋亡作用来抑制肿瘤生长[83], 其还能通过激活肿瘤微环境中M1巨噬细胞和T细胞的肿瘤杀伤功能来改善免疫系统。黄芪不仅能提高全身免疫力, 对提高化疗效果和预防肿瘤转移也有一定的疗效。Zhang等[84]将黄芪与苦参和皂角刺的抗肝癌作用进行对比后发现, 黄芪在提高机体免疫功能方面尤为显著。Yang等[85]构建了C57BL/6小鼠Lewis肺癌模型, 采用VisualSonics-Vevo2100小动物专用高频彩超评估了黄芪中的黄酮组分对肿瘤体积的抑制作用, 发现黄芪中的黄酮对肿瘤增长有明显抑制作用, 并发现荷瘤小鼠的胸腺和脾腺指数均有明显提高, 为黄芪中的黄酮组分能够通过调节机体免疫功能抑制荷瘤小鼠肿瘤生长的观点提供了新证据。Ni等[86]应用斑马鱼模型对黄芪等11种中药提取物进行的研究发现, 黄芪具有抑制血管生长作用。

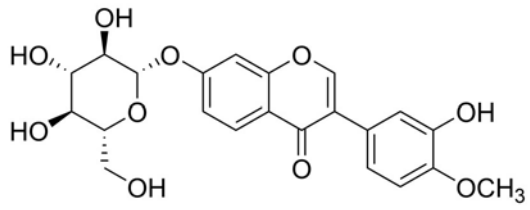

Figure 7 Structure of calycosin-7-*O*-glucoside

黄芪同样也可以通过阻断信号通路的方式抗肿瘤。Zhang等[87]研究发现, 黄芪总黄酮及其单体化合物具有抑制肝癌细胞株BEL-7402生长的作用, 药物处理组细胞G0/G1期比例明显增加, S期细胞明显减少。毛蕊异黄酮葡萄糖苷可通过下调重组人细胞周期素A2 (recombinant human cyclin-A2, CCNA2)、周期蛋白依赖性激酶2 (cyclin-dependent kinase 2, CDK2) 和细胞周期蛋白B1 (mitotic-specific cyclin-B1, CCNB1) 等蛋白水平, 将肝癌BEL-7402细胞周期阻滞在G0/G1期。Zhang等[88]研究了毛蕊异黄酮-7-*O*-*β*-*D*-葡萄糖苷 (calycosin-7-*O*-*β*-*D*-glucoside, CG) 对人宫颈癌HeLa细胞的作用, 发现CG可通过增加caspase-3活性、下调Bcl-2蛋白表达并同时上调Bax表达来抑制宫颈癌细胞增殖并诱导其凋亡。这些研究为黄芪的抗肿瘤作用机制提供了合理的解释。

3 总结与展望

本文主要介绍了黄酮类化合物的抗肿瘤作用及其作用机制。各类天然植物中所含的不同黄酮类化合物均表现了不同的抗肿瘤活性, 而同一黄酮类化合物对不同的肿瘤也显示出不一样的活性, 在肿瘤细胞生长、扩散等多个阶段均能发挥有效作用。当前的主要研究方向主要集中在对天然黄酮类化合物的结构修饰和优化方面, 而与其他抗肿瘤药物联合应用则可能取得更有临床意义的效果。黄酮类化合物及其衍生物有可能

开发成为一类新型的抗肿瘤药, 其相关作用机制和靶点、毒性研究以及其他生物学活性, 仍然值得人们做进一步的研究与探索。

作者贡献: 李鑫萍构建了全文; 于洣璇和况婷瑞对内容做了补充工作; 延玺、李春颖和郝海军审阅了全文并提出了合理的修改意见。

利益冲突: 所有作者均声明不存在利益冲突。

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