基于有氧糖酵解的天然小分子抑制剂的研究进展

罗煜瑾,戴子琦,徐　冰*,雷海民*

北京中医药大学中药学院,北京 102488

摘　要:有氧糖酵解作为恶性肿瘤最显著的能量代谢特征之一,肿瘤细胞中大约有 50% 的 ATP 是通过有氧糖酵解途径合成的,同时糖酵解过程中产生的各种中间代谢产物也是合成蛋白质等生物大分子重要的原料来源。此外酵解途径导致的乳酸增加为肿瘤细胞提供了一个酸性成长环境,有利于其浸润和转移,因此其在维持肿瘤细胞能量需求、合成代谢平衡和肿瘤浸润和转移方面发挥着重要作用。研究表明有氧糖酵解的过程与葡萄糖转运蛋白、己糖激酶、丙酮酸激酶、磷酸果糖激酶等密切相关。目前靶向有氧糖酵解相关转运蛋白和关键限速酶已经成为抗肿瘤药物研发的有效途径,本文对目前天然产物中靶向有氧糖酵解相关蛋白的小分子抑制剂研究最新进展及作用机理进行总结,以期为相关领域药物研究人员提供新的思路和参考。

关键词:有氧糖酵解;小分子抑制剂;葡萄糖转运蛋白;己糖激酶;丙酮酸激酶;磷酸果糖激酶

中图分类号:R285;R73-36　　**文献标识码**:A　　**文章编号**:1001-6880(2023)1-0171-11

DOI:10.16333/j.1001-6880.2023.1.018

Research progress of natural small molecule inhibitors based on aerobic glycolysis

LUO Yu-jin, DAI Zi-qi, XU Bing*, LEI Hai-min*

School of Chinese Materia Medica, Beijing University of Chinese Medicine, Beijing 102488, China

Abstract: Aerobic glycolysis is the most prominent feature of energy metabolism in malignant tumors. About 50% of ATP in tumor cells was synthesized through aerobic glycolysis. At the same time, various intermediate metabolites produced in the process of glycolysis were also important raw materials for the synthesis of proteins and other biological macromolecules. In addition, the increase of lactic acid caused by glycolysis pathway could provide an acidic growth environment for tumor cells, which was conducive to their infiltration and metastasis. Therefore, it played an important role in maintaining the energy demand of tumor cells, the balance of anabolism and tumor invasion and metastasis. Studies have shown that the process of aerobic glycolysis was closely related to glucose transporter, hexokinase, pyruvate kinase and phosphofructokinase. At present, targeting aerobic glycolysis-related transporters and key rate-limiting enzymes have become an effective way for the development of antineoplastic drugs. This paper summarizes the latest progress and mechanism of action of small molecule inhibitors targeting aerobic glycolysis-related proteins in natural products, with a view to providing new ideas and references for drug researchers in related fields.

Key words: aerobic glycolysis; small molecule inhibitor; glucose transporters; hexokinase; pyruvate kinase; phosphofructokinase

恶性肿瘤仍是目前严重威胁人类生命和健康的重要疾病。肿瘤细胞表现出异常的能量代谢特征,其即使在氧气充足的环境里,仍使用糖酵解的方式来提供能量,肿瘤细胞中大约有 50% 的 ATP 是通过有氧糖酵解途径合成的。肿瘤细胞不仅能够利用糖酵解通路的中间产物为合成代谢提供原料,而且糖酵解途径导致的乳酸增加可以为肿瘤细胞提供了一个酸性成长环境,有利于其浸润和转移[1-4]。肿瘤细胞中糖酵解活跃的机制复杂,其有多种因素综合作用引起的,主要包括有利于糖酵解的跨膜结构、糖酵解关键酶代谢异常、癌基因及信号传导通路异常等,而调整糖酵解酶,是糖酵解活性增强、过表达的最主要途径。研究表明葡萄糖转运蛋白(GLUTs)可促进葡萄糖在细胞膜上的转运,而丙酮酸脱氢酶激酶

收稿日期:2022-05-11　　接受日期:2022-09-26

基金项目:科技部重大专项(2020YFF0426288);中央高校基本科研经费(2020-JYB-XJSJJ-007);国家自然科学基金(81173519);中国科学院青年人才培养项目(CACM-2018-QNRC2-B08)

*通信作者 Tel:86-10-84738641;E-mail:hm_lei@126.com,weichenxubing@126.com

(PDK)、乳酸脱氢酶 A(LDHA)、丙酮酸激酶(PKM)、己糖激酶(HK)均为糖酵解过程的关键限速酶,且在肝癌、肺癌和乳腺癌等多种恶性肿瘤上高表达[5,6]。此外抑制有氧糖酵解可以有效抑制肿瘤细胞增殖,促进肿瘤细胞凋亡;通过改变缺氧诱导的代谢开关,靶向这些特定的酶来降低肿瘤细胞的活性,从而逆转 Warburg 效应已成为了抗肿瘤的重要途径[7]。

研究表明,葡萄糖转运体(GLUT)家族 14 个成员都能转运己糖和多元醇。GLUT1 ~5 在各种组织和细胞类型中都具有葡萄糖和/或果糖转运蛋白的作用。有氧糖酵解的代谢过程(见图 1)表明,在大部分肿瘤中缺氧的微环境能够诱导 GLUT1 的高表达,进而提高肿瘤细胞对葡萄糖的摄取能力,该过程是肿瘤细胞产生 Warburg 效应的基础。己糖激酶(HK)是糖酵解过程中的第一个限速酶,磷酸果糖激酶 1(PFK1)是糖酵解过程中的第二个限速酶,其活性被磷酸果糖激酶-2/果糖-2,6-二磷酸酶(PFKFB3)调控,丙酮酸激酶(PK)有 4 种同工酶,分别是 M1、M2、L 和 R 型,是糖酵解过程中的第三个限速酶,其中 PKM2 在肿瘤组织中广泛高表达。而乳酸脱氢酶(LDH)催化糖酵解过程中的最后一步——乳酸和丙酮酸的相互转化过程[8,9]。肿瘤细胞主要表达乳酸脱氢酶亚型 LDHA,LDHA 在肿瘤中升高除了促进糖酵解,还可以促进乳酸的产生从而重塑肿瘤微环境[1-3,10]。本文对以上相关蛋白及其天然小分子靶向抑制剂的研究最新进展及作用机理进行总结,为相关领域研究提供参考。

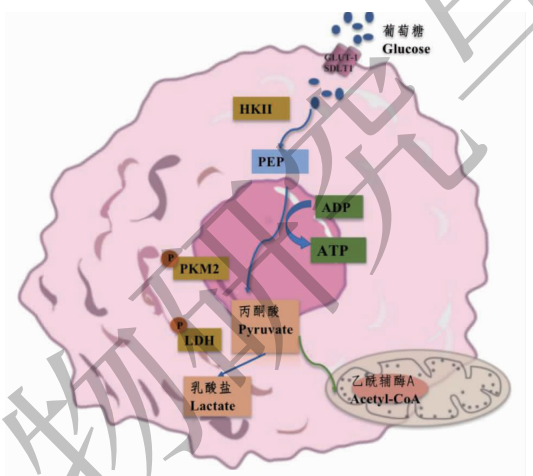

图 1 有氧糖酵解过程

Fig. 1 Aerobic glycolysis process

1 有氧糖酵解相关蛋白及其小分子抑制剂

1.1 葡萄糖转运蛋白(glucose transports,GLUTs)及其抑制剂

肿瘤细胞糖酵解的第一个限速步骤是葡萄糖在 GLUT 的帮助下通过细胞膜进入细胞进行氧化[11]。葡萄糖转运体是一种跨膜糖蛋白,它分布在细胞膜上,介导葡萄糖在细胞膜两侧的跨膜转运。相比于正常细胞的氧化磷酸化,肿瘤细胞的有氧糖酵解是一种低效能的能量代谢方式。因此,为了满足快速生长的需求,肿瘤细胞增加了 GLUT1 的表达,增加了葡萄糖的吸收,为其能量代谢提供了丰富的原料,从而促进其自身的增殖和浸润。在目前已发现的 14 种 GLUTs 亚型中,GLUT1 在多种癌症中呈异常高水平表达,包括肝、结肠、卵巢等癌症,表达水平均达到甚至超过 50%。研究表明 GLUT1 在人肠腺癌细胞、胃癌、肺癌等肿瘤细胞系的表达量显著高于正常细胞系[12-14],其表达水平的升高在肿瘤细胞的分化和自我更新中起关键作用[15-17]。

Hsieh[18] 和 Zhou 等[19] 发现黄酮类化合物根皮素(phloretin,**1**,见图 2)可以通过抑制 GLUT 直接诱导癌细胞的凋亡。低剂量的根皮素与化疗药物联用可以显著提高其抗肿瘤效果。除此之外,天然黄酮类化合物中,Zhan[20] 发现水飞蓟宾(silybin,**2**)也具有 GLUT 抑制作用。芹黄素(apigenin,**3**)一种广泛存在于水果中的黄酮类化合物,研究表明其具有良好的抗肿瘤活性,在低氧条件下,芹黄素的抗卵巢癌活性可能与其抑制 HIF-1α 的合成进而下调 SKOV3 的 GLUT1 的表达发挥抗肿瘤活性[21]。

青蒿素是一种由中国科学家从青蒿中提取分离得到的倍半萜内酯类化合物,其不仅具有极好的抗

疟活性,而且其还能显著抑制多种恶性肿瘤的增殖。Li 等[22]研究显示其衍生物双氢青蒿素(dihydroartemisinin,**4**)调节糖酵解抑制癌细胞生长,通过下调 GLUT1 和 PKM2 的表达发挥抗肿瘤活性,同时其与其他抗肿瘤药物不产生交叉耐药性。

近年研究发现,白花丹素(plumbagin,**5**)具有抑制肿瘤细胞增殖和迁移的活性,可呈浓度和时间依赖性地通过阻断 PIK3/AKT 信号通路来抑制直肠癌细胞中 GLUT1 的表达,从而发挥抗直肠癌活性[23]。

也有研究表明天然糖类具有抑制 GLUT1 的作用,Chang 等[24]研究发现,银杏多糖和银杏外种皮多糖能够从转录水平上抑制 GLUT1 基因表达从而抑制乳腺癌细胞 4T1 增殖。Wang 等[25]研究发现,甘露糖(mannose,**6**)可在一定剂量范围内通过抑制直肠癌细胞内 GlUT1 的表达,体外抑制结直肠癌细胞 HCT116 的增殖。

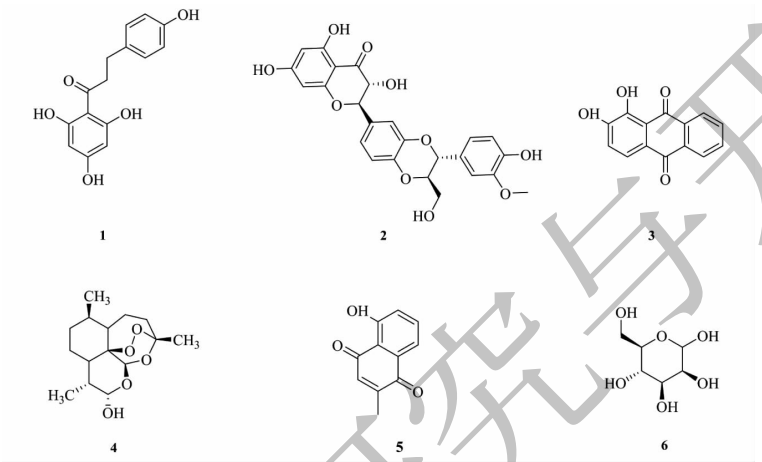

图 2　GLUTs 抑制剂结构

Fig. 2　Structures of GLUTs inhibitors

1.2　己糖激酶(hexokinase,HK)及其抑制剂

己糖激酶是肿瘤细胞有氧糖酵解的第一个关键的速度限制酶[26],负责催化葡萄糖磷酸化生成 6-磷酸葡萄糖,促进有氧糖酵解,为戊糖磷酸途径提供原料[27]。HK 家族有四种亚型,包括 HKI ~ IV,研究发现 HK II 在肝癌、结肠癌和卵巢癌等多种肿瘤细胞中高表达;与恶性肿瘤细胞的异常能量代谢和增殖中起着关键作用,其能抑制促凋亡因子的释放,有利于肿瘤细胞逃逸凋亡,促进肿瘤的发展[28-30]。通过有效抑制 HK II 的活性进而抑制肿瘤细胞的有氧糖酵解,诱导肿瘤细胞凋亡,能有效杀死肿瘤细胞。靶向 HK II 的治疗恶性肿瘤的策略可以分为抑制 HK II 的活性、抑制 HK II 的表达、抑制 HK II 与线粒体结合[31-35]。

2-脱氧葡萄糖(2-deoxy-*D*-glucose,2-DG,**7**,见图 3)为 2-OH 被氢取代的葡萄糖类似物,能与葡萄糖竞争 HK II;HK II 可以将其磷酸化生成 2-脱氧-*D*-葡萄-6-磷酸酯(2-DG-P),而 2-DG-P 则无法被代谢,在机体内蓄积,因此其能显著性抑制细胞糖酵解,使细胞内 ATP 耗竭,破坏蛋白质 N 糖基化,进而杀死肿瘤细胞[10,36]。

小檗碱又名黄连素(berberine,BBR,**8**),来源于小檗科、罂粟科、芸香科、防己科等的一种异喹啉类生物碱。近年来,许多研究表明其对多种肿瘤具有抑制作用。Ren[37]研究发现 BBR 通过抑制乳腺癌细胞 HK2 的表达,从而影响肿瘤细胞糖酵解的发生进而抑制乳腺癌细胞的增殖。Wu 等[38]发现 BBR 对 A549 的增殖活性的影响与其剂量相关,在高浓度下其通过抑制 A549 的 EMT 过程抑制肿瘤细胞迁移,并显著下调 HK II 的基因表达,BBR 对于 EMT 的抑制作用具有 HK II 依赖性。

麦冬皂苷 B(ophiopogonin B,OP-B,**9**)是由麦冬中分离出的单体物质,可抑制多种癌症细胞株的增殖活性。Bai 等[39]研究发现 OP-B 可直接抑制 HK II 蛋白的表达,并上调人肺癌 A549 细胞中促凋亡蛋白 BAX 表达。当 HK II 活性较低时,BAX 与线粒体外膜通道蛋白 VDAC 结合增多,从而削弱 A549 的增殖能力。

薯蓣次苷 A(prosapogenin A,PSA,**10**)是来源于中药藜芦中的单体化合物,Wang 等[40]的研究发现 PSA 可明显抑制人乳腺癌 MCF7 细胞株的增殖,通

过下调转录因子 STAT3 进而明显降低 MCF7 中 HK II 的水平,从而抑制其糖酵解过程使细胞增殖活性明显降低。

白头翁皂苷(Pulsatilla chinensis,**11**)为中药白头翁中具有抗肿瘤活性的主要成分。Luo 等[41]研究表示高剂量白头翁皂苷可通过抑制 HK II 表达明显降低肿瘤组织中 HK II 的水平,降低糖酵解的速度从而抑制肝癌细胞的增殖。

20(*S*)-人参皂苷 Rg_3(20(*S*)-ginsenoside Rg_3,**12**)是中药人参的药理活性成分之一,研究表明其可通过调节己糖激酶 2(HK II)和丙酮酸激酶 M2(PKM2)抑制卵巢癌细胞的糖酵解,其与抗肿瘤药物顺铂联合应用可显著增强顺铂类药物的抗癌作用,降低其耐药性,减小其毒副作用[42-44]。

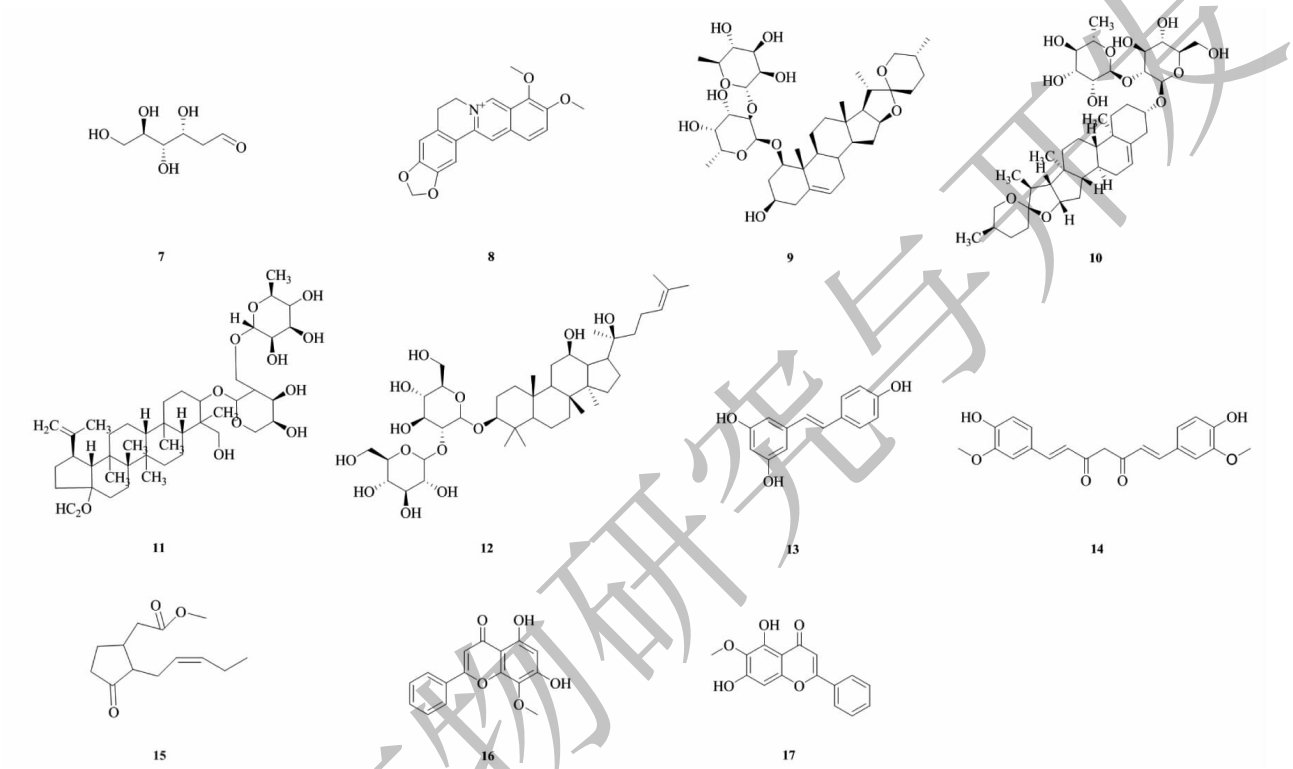

图 3 HK II 抑制剂结构

Fig. 3 Structures of HK II inhibitors

白藜芦醇(resveratrol,**13**)是存在于桑树和花生等植物中非黄酮类多酚化合物,研究发现白藜芦醇在体内外均对非小细胞肺癌(NSCLC)表现出很强的抑制作用,其可通过 Akts 信号通路介导,降低 NSCLC 中 HK II 表达,抑制了糖酵解的发生,从而抑制 NSCLC 的增殖[45-47],此外白藜芦醇也可减轻肝细胞癌 HCC 对索拉非尼的耐药性[48,49]。此外研究表明其二甲基衍生物紫檀芪亦可通过 STAT3/HK2 途径抑制糖酵解进而抑制卵巢癌细胞增殖[50]。

除此之外,姜黄素[51](curcumin,**14**)、茉莉酮酸甲酯(methyl jasmonate,**15**)可通过影响 VDAC 与 HK2 的相互作用从而抑制癌症细胞的生长。汉黄芩素(wogonin,**16**)可抑制缺氧介导的 PI3K/AKT、HIF-1α 信号通路活化,减少细胞葡萄糖摄取,抑制糖酵解酶 HK2、PDK1 和 LDHA 表达进而抑制肿瘤糖酵解[52]。此外研究表明千层纸素 A(oroxylin A,**17**)依赖于 SIRT3 去乙酰化亲环素 D 抑制 HK2 与线粒体 VDAC 结合,降低癌细胞的 ATP 水平发挥抗肿瘤作用[53,54]。

1.3 丙酮酸脱氢酶激酶(pyruvate dehydrogenase kinase,PDK)及其抑制剂

丙酮酸脱氢酶(pyruvate dehydrogenase,PDH)是一种限制细胞氧化磷酸化第一阶段的酶。它负责将糖酵解产物丙酮酸催化转化为乙酰辅酶 A,乙酰辅酶 A 被引入三羧酸循环,生成 ATP 供细胞使用。PDK 催化 PDH 磷酸化,抑制氧化磷酸化,使得肿瘤细胞能够适应缺氧环境,加强有氧糖酵解供能,从而促进肿瘤发展[10,55]。越来越多研究显示 PDK 在肿瘤细胞及其耐药株中受到原癌基因、转录因子及生长因子等的调节异常高水平表达,进而促进肿瘤细

胞的增殖并获得耐药[56-61]。因此,PDK 可成为一个新的杀死肿瘤细胞并提高化疗疗效靶点,若能抑制 PDK 的活性,使其不能催化丙酮酸脱氢酶磷酸化,进而促进葡萄糖氧化代谢途径,诱导肿瘤细胞的凋亡,就能达到抗肿瘤的目的。

目前文献报道了多种结构类型的 PDK 抑制剂,但是表现出理想的抗肿瘤作用的化合物却并不多,其中活性最理想的是氯代羰基类的二氯乙酸(dichloroacetic acid,DCA)(化合物 **18**,见图 4),其也是目前唯一进入 II 期临床的 PDK 抑制剂[57]。研究表明 DCA 是作为小分子化合物,其能够很好渗透组织,甚至进入大脑,具有较高的生物利用度。大量临床前体内外活性评价表明 DCA 可以其可通过抑制 PDK 活性而对胶质母细胞瘤、非小细胞肺癌和子宫内膜癌等恶性肿瘤表现出良好的杀伤力[10,60-62]。Sun 等[63]设计出一系列 PDK 的共价抑制剂,其中 JX06(**19**)在 A549 异种移植瘤模型中通过作用于细胞内的 PDK 半胱氨酸残基附近的结合位点,从而使酶的构象发生改变,降低与 ATP 的结合能力,从而抑制肿瘤的生长。

随着传统中药研究的深入,人们发现它们含有的一些活性成分也可以通过抑制 PDK 活性发挥抗肿瘤作用。Lee 等[64]发现桂皮的水提物中活性成分肉桂酸(trans-cinnamic acid,**20**)能够抑制 PDK,促进线粒体的氧化磷酸化,增加 ROS 的生成,并且通过 ROS 和线粒体依赖的凋亡途径诱导细胞发生凋亡,而对正常细胞表现出较低的细胞毒性。Tangchirakhaphan 等[65]研究发现番荔枝科植物中的抗癌活性成分 goniothalamin(**21**)可以通过同时抑制 PDK 和 AKT 的活性起到抑制黑色素瘤细胞的增殖并诱导其凋亡的作用。Chung 等[66]发现草玉梅中的有效成分可以通过抑制 PDK1 的活性,降低 PDH 的磷酸化,增加 ROS 的产生并诱导线粒体损伤,杀伤小鼠肺癌细胞。Kwak 等[67]研究表明 Huzhangoside A(**22**)可以通过抑制 PDK 的活性,降低多种人癌细胞株的细胞活力。Song 等[68]发现紫云英苷(astragalin,**23**)能够抑制 PDK 的表达,激活线粒体介导的凋亡通路促进卵巢癌细胞凋亡。

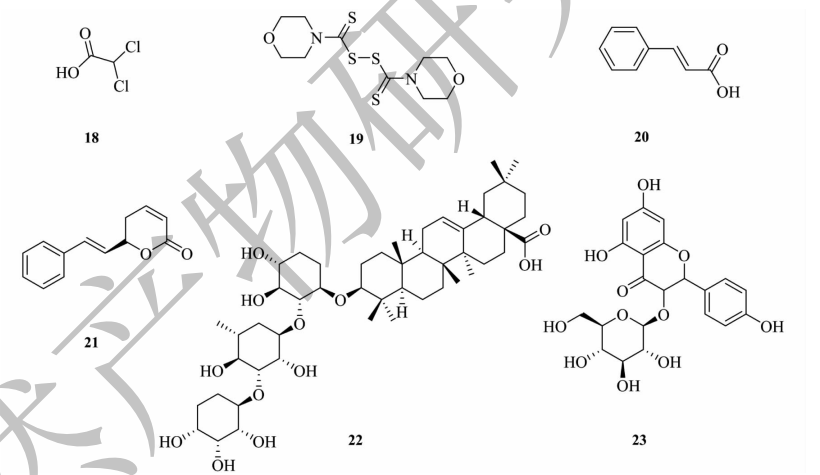

图 4　PDK II 抑制剂结构

Fig. 4　Structures of PDK II inhibitors

1.4　丙酮酸激酶(pyruvate kinase,PK)及其抑制剂

丙酮酸激酶也是肿瘤细胞有氧糖酵解过程中的一种重要的限速酶,其是去磷酸化磷酸烯醇式丙酮酸(phosphoenol pyruvate,PEP)后异构为丙酮酸的催化剂,从而累积大量丙酮酸,促进磷酸戊糖途径以及磷脂和氨基酸等的合成途径并降低细胞内 NADPH 的水平,促进肿瘤细胞增殖[10,69]。PK 存在四种异构体,分别是 L 型丙酮酸激酶、R 型丙酮酸激酶、M1 型丙酮酸激酶和 M2 型丙酮酸激酶,其中 PKM2 是胚胎表型,主要表达胚胎细胞等在合成代谢需求量高的分裂细胞中表达但它会在分化过程中逐步转化为其他亚型。此外成熟组织或细胞发生重新获得无限增殖的能力的时候,也将再次重新表达 PKM2[70,71]。研究发现当细胞发生癌变时,稳定表达的其他类型的异构体会重新转变成 PKM2,而肿瘤细胞内高表达的 PKM2 会进一步促进肿瘤细胞的异常能量代谢并参与和调控其他与肿瘤增殖有关的基因和信号通路,促进肿瘤细胞的生长、浸润和转

移。因此，PKM2 现已成为肿瘤治疗的重要靶标[72-77]。

目前研究表明，紫草素（shikonin，**24**，见图 5）是 PKM2 最有效且特异性最高的抑制剂，其能够显著抑制高表达 PKM2 肿瘤细胞的糖酵解速率，杀伤肿瘤细胞的同时逆转耐药[78,79]。Tang 和 Tao 等[80,81]发现紫草素通过抑制 PKM2 的表达抑制食管癌和膀胱癌的进展并诱导细胞凋亡。无独有偶，萘醌类化合物—维生素 K，一种人体必需的脂溶性维生素，在临床上主要用作凝血剂。近年来，体内外研究显示维生素 K 也有抑制 PKM2 发挥抗肿瘤的作用，特别是维生素 K3（vitamin K3，**25**）和 K5（vitamin K5，**26**）。Chen 等[82]发现，维生素 K3 和 K5 对 PKM 的抑制作用明显高其对于 PKM1 和 PKL。更有研究显示维生素 K3 和 K5 对结肠癌、肝癌、乳腺癌等多种肿瘤细胞和组织的杀伤效果显著强于维生素 K1 和 K2。

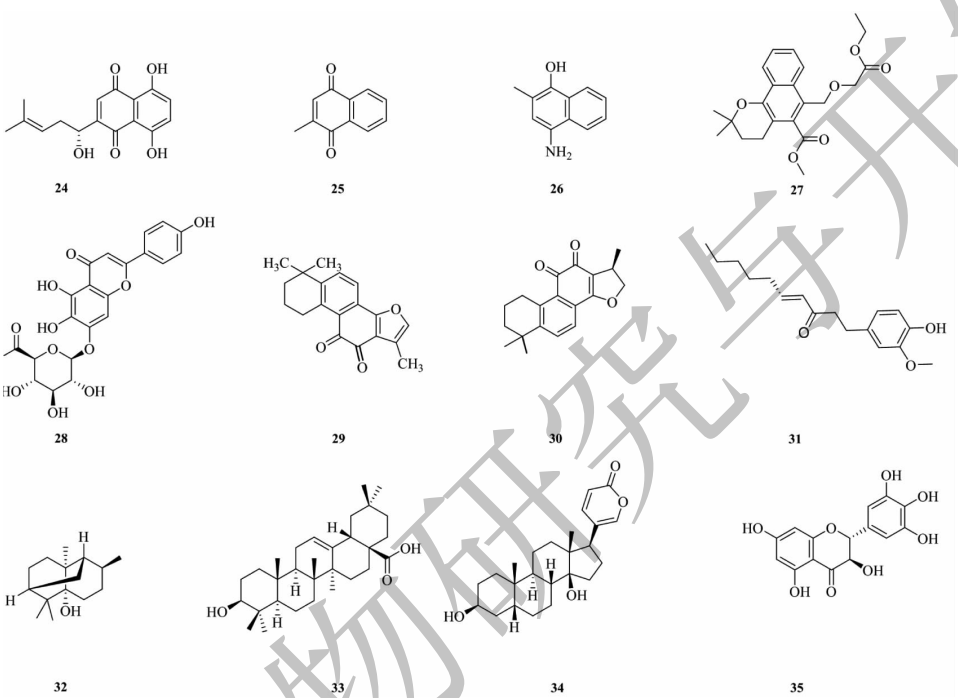

图 5 PK 抑制剂结构

Fig. 5 Structures of PK inhibitors

Wei 等[83]在中药茜草提取物的结构基础上合成了 FFJ-5（**27**），经研究发现其能下调 PKM2，抑制 MCF7、A549、HepC2 的生长并逆转 MCF7 耐药细胞对化疗药多柔比星的耐药性。Zhao 等[84]发现穿心莲有效部位 AEP 可通过抑制 PKM2 和脂肪酸合酶（fatty acid synthase，FASN）的表达诱导肿瘤细胞凋亡及坏死。

在中药方剂方面，Zhu 等[85]发现白露汤能通过抑制 GLUT1、HK2、PKM2、LDH-A、HIF-1α 的表达调控，从而调控人肺腺癌 A549 糖酵解过程，表现出对人肺腺癌 A549 明显的抑制作用并恢复其对化疗药物顺铂的敏感性。灯盏花乙素（scutellarin，**28**）是黄酮类天然化合物，研究表明其可直接靶向抑制 PKM2，降低其胞浆活性从而抑制糖酵解代谢发挥抗结肠癌作用，此外其还可通过激活 MEK/ERK/PIN1 途径参与细胞周期和凋亡过程的调节，促进 PKM2 的核转位[86]。

丹参酮中的有效成分丹参酮 II_A（tanshinoneII_A，**29**）和隐丹参酮（cryptotanshinone，**30**）均具有显著的抗肿瘤活性，Zhang 等[87]研究显示，丹参酮 II_A 可通过上调 miR-122 通过抑制食道癌细胞中 PKM2 的表达发挥抗肿瘤活性；隐丹参酮可以通过靶向抑制 PKM2/β-catenin 信号的表达发挥抗乳腺癌作用[88]。6-姜烯酚（6-gingerol，**31**）可以通过调控人胃癌 BGC-823 细胞中 PKM2 磷酸化干扰癌细胞能量代谢过程以发挥抗胃癌功效[89]。

有研究发现，三环倍半萜类化合物广藿香醇（patchouli alcohol，**32**）可以诱导 MVP 细胞的凋亡，其主要机制是下调磷酸化酶 PKM2 并抑制核转录因子 NF-κB 活性而发挥作用[90]。

五环三萜类成分齐墩果酸(oleanolic acid,**33**)具有良好的抗肿瘤活性,其抗癌活性可能与 PKM2/PKM1 介导抑制有氧糖酵解相关,且在齐墩果酸处理的癌细胞处发现被灭活的雷帕霉素靶蛋白 mTOR,而 mTOR 与诱导 PMK2/PMK1 直接相关[91]。

除此之外,天南星水提物蟾毒灵[92](bufalin,**34**)、蛇葡萄素[93](ampelopsin,**35**)和苦参碱[94](matrine,**36**)对于癌症细胞的抑制作用也均与 mTOR/PMK 信号通路直接相关。

大黄提取物中的蒽醌类有效成分大黄素(emodin,**37**)可明显降低 HepG2 细胞中 PKM2,HK2 等的表达水平有效抑制其增殖活性[95]。大黄素抑制胃癌细胞 MGC803 细胞生长的机制与抑制细胞外信号调节激酶 ERK1/2 和 PKM2 的表达,促进凋亡因子 p53 表达相关[96]。

Feng 等[97]研究发现二聚体黄酮原花青素 B2(proanthocyanidin B2,**38**)可通过抑制 PKM2 表达和核转位阻断 PKM2/HSP90/HIF-1α 的相互作用,进而抑制肝癌细胞有氧糖酵解而发挥抗肿瘤作用。

1.5　乳酸脱氢酶(lactate dehydrogenase,LDH)及其抑制剂

乳酸脱氢酶是细胞糖酵解最后阶段的一种关键的速度限制酶,其能催化 NADH 和丙酮酸发生氧化还原反应生成乳酸和 NAD^+,对形成肿瘤中具有重要作用[10]。乳酸脱氢酶由 4 个亚基构成,分别为两个 M 亚基(muscle-type,LDHA)和 2 个 H 亚基(heart-type,LDHB),其中 LDHA 亚基在有氧糖酵解里发挥关键性作用[98]。许多研究表明 LDHA 在胰腺癌、骨髓瘤和前列腺癌等肿瘤细胞中表达异常高[99-101]。Fantin 等[102]研究发现用当用 siRNA 敲除 LDHA 的表达时,在低氧条件下细胞呼吸受到抑制,线粒体膜电位降低,肿瘤细胞扩增能力降低。Le 等[103]发现用肿瘤细胞经 siRNA 或 FX11 处理后,LDHA 的活性受到抑制,ATP 的生成减少,并且发生氧化应激反应,最终导致细胞死亡。不仅如此,Zhou 等[104]还观察到紫杉醇耐药乳腺癌细胞中 LDHA 的表达和活性增加,siRNA 对 LDHA 表达的降解显著恢复了紫杉醇耐药细胞的敏感性。很明显,LDH 现在可以成为肿瘤诊断和治疗的关键目标。

LDH 的抑制剂主要是丙酮酸类似物或者 NADH 类似物,该类化合物通过发挥竞争性抑制作用遏制肿瘤细胞能量代谢,杀死肿瘤细胞的同时逆转耐药。草氨酸(oxamate,**39**,见图 6)作为 LDH 的特异性抑制剂,其是丙酮酸的结构类似物[10,104-106]。根据 Zhou 等[104]的研究显示草氨酸通过抑制 LDH 使丙酮酸无法转化为乳酸,抑制糖酵解,进而促进乳腺癌细胞的凋亡;同时与紫杉醇联用对肿瘤细胞有很好的协同抑制作用。FX-11(**40**)是和 NADH 竞争的 LDH 的选择性可逆抑制剂,在淋巴瘤和胰腺癌异种移植物中表现出明显的抗肿瘤活性[107]。

中药雷公藤中的活性成分雷公藤红素(celastrol,**41**)对于人肺癌细胞 A549 具有体外抑制作用,研究表明其能通过下调 LDHA 的表达抑制肿瘤细胞有氧糖酵解[108]。与之类似,光甘草定[109](glabridin,**42**)和甜菜碱[110](betaine,**43**)也通过抑制 LDHA 的表达发挥抑制作用。

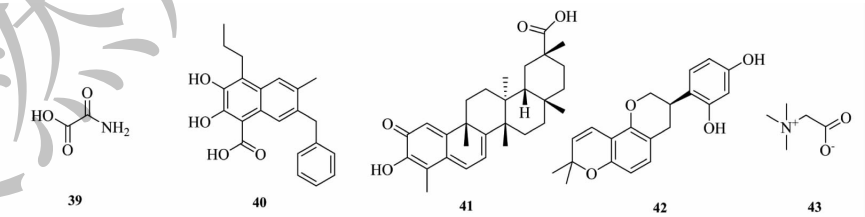

图 6　LDH 抑制剂结构

Fig. 6　Structures of LDH inhibitors

2　小结与展望

有氧糖酵解作为肿瘤细胞特异的能量代谢特征,不仅为肿瘤细胞的增殖提供了能量和物质供应,同时酵解途径导致的乳酸增加还为肿瘤细胞提供了一个酸性成长环境而利于其浸润和转移。本文通过对现有研究发现的天然来源靶向肿瘤糖酵解关键酶的小分子抑制剂进行总结,直观地体现其结构并为结构改造提供新思路,以期为更有效地开发利用天然产物中的抗肿瘤活性成分,对逆转肿瘤的多药耐药,改善常规化疗的治疗效果,提高肿瘤对化疗药物

的敏感性等多个抗肿瘤方面的研究提供灵感。

参考文献

1 Wei HR, et al. Regulation of cancer cell metabolism[J]. Sci Sin Vitae(中国科学), 2017, 47: 32-139.

2 Pavlova NN, et al. The hallmarks of cancer metabolism: still emerging[J]. Cell Metabolism, 2022, 34: 355-377.

3 Pan ZD, et al. Research progress on the effect of glucose and lipid metabolism disorders on malignant cancer[J]. Electron J Metab Nutr Cancer(肿瘤代谢与营养电子杂志), 2021, 8: 582-587.

4 Vaupel P, et al. Revisiting the Warburg effect: historical dogma versus current understanding[J]. J Physiol, 2021, 599: 1745-1757.

5 Yoshida GJ. Metabolic reprogramming: the emerging concept and associated therapeutic strategies[J]. J Exp Clin Cancer Res, 2015, 34: 111.

6 Abdel-Wahab AF, et al. Targeting glucose metabolism to suppress cancer progression: prospective of anti-glycolytic cancer therapy[J]. Pharmacol Res, 2019, 150: 104511.

7 Abbaszadeh Z, et al. Crucial players in glycolysis: cancer progress[J]. Gene, 2020, 726: 144158.

8 Icard P, et al. How the Warburg effect supports aggressiveness and drug resistance of cancer cells? [J]. Drug Resist Updat, 2018, 38: 1-11.

9 Bhattacharya B, et al. The Warburg effect and drug resistance [J]. Br J Pharmacol, 2016, 173: 970-979.

10 Li XX, et al. Research progress of therapeutic targets and targeted medicine of aerobic glycolysis[J]. Electron J Metab Nutr Cancer(肿瘤代谢与营养电子杂志), 2022, 9: 253-258.

11 Davis-Yadley A H, et al. Increased expression of the GLUT-1 gene is associated with worse overall survival in resected pancreatic adenocarcinoma[J]. Pancreas, 2016, 45: 974.

12 Zhang JG, et al. Role of glucose transporter type 1 inhibitor in oxaliplatin resistance in colon cancer cells[J]. (中国现代医生), 2020, 58: 14-17.

13 Wang T, et al. Elevated expression of TrpC5 and GLUT1 is associated with chemoresistance in colorectal cancer[J]. Oncol Rep, 2017, 37: 1059-1065.

14 Meng Y, et al. The progress and development of GLUT1 inhibitors targeting cancer energy metabolism[J]. Future Med Chem, 2019, 11: 2333-2352.

15 Ancey PB, et al. Glucose transporters in cancer-from tumor cells to the tumor microenvironment[J]. FEBS J, 2018, 285: 2926-2943.

16 Tiemin P, et al. Dysregulation of the miR-148a-GLUT1 axis promotes the progression and chemoresistance of human intrahepatic cholangiocarcinoma[J]. Oncogenesis, 2020, 9: 19.

17 Xu YY, et al. Apigenin suppresses GLUT-1 and p-AKT expression to enhance the chemosensitivity to cisplatin of laryngeal carcinoma HepG-2 cells: an *in vitro* study[J]. Int J Clin Exp Pathol, 2014, 7: 3938.

18 Hsieh IS, et al. Osteopontin upregulates the expression of glucose transporters in osteosarcoma cells[J] PLoS One, 2014, 9: e109550.

19 Zhou LQ, et al. Phloretin regulates aerobic glycolysis to activate AMPK- P53 signaling pathway and inhibit colon cancer cell growth[J]. Tradit Chin Drug Res Clin Pharmacol(中药新药与临床药理), 2020, 31: 515-522.

20 Zhan T, et al. Silybin and dehydrosilybin decrease glucose uptake by inhibiting GLUT proteins[J]. J Cell Biochem, 2011, 112: 849-859.

21 Deng J, et al. Apigenin reduces GluT-1expression via down-regulation of HIF-1α under hypoxic conditions in ovarian cancer SKOV3 cells[J]. Med J West China(西部医学), 2016, 28: 611-614.

22 Li S, et al. Dihydroartemisinin represses esophageal cancer glycolysis by down-regulating pyruvate kinase M2[J]. Eur J Pharmacol, 2019, 854: 232-239.

23 Sun H. Study on mechanism of plumbagin inhibiting expression of Glut-1 in rectal cancer cells[J]. J Med Forum(医药论坛杂志), 2021, 42: 51-54.

24 Chang P, et al. Effects of polysaccharides from ginkgo biloba on proliferation of 4T1 breast cancer cells and expression of GLUT family genes[J]. Chin Pharmacol Bull(中国药理学通报), 2018, 34: 1301-1307.

25 Wang H, et al. Mannose inhibits the occurrence and proliferation of colorectal cancer by down-regulationg GLUT1[J]. Food Ferment Ind(食品与发酵工业), 2020, 46: 53-59.

26 Ciscato F, et al. Hexokinase 2 in cancer: a prima donna playing multiple characters[J]. Int J Mol Sci, 2021, 22: 4716.

27 Deng XL, et al. Research progress of the effects of hexohinase on tumor[J]. Chin Mord Doct(中国现代医生), 2018, 56: 164-168.

28 Wilson JE. Isozymes of mammalian hexokinase: structure, subcellular localization and metabolic function[J]. J Exp Bio, 2003, 206: 2049-2057.

29 Pelicano H, et al. Glycolysis inhibition for anticancer treatment[J]. Oncogene, 2006, 25: 4633-4646.

30 Garcia SN, et al. Unlocking the potential of HK2 in cancer metabolism and therapeutics[J]. Curr Med Chem, 2019, 26: 7285-7322.

31 Suh DH, et al. Association of overexpression of hexokinase II

with chemoresistance in epithelial ovarian cancer[J]. Clin Exp Med,2014,14:345-353.

32 Jiang M,et al. A pan-cancer analysis of molecular characteristics and oncogenic role of hexokinase family genes in human tumors[J]. Life Sci,2021,264:118669.

33 Shi ZF,et al. Study on the effects of 3-bromopyruvate on the sensitivity of human nasopharyngeal carcinoma cells to cisplatin and its mechanism[J]. J Bengbu Med Coll(蚌埠医学院学报),2020,45:1147-1153.

34 Fan HL,et al. Apoptosis of human lung adenocarcinoma cells H1975 induced by 3-bromopyruvate[J]. Chin Pharmacol Bull(中国药理学通报),2020,36:1520-1526.

35 Kim W,et al. Apoptosis-inducing antitumor efficacy of hexokinase II inhibitor in hepatocellular carcinoma[J]. Mol Cancer Ther,2007,6:2554-2562.

36 Wang L,et al. The combination of the glycolysis inhibitor 2-DG and sorafenib can be effective against sorafenib-tolerant persister cancer cells[J]. Onco Targets Ther,2019,12:5359-5373.

37 Ren H,et al. Regulation effect of berberine on the inhibition of hexokinase Ⅱ in breast cancer cells glycolysis[J]. China Pharm(中国药师),2017,20:1945-1949.

38 Wu GC,et al. Berberine regulating the level of glycolysis in non-small cell lung cancer cells and inhibiting the occurrence of EMT in A549 cells[J]. J Clin Pulmon Med(临床肺科杂志),2020,25:1206-1211.

39 Bai J. Inhibitory effect and mechanism of ophiopogonin B on human lung adenocarcinoma A549 xenografted in nude mice[D]. Changchun:Jilin University(吉林大学),2021.

40 Wang XT,et al. Prosapogenin A inhibits cell growth of MCF7 via downregulating STAT3 and glycometabolism-related gene[J]. Acta Pharm Sin(药学学报),2013,48:1510-1514.

41 Luo YY,et al. Regulation of saponins from *Pulsatilla chinensis* on energy metabolism of BEL-7402 xenograft in nude mice[J]. Chin Tradit Herb Drugs(中草药),2014,45:973-977.

42 Li J,et al. 20(*S*)-Ginsenoside Rg_3 inhibits the Warburg effect through STAT3 pathways in ovarian cancer cells[J]. Int J Oncol,2015,46:775-781.

43 Wu SB,et al. Protective effects of ginsenoside 20(*S*)-Rg_3 on renal toxicity induced by cisplatin in LLC-PK1 cells[J]. World Chin Med(世界中医药),2018,13:2010-2014.

44 Chen Y,et al. Ginsenoside Rh_2 improves the cisplatin antitumor effect in lung adenocarcinoma A549 cells via superoxide and PD-L1[J]. Anticancer Agents Med Chem,2020,20:495-503.

45 Liu XM,et al. Resveratrol promotes cisplatin-induced cell death by inhibiting the glucose metabolism in bladder cancer[J]. Chin J Mod Appl Pharm(中国现代应用药学),2016,33:1262-1267.

46 Yu M,et al. Research stotus of the anti-tumor mechanism of resveratrol[J]. J Jilin Inst Chem Technol(吉林化工学院学报),2019,36:12-15.

47 Xiong J,et al. Resveratrol inhubits glycolusis of oral squamous cell regulating mTOR/PKM2 Axis[J]. J Oral Sci Res(口腔医学研究),2019,35:573-577.

48 Li W,et al. Resveratrol inhibits hexokinases II mediated glycolysis in non-small cell lung cancer via targeting Akt signaling pathway[J]. Exp Cell Res,2016,349:320-327.

49 Dai W,et al. By reducing hexokinase 2,resveratrol induces apoptosis in HCC cells addicted to aerobic glycolysis and inhibits tumor growth in mice[J]. Oncotarget,2015,6:13703-13717.

50 Vaughan RA,et al. Tumor necrosis factor alpha induces Warburg-like metabolism and is reversed by anti-inflammatory curcumin in breast epithelial cells[J]. Int J Cancer,2013,133:2504-2510.

51 Wang K,et al. Curcumin inhibits aerobic glycolysis and induces mitochondrial-mediated apoptosis through hexokinase II in human colorectal cancer cells in vitro[J]. Anticancer Drugs,2015,26:15-24.

52 Wang H,et al. Wogonin reverses hypoxia resistance of human colon cancer HCT116 cells via downregulation of HIF-1α and glycolysis,by inhibiting PI3K/Akt signaling pathway[J]. Mol Carcinog,2014,53(S1):E107-E118.

53 Wei L,et al. Oroxylin A inhibits glycolysis-dependent proliferation of human breast cancer via promoting SIRT3-mediated SOD2 transcription and HIF1α destabilization[J]. Cell Death Dis,2015,6:e1714-e1714.

54 Zhang D,et al. Effects of silencing HK2 gene through siRNA on radionsensitivity of MDA-MB231 cell line[J]. Acta Univ Med Anhui(安徽医科大学学报),2017,52:95-99.

55 He JQ,et al. Research progress of pyruvate dehydrognenase kinases in maliganancies[J]. Electron J Pract Gynecol Endocrinol(实用妇科内分泌杂志电子版),2018,5:9.

56 Wang QY,et al. Research progress of pyruvate dehydrognenase kinases in maliganancies[J]. J Cancer Control Treat(肿瘤预防与治疗),2019,32:945-950.

57 Li B,et al. Reversal of the Warburg effect with DCA in PDGF-treated human PASMC is potentiated by pyruvate dehydrogenase kinase-1 inhibition mediated through blocking Akt/GSK-3β signalling[J]. Int J Mol Med,2018,2:1391-1400.

58 Emmanouilidi A,et al. Targeting PDK1 for chemosensitization

of cancer cells[J]. Cancers,2017,9:140.
59 Peng F,et al. Glycolysis gatekeeper PDK1 reprograms breast cancer stem cells under hypoxia[J]. Oncogene,2018,37:1062-1074.
60 Xiao D. Role of pyruvate dehydrogenase kinase 4 in chemotherapy resistance of bladder cancer[J]. Lab Med Clin(检验医学与临床),2020,17:344-346.
61 Xu XW,et al. Mechanism of HIF-1α-PDK1 signaling system involved in glucose metabolism and drug resistance in acute monocytic leukemia[J]. J Shanghai Jiaotong Univ Med Sci(上海交通大学学报医学版),2018,38:1294-1299.
62 Tataranni T,et al. Dichloroacetate(DCA) and cancer:an overview towards clinical applications[J]. Oxid Med Cell Longev,2019,2019:8201079.
63 Sun W,et al. JX06 selectively inhibits pyruvate dehydrogenase kinase PDK1 by a covalent cysteine modification[J]. Cancer Res,2015,75:4923-4936.
64 Lee EJ,et al. Water-extracted branch of *Cinnamomum cassia* promotes lung cancer cell apoptosis by inhibiting pyruvate dehydrogenase kinase activity[J]. J Pharmacol Sci,2018,138:146-154.
65 Tangchirakhaphan S,et al. Mechanism of apoptosis induction associated with ERK1/2 upregulation via goniothalamin in melanoma cells[J]. Ther Med,2018,15:3052-3058.
66 Chung TW,et al. *Anemone rivularis* inhibits pyruvate dehydrogenase kinase activity and tumor growth[J]. J Ethnopharmacol,2017,203:47-54.
67 Kwak CH,et al. Huzhangoside a suppresses tumor growth through inhibition of pyruvate dehydrogenase kinase activity[J]. Cancers,2019,11:712.
68 Song L,et al. Study of the effect of astragalin on prolifreation of ovarian cancer inhubiting the glycolytic pathway induced via HIF-1α[J]. Pract Oncol J(实用肿瘤学杂志),2018,32:503-509.
69 Christofk HR,et al. The M2 splice isoform of pyruvate kinase is important for cancer metabolism and tumour growth[J]. Nature,2008,452:230-233.
70 Wang JZ,et al. The role of the HIF-1α/ALYREF/PKM2 axis in glycolysis and tumorigenesis of bladder cancer[J]. Cancer Commun,2021,41:560-575.
71 Li Y,et al. PKM2,a potential target for regulating cancer[J]. Gene,2018,668:48-53.
72 Zhu S,et al. Pyruvate kinase M2(PKM2) in cancer and cancer therapeutics[J]. Cancer Lett,2021,503:240-248.
73 Zahra K,et al. Pyruvate kinase M2 and cancer:the role of PKM2 in promoting tumorigenesis[J]. Front Oncol,2020,10:159.
74 Li SM,et al. Research progress on anticancer effects of pyruvate kinase M2[J]. China Cancer(中国肿瘤),2019,28(3):202-207.
75 Li TE,et al. PKM2 drives hepatocellular carcinoma progression by inducing immunosuppressive microenvironment[J]. Front Immunol,2020,11:589997.
76 Liu K,et al. The role of PKM1 in tumors[J]. Prog Physiol Sci(生理科学进展),2020,51:437-442.
77 Su Q,et al. The role of pyruvate kinase M2 in anticancer therapeutic treatments[J]. Oncol Lett,2019,18:5663-5672.
78 Zhao J.[D]. Shikonin enhances the sensitivity of non-small cell lung cancer to gefitinib and its mechanism[D]. Nanchong:North Sichuan Medical College(川北医学院),2018.
79 Wang Z,et al. Combination of shikonin with paclitaxel overcomes multidrug resistance in human ovarian carcinoma cells in a P-gp-independent manner through enhanced ROS generation[J]. Chin Med,2019,14:7.
80 Tang JC,et al. Efficacy of shikonin against esophageal cancer cells and its possible mechanisms *in vitro* and *in vivo*[J]. J Cancer,2018,9:32.
81 Tao T,et al. Down-regulation of PKM2 decreases FASN expression in bladder cancer cells through AKT/mTOR/SREBP-1c axis[J]. J Cell Physiol,2019,234:3088-3104.
82 Chen J,et al. Vitamin K3 and K5 are inhibitors of tumor pyruvate kinase M2[J]. Cancer letters,2012,316:204-210.
83 Wei X,et al. Induction of apoptosis by FFJ-5,a novel naphthoquinone compound,occurs via downregulation of PKM2 in A549 and HepG2 cells[J]. Oncol Lett,2017,13:791-799.
84 Zhao JL. Effect of andrographis effective parts on MCF-7/ADM and its mechanisms[D]. Kaifeng:Henan Univ(河南大学),2016.
85 Zhu ZW,et al. Effects of serum containing Bailu decoction on the activity,apoptosis and cell cycle of human lung cancer A549 cells[J]. Inf Tradit Chin Med(中医药信息),2019,36(3):1-4.
86 Sun W,et al. Scutellarin resensitizes oxaliplatin-resistant colorectal cancer cells to oxaliplatin treatment through inhibition of PKM2[J]. Mol Ther Oncolytics,2021,21:87-97.
87 Zhang HS,et al. Tanshinone ⅡA inhibits human esophageal cancer cell growth through miR-122-mediated PKM2 downregulation[J]. Arch Biochem Biophys,2016,598:50-56.
88 Zhou J,et al. Cryptanshinone inhibits the glycolysis and inhibits cell migration through PKM2/β-catenin axis in breast cancer[J]. Onco Targets Ther,2020,13:8629.
89 Zhao XY,et al. Effect of 6-gingerol on glycolysis in human gastric cancer BGC-823 cells[J]. Acta Nutr Sin(营养学报),2019,41:411-413.

90 Yang YT, et al. Patchouli alcohol induces apoptosis of MV4-11 leukemia cells by PKM2 and NF-κB[J]. Chin J Exp Tradit Med Form(中国实验方剂学杂志),2016,22:99-103.

91 Liu J, et al. Oleanolic acid suppresses aerobic glycolysis in cancer cells by switching pyruvate kinase type M isoforms [J]. PLoS One,2014,9:e91606.

92 Shi T, et al.. Bufalin inhibits the proliferation, migaration and invasion of melanoma cells by down-regulating PKM2 [J]. Pharm Clin Res(药学与临床研究),2019,27:327-331.

93 Zhou Y, et al. Ampelopsin induces cell growth inhibition in breast cancer cells by suppressing pyruvate kinase M2-dependent glycolysis[J]. Electron J Metabol Nutr Cancer(肿瘤代谢与营养电子杂志),2019,6:236-241.

94 Li Z, et al. Research progress of matrine in treatment of liver cancer[J], Chin Arch Tradit Chin Med(中华中医药学刊),2022,40(4):99-103.

95 Han RL, et al. Effects of Emodin on the proliferation and apoptosis of gastric cancer cells and ERK1/2-PKM2/P53 pathway[J]. China Pharm(中国药师),2019,22:2208-2213.

96 Li YQ, et al. Effects of Emodin-8-*O*-*β*-*D*-glucopyranoside on tumor cell migration and mestastasis *in vitro* and *in vivo* [J]. Chin J Pharmacov(中国药物警戒),2019,16:705.

97 Feng J, et al. PKM2 is the target of proanthocyanidin B2 during the inhibition of hepatocellular carcinoma[J]. J Exp Clin Cancer Res,2019,38:204.

98 Zhou WL, et al. Research progress on lactatedehydrogenase and immunometabolism in cancer[J]. Electron J Metab Nutr Cancer(肿瘤代谢与营养电子杂志),2020,7:396-401.

99 Pathria G, et al. Targeting the Warburg effect via LDHA inhibition engages ATF 4 signaling for cancer cell survival [J]. EMBOJ,2018,37:e99735.

100 Li X, et al. The research progress of lactate dehydrogenase A(LDH-A) in malignant tumor[J]. J Mod Oncol(现代肿瘤医学),2020,28:2532-2535.

101 Feng Y, et al. Lactate dehydrogenase A:a key player in carcinogenesis and potential target in cancer therapy [J]. Cancer Med,2018,7:6124-6136.

102 Fantin VR, et al. Attenuation of LDH-A expression uncovers a link between glycolysis, mitochondrial physiology, and tumor maintenance[J]. Cancer Cell,2006,9:425-434.

103 Le A, et al. Inhibition of lactate dehydrogenase A induces oxidative stress and inhibits tumor progression [J]. Proc Natl Acad Sci USA,2010,107:2037-2042.

104 Zhou M, et al. Warburg effect in chemosensitivity:targeting lactate dehydrogenase-A re-sensitizes taxol-resistant cancer cells to taxol[J]. Mol Cancer,2010,9:33.

105 Xing BC, et al. Synergistically suppressive effects on colorectal cancer cells by combination of mTOR inhibitor and glycolysis inhibitor, Oxamate [J]. Int J Clin Exp Pathol, 2018,11:4439.

106 Solyanik GI, et al. Oxamate, an inhibitor of lactate dehydrogenase, can stimulate M2 polarization of peritoneal macrophages in mice with Lewis lung carcinoma[J]. Exp Oncol. 2021,43:270-273.

107 Mohammad GH, et al. Targeting pyruvate kinase M2 and lactate dehydrogenase a is an effective combination strategy for the treatment of pancreatic cancer[J]. Cancers, 2019, 11:1372.

108 Wang SJ, et al. Effect of celastrol on proliferation and energy metabolism of human lung cancer A549 cell line [J]. J Pharm Res(药学研究),2020,39:266-268.

109 Li LJ, et al. Regulatory effects of glabridin and quercetin on energy metabolism of breast cancer cells[J]. China J Chin Mater Med(中国中药杂志),2019,44:3786-3791.

110 Zhang JY, et al. Antitumor effect of betaine and its influence on glycometabolism of S_{180} bearing mice [J]. Chin Tradit Herb Drugs(中草药),2006,37:1378-1380.