中药有效成分防治骨肉瘤转移作用与机制研究进展*

赵福来[1,2]，常君丽[1,2]，支文兰[1,2]，王晓波[1,2]，孙星媛[1,2]，马小平[1,2]，施 杞[1,2]，王拥军[1,2**]，杨燕萍[1,2**]

（1. 上海中医药大学附属龙华医院 上海 200032；2. 教育部筋骨理论与治法重点实验室 上海 200032）

摘 要：骨肉瘤是一种原发性、侵袭性恶性肿瘤，极易发生早期转移且缺乏有效治疗靶点。手术和放化疗及二者的联合应用是目前主要的治疗手段，但对转移性骨肉瘤患者总体生存率的提高仍未有明显改善。骨肉瘤在中医属于“癌病”范畴，中医药治疗骨肉瘤历史悠久，并且在防治骨肉瘤转移方面能够发挥积极作用，但其机制尚未十分明确。信号通路在骨肉瘤转移的发生过程中具有重要作用，并且是研究骨肉瘤转移的病理机制和治疗药物的作用靶点之一。近年来，针对中药有效成分防治骨肉瘤转移相关信号通路进行了大量研究，现将此做一综述，以期为中医药防治骨肉瘤转移、新药开发和临床诊治提供借鉴。

关键词：中药有效成分 骨肉瘤 转移 作用机制 研究进展

doi: 10.11842/wst.20210328001 中图分类号：R285 文献标识码：A

骨肉瘤是一种原发性、恶性骨肿瘤，好发于儿童和青少年[1-2]，具有较高的局部侵袭和远处转移率，易发生早期转移，最常发生转移的部位在肺部，预后较差。据报道未发生转移的骨肉瘤患者5年生存率在70-80%之间[3]，而转移性患者5年生存率仅10-30%[4]。目前临床上常规治疗主要是手术和放化疗以及两者的联合应用，而转移性患者对常规治疗手段更容易产生抗性。针对骨肉瘤转移的治疗可有效延长患者生存时间，因此急需开发防治骨肉瘤转移的新药物。

骨肉瘤属于中医“癌病”范畴，病因病机为正虚热毒蕴结，中药治疗骨肉瘤能够起到清热解毒，驱邪扶正之功效。探究骨肉瘤转移的分子机制能够帮助寻找中药防治骨肉瘤转移的治疗靶点。研究表明，多种信号通路参与到骨肉瘤的转移进程[5-6]。近年来关于中药及其有效单体防治肿瘤的报道越来越多，并且有研究表明中药在抗骨肉瘤转移方面表现出不同于西药的独特效果[7-8]，且中药有效成分能够通过多种信号途径发挥作用[9]。现将中药有效成分通过相关信号通路防治骨肉瘤转移的研究进展综述如下（表1），以便为后期骨肉瘤转移的分子机制研究、靶点药物开发以及临床治疗提供借鉴和参考。

1 PI3K/Akt信号通路

磷脂酰肌醇3-激酶（Phosphoinositide 3 kinase, PI3K）/蛋白激酶B（Protein kinase B, Akt）通路是调节肿瘤细胞分化、迁移和浸润的重要细胞内信号通路之一[10]。PI3Ks构成了一个脂质激酶家族，可分为3类，其中Ⅰ型PI3K和肿瘤的关系最为密切[11]。原发骨肉

收稿日期：2021-03-28

修回日期：2022-01-14

* 国家科学技术部国家重点研发计划-中医药现代化研究重点专项（2018YFC1704300）：“肾阳虚证”辨识方法和临床指导原则的应用研究，负责人：王拥军；国家自然科学基金委员会面上项目（81973877）：LncRNA-CCDC18-AS1激活YAP/TAZ诱导骨肉瘤发生及加味芪重散作用的机制研究，负责人：杨燕萍；国家科学技术部战略性国际科技创新合作重点专项（2020YFE0201600）：人类表型组测量技术及表型数据跨尺度关联合作研究，负责人：杨燕萍。

** 通讯作者：杨燕萍，研究员，博士生导师，主要研究方向：中医药防治骨肉瘤的基础研究；王拥军，本刊编委，教授，博士生导师，主要研究方向：中医药防治慢性筋骨疾病的基础与临床研究。

瘤病例中PI3K/Akt信号通路的激活与患者的不良预后有明显的相关性[12]，且在中国人群中携带Akt基因型AA的骨肉瘤患者具有更高的转移风险[13]。上皮间充质转化（Epithelial to mesenchymal transition，EMT）与骨肉瘤的侵袭和远处转移高度相关[14-15]，通过EMT过程上皮细胞获得更高的迁移和浸润能力，以及更强的降解细胞外基质的间质细胞表型。基质金属蛋白酶（Matrix metallo proteinases，MMPs）的上调能够降解细胞外基质和基底膜，从而参与并促进EMT进程[16-17]。有研究表明侵袭相关蛋白MMP-2和MMP-9受到Akt的调控[18]。Yang[19]等的研究发现，卷柏在U-2 OS细胞中表现出与PI3K抑制剂相似的抗骨肉瘤转移的效果，在0-50 μg·mL^{-1}范围内呈剂量依赖性通过抑制Akt的磷酸化减少MMP-2和MMP-9的产生，同时增加基质金属蛋白酶抑制因子（Tissue inhibitor of metalloproteinase，TIMP）-1/-2的表达，可抑制U-2 OS细胞的迁移和浸润。Chen[20]等发现苦豆碱能够下调Akt的磷酸化水平和PI3K的表达抑制PI3K/Akt信号通路的活性，下调MMP-2和MMP-9，抑制骨肉瘤细胞的迁移和侵袭。骨肉瘤肿瘤干细胞具有更高的成瘤率和更强的转移能力，且在骨肉瘤的发生和转移中发挥重要作用[21]。有研究表明PI3K/Akt信号通路参与骨肉瘤细胞的干性调节[22-23]。Ma[24]等研究发现和蟾蜍他灵能够降低U-2 OS和MG63细胞中MMP-2、干细胞标记物CD133等的表达，并在体内实验证实和蟾蜍他灵能够有效降低骨肉瘤肺转移的数量，而这些都是通过对PI3K/Akt信号通路的调节作用实现的。此外还有雷公藤红素[25]、异甘草素[26]、羽扇豆醇[27]等多种中药有效成分可通过PI3K/Akt信号通路降低MMP-2、MMP-9的表达水平抑制骨肉瘤转移。

2 Wnt/β-Catenin信号通路

Wnt/β-catenin信号通路在细胞的增殖和迁移中发挥重要作用，其核心蛋白β-catenin受磷酸化调节，在Wnt信号缺乏时糖原合成酶激酶-3β（Glycogen synthase kinase-3β，GSK-3β）促进β-catenin磷酸化，从而阻止β-catenin入核，并使其在胞浆中通过泛素/蛋白酶体通路降解[28]。当Wnt信号被激活后β-catenin的磷酸化受到抑制，未磷酸化的β-catenin进入核内激活其下游靶基因。Wnt靶基因的异常激活参与到癌症的发展进程。在骨肉瘤临床样本中发现，患者瘤体组织中β-catenin表达量增高，且与不良预后和肺转移的发生高度相关[29-30]，激活Wnt/β-catenin信号通路能够促进骨肉瘤细胞的侵袭和转移[31-32]。Nomura等也发现与原位骨肉瘤细胞相比，Wnt/β-catenin信号在远处转移瘤细胞中的活性显著增强[33]。Wnt/β-catenin信号通路激活与EMT相关，骨形成蛋白-2（Bone morphogenetic protein-2，BMP-2）[34]和细胞外基质蛋白-3（Fibulin-3）[35]都可通过激活Wnt/β-catenin信号通路诱导EMT进程，促进骨肉瘤细胞的迁移和浸润。Fang等发现Wnt/β-catenin信号通路抑制剂PRI-724能够有效抑制骨肉瘤细胞的增殖、迁移和浸润[36]。天然药物的有效成分也能够有效抑制Wnt/β-catenin信号通路活性，且无明显副作用[37]。重楼皂苷Ⅰ是中药重楼的主要有效成分之一，Chang[38]等在体内外研究中发现重楼皂苷Ⅰ能降低骨肉瘤细胞中GSK-3β的磷酸化水平，导致活性β-catenin和Vimentin水平下调；使用GSK-3β特异性抑制剂后，重楼皂苷Ⅰ对骨肉瘤细胞活性和转移的抑制作用被消除，而β-catenin沉默增强了重楼皂苷Ⅰ对骨肉瘤细胞活性和迁移的抑制作用。Dai[39]等发现黄芩素能够通过下调Wnt/β-catenin信号通路的活性降低MMP-2和MMP-9的表达，抑制143B和MG63细胞的迁移和侵袭；当上调Wnt/β-catenin信号通路时，黄芩素的这种作用被拮抗掉。Yang[40]等研究发现吴茱萸碱能够促进GSK-3β的表达抑制β-catenin的活性，下调Snail、Vimentin、MMP-2、MMP-7、MMP-9、N-Cadherin等的表达并上调E-Cadherin，抑制骨肉瘤细胞的迁移和浸润。二氢丹参酮Ⅰ是中药丹参的有效成分之一，Tan[41]等研究发现，经二氢丹参酮Ⅰ处理后的骨肉瘤细胞中Snail、MMP-2、MMP-7、MMP-9、N-Cadherin的表达量明显降低，β-catenin和低密度脂蛋白受体相关蛋白6（Low density lipoprotein receptor-related protein 6，LRP6）等的活性被抑制，表明二氢丹参酮Ⅰ能够通过Wnt/β-catenin信号通路抑制骨肉瘤细胞的EMT进程实现抑制骨肉瘤细胞的迁移和侵袭。

3 NF-κB信号通路

核因子κB（Nuclear factor kappa-B，NF-κB）广泛参与肿瘤细胞（包括骨肉瘤）的迁移、浸润、增殖、凋亡、周期、化疗耐药等过程[42-43]。NF-κB抑制蛋白（Inhibitor of NF-κB，IκB）的磷酸化促进NF-κB的释

放入核，并促进Twist、Vimentin等EMT相关靶基因的激活，导致白细胞介素8（Interleukin-8，IL-8）和血管内皮生长因子（Vascular endothelial growth factor，VEGF）的释放，促进MMPs的表达[44-45]。NF-κB的诱杀性寡脱氧核苷酸（Decoy oligodeoxynucleotide，Decoy DON）转染到具有高肺转移能力的小鼠骨肉瘤细胞核中，发现细胞中VEGF和细胞间粘附分子1（Intercellular adhesion molecule 1，ICAM-1）mRNA表达明显降低，在体内实验中NF-κB Decoy DON能够显著降低肺转移的数量，说明NF-κB在骨肉瘤转移的调控中具有独特作用[46]，因此NF-κB可作为抑制骨肉瘤转移的潜在作用靶点。小白菊内酯是一种天然的NF-κB抑制剂，Kishida等研究发现小白菊内酯能够使NF-κB失活并下调VEGF的表达[47]。中药苦参的有效成分苦参碱，长期被用于炎症和癌症的治疗中。Li[48]等发现苦参碱能够明显抑制NF-κ B/p50、p65/RelA的表达和IκB-β的磷酸化，并能够降低MMP-2和MMP-9的表达，发挥抑制骨肉瘤细胞侵袭能力的作用。Zhang[49]等研究发现莱菔硫烯能够明显促进E-Cadherin的表达，并能抑制Vimentin、Snail、Twist的表达，说明莱菔硫烯能够抑制骨肉瘤细胞的EMT进程，进一步探究发现莱菔硫烯能够呈剂量依赖性的降低跨膜糖蛋白滤泡抑制素样蛋白1（Transmembrane glycoprotein folliclestatin like protein 1，FSTL1）和NF-κB的磷酸化水平，说明莱菔硫烯是通过下调FSTL1/NF-κB通路抑制U-2 OS和Saos-2细胞的迁移和浸润能力。此外，Chang[50]等的研究发现重楼皂苷Ⅰ也能够通过下调NF-κB信号通路的活性，逆转EMT进程发挥抑制骨肉瘤细胞转移的作用。

4 MAPKs信号通路

丝裂原活化蛋白激酶（Mitogen-activated protein kinases, MAPKs）是细胞在生长、生存和生理、病理过程的重要调控因子，MPAKs的异常活化在肿瘤的恶性化进程中发挥了重要作用[51]。在高等真核生物中MAPKs信号通路主要有5个亚家族成员分别是：细胞外信号调节激酶1/2（Extracellular signal regulated kinases 1/2, ERK1/2）、p38丝裂原活化蛋白激酶（p38 mitogen-activated protein kinase, p38 MAPK）、c-Jun氨基末端激酶（c-Jun N-terminal kinase, JNK）、细胞外信号调节激酶5（Extracellular signal regulated kinases 5, ERK5）和非典型性MAPKs（Atypical MAPKs）[52]。MAPKs信号通路的家族成员是启动EMT的关键因子，其失活可以抑制肿瘤细胞的迁移和浸润[53]；同时ERK1/2、JNK和p38 MAPK等MAPKs成员在调控MMPs的表达中也起着核心作用[54-56]。巴黎皂苷Ⅶ是中药延龄草的有效成分之一，巴黎皂苷Ⅶ能够通过抑制p38 MAPK的活性，从而导致MMP-2和MMP-9的下调抑制骨肉瘤细胞的迁移和浸润[57]。中药飞燕草的有效成分飞燕草素能降低ERK1/2和p38 MAPK的磷酸化水平，抑制N-Cadherin的表达，促进E-Cadherin的表达，从而抑制骨肉瘤细胞的迁移能力，且飞燕草素的这些作用在与ERK1/2和p38的抑制剂联用后被增强[58]。Cheng[59]等发现中药陈皮的有效成分川陈皮素能够通过抑制NF-κB和环磷腺苷效应元件结合蛋白（cAMP-response element binding protein，CREB）在MMP-2和MMP-9启动子上的表达和结合活性来抑制骨肉瘤细胞转移，且这种抑制作用主要是通过下调ERK和JNK的磷酸化水平，降低MMP-2和MMP-9的表达逆转EMT进程实现抑制骨肉瘤转移的作用。三氧化二砷（As_2O_3）是目前研究最广泛的抗癌药。Ren[60]等研究发现As_2O_3能够降低ERK1/2和MAPK激酶（MAP kinase，MEK）的磷酸化水平，抑制MMP-9的表达，进而抑制骨肉瘤细胞的迁移和浸润能力。光甘草定[61]，山奈酚[62]也能够通过抑制MAPKs信号通路下调MMPs的表达抑制骨肉瘤细胞转移。

5 其他信号通路

Notch同源物1（Notch homolog 1，Notch-1）和信号传导及转录激活蛋白3（signal transducer and activator of transcription 3，STAT3）信号通路的异常激活能够促进多种细胞进程，包括肿瘤细胞的增殖，迁移和浸润[63-64]。Li等研究发现姜黄素和大蒜素分别能够通过抑制Notch-1信号通路下调VEGF、MMP-2和MMP-9的表达抑制骨肉瘤细胞的侵袭[65-66]。Jin[67]等研究发现木香烃内酯能够抑制STAT3信号通路活性下调MMP-2的表达抑制骨肉瘤转移，体内实验表明木香烃内酯明显减少了肿瘤肺转移的数量；趋化因子受体4（Chemokine receptor 4，CXCR4）能够激活STAT3信号通路参与多种肿瘤细胞过程，XIE [68]等的研究发现汉防己碱能够通过抑制MMP-2和MMP-9的分泌和

表1　中药有效成分对骨肉瘤转移相关信号通路的调控作用

中药有效成分	作用细胞	作用机制	相关信号通路	参考文献
卷柏	U-2 OS	MMP-2↓，MMP-9↓，TIMP-1↑，TIMP-2↑，p38↓，p-Akt↓	PI3K/Akt，p38	19
苦豆碱	U-2 OS MG63	MMP-2↓，MMP-9↓，PI3K↓，p-AKT1↓	PI3K/Akt	20
和蟾蜍他灵	U-2 OS MG63	E-Cad↑，MMP-2↓，p-PI3K↓，p-Akt↓	PI3K/Akt	24
雷公藤红素	U-2 OS	MMP-2↓，MMP-9↓，p-PI3K↓，p-Akt↓，p-IKKα/β↓，p-IκBα↓，p-p65↓	PI3K/Akt，NF-κB	25
异甘草素	U-2 OS	p-Akt↓，p-PI3K↓，MMP-2↓，MMP-9↓	PI3K/Akt	26
羽扇豆醇	U-2 OS	MMP-2↓，MMP-9↓，p-p38↓，β-catenin↓	PI3K/Akt/GSK-3β，p-38/MAPK	27
重楼皂苷Ⅰ	143B HOS	p- GSK-3β↓，Vimentin↓，Active β-catenin↓	Wnt/β-catenin	38
黄芩素	143B MG63	MMP-2↓，MMP-9↓，β-catenin↓	Wnt/β-catenin	39
吴茱萸碱	143B MG63	MMP-2↓，MMP-7↓，MMP-9↓，Snail↓，N-Cadherin↓，GSK-3β↑，β-catenin↓，Vimentin↓，E-Cadherin↑，	Wnt/β-catenin	40
二氢丹参酮Ⅰ	143B MG63 Saos-2 U-2 OS	MMP-2↓，MMP-7↓，MMP-9↓，LRP6↓，N-Cadherin↓，Snail↓，β-catenin↓	Wnt/β-catenin	41
苦参碱	Saos-2 U-2 OS MG63	NF-κB/p50↓，p65/RelA↓，p-IκB-β↓，ERK↑，MMP-2↓，MMP-9↓，P-IκB-β↓，p-ERK↓，	ERK/NF-κB	48
莱菔硫烯	Saos-2 U-2 OS	E-Cadherin↑，FSTL1↓，Vimentin↓，Snail↓，Twist↓，p-NF-κB↓	FSTL1/NF-κB	49
重楼皂苷Ⅰ	MG-63 Saos-2 U-2 OS	p-Erk1/2↓，p-NF-κBp65↓，E-Cadherin↑，Vimentin↓	NF-κB	50
巴黎皂苷Ⅶ	U-2 OS	MMP-2↓，MMP-9↓，p-38↓	p-38 MAPK	57
飞燕草素	U-2 OS HOS MG63	E-Cadherin↑，Slug↓，N-Cadherin↓，Snail↓，p-p38↓，p-ERK1/2↓	ERK/p38 MAPK	58
川陈皮素	U-2 OS HOS	p-ERK↓，p-JNK↓，MMP-2↓，MMP-9↓	MAPK	59
三氧化二砷	HOS MNNG	p-ERK↓，p-MEK↓，MMP-9↓	MAPK	60
光甘草定	MG63 HOS	p-p38↓，p-ERK↓，c-Jun↓，p-JNK↓，MMP-2↓，MMP-9↓	MAPK	61
山柰酚	U-2 OS	p-p38↓，p-ERK↓，p-JNK↓，MMP-2↓，MMP-9↓	MAPK	62
姜黄素	U-2 OS Saos-2 MG63	Notch-1↓，Hes-1↓，MMP-2↓，MMP-9↓，Cyclin D1↓	Notch	65
大蒜素	U-2 OS Saos-2 MG63	Notch-1↓，VEGF↓，Hes-1↓，Cyclin D1↓，MMP-2↓，MMP-9↓	Notch	66
木香烃内酯	143B HOS MG63	PCNA↓，p-STAT3↓，MMP-2↓	STAT3	67
汉防己碱	U-2 OS HOS	p-STAT3↓，CRCX4↑，MMP-2↓，MMP-9↓，TIMP-1↑，TIMP-2↑	CRCX4-STAT3	68

注：↑升高，↓降低。

CXCR4-STAT3轴的激活来抑制骨肉瘤的侵袭和迁移。

6 小结与展望

综上所述，多条信号通路参与骨肉瘤细胞的EMT进程和细胞外基质降解是骨肉瘤发生转移的根本原因，信号通路的激活或失活均可导致骨肉瘤转移的发生，因此针对信号通路的治疗有望成为防治骨肉瘤转移的有效靶点。中药有效成分能够抑制骨肉瘤转移相关的PI3K/Akt、Wnt/β-Catenin、NF-κB、MAPKs、Notch-1、STAT3等多种信号通路活性，主要是下调通路中关键信号分子的磷酸化水平，抑制骨肉瘤细胞的EMT进程及细胞外基质的降解，发挥抑制骨肉瘤细胞转移的作用。中药有效成分对于防治骨肉瘤的转移具有重要意义，能够以其独特的作用方式和作用特点调控骨肉瘤发展进程，发挥抑制骨肉瘤转移的作用。此外，中药属于天然药物经过特殊的炮制后能够起到不同于西药的独特疗效且具有副作用低的特点。随着科技的发展和对中医药理论及实践研究的逐渐深入，中医药的作用方式和作用途径被进一步发展扩大。

但现有的研究仅对骨肉瘤转移过程中的单一相关信号通路、部分细胞因子或相关基因进行研究；在使用中药过程中缺乏对该疾病证型的分析和中医整体观以及个性化治疗的把握，关于信号通路的研究也较为单一，未涉及到信号通路间相互作用的研究。通过梳理本文发现，重楼皂苷Ⅰ能够同时通过调控Wnt/β-catenin和NF-κB信号通路逆转骨肉瘤细胞的EMT进程实现抑制骨肉瘤细胞侵袭和迁移的作用[38,50]。在今后的研究中应该着重关注中药同时调控多条信号通路发挥协同作用抑制骨肉瘤转移的作用，以发掘中药通过多靶点、多途径发挥作用的优势和特色。

针对中药及有效成分防治骨肉瘤转移的研究要深入结合现代研究手段及多学科交叉的研究思路，持续深化中药药理作用研究、并开展辨证论治用药治疗骨肉瘤，推进中医药防治骨肉瘤转移的机制研究，以便在未来应用中医药防治骨肉瘤转移方面发挥更大的作用和潜力。

参考文献

1 Rothzerg E, Ingley E, Mullin B, *et al*. The Hippo in the room: Targeting the Hippo signalling pathway for osteosarcoma therapies. *J Cell Physiol*, 2021, 236(3):1606-1615.

2 Liu Y, Qiao Z, Gao J, *et al*. Hydroxyapatite-bovine serum albumin-paclitaxel nanoparticles for locoregional treatment of osteosarcoma. *Adv Healthc Mater*, 2021, 10(2):e2000573.

3 Anderson M E. Update on survival in osteosarcoma. *Orthop Clin North Am*, 2016, 47(1):283-292.

4 Farfalli G L, Albergo J I, Lobos P A, *et al*. Osteosarcoma lung metastases. *Survival after chemotherapy and surgery. Medicina (B Aires)*, 2015, 75(2):87-90.

5 Moriarity B S, Otto G M, Rahrmann E P, *et al*. A Sleeping Beauty forward genetic screen identifies new genes and pathways driving osteosarcoma development and metastasis. *Nat Genet*, 2015, 47(6): 615-624.

6 Yang C, Tian Y, Zhao F, *et al*. Bone microenvironment and osteosarcoma metastasis. *Int J Mol Sci*, 2020, 21(19):6985.

7 Parekh H S, Liu G, Wei M Q. A new dawn for the use of traditional Chinese medicine in cancer therapy. *Mol Cancer*, 2009, 8(1):21.

8 Wu B, Cui J, Zhang C, *et al*. A polysaccharide from Agaricus blazei inhibits proliferation and promotes apoptosis of osteosarcoma cells. *Int J Biol Macromol*, 2012, 50(4):1116-1120.

9 Zhang L, Wei W. Anti-inflammatory and immunoregulatory effects of paeoniflorin and total glucosides of paeony. *Pharmacol Ther*, 2020, 207:107452.

10 Tang R, Zhang Z, Han W. CircLRRK1 targets miR-223-3p to inhibit the proliferation, migration and invasion of trophoblast cells by regulating the PI3K/AKT signaling pathway. *Placenta*, 2021, 104: 110-118.

11 Zhang J, Yu X H, Yan Y G, *et al*. PI3K/Akt signaling in osteosarcoma. *Clin Chim Acta*, 2015, 444:182-192.

12 Zhao G, Cai C, Yang T, *et al*. MicroRNA-221 induces cell survival and cisplatin resistance through PI3K/Akt pathway in human osteosarcoma. *PLoS One*, 2013, 8(1):e53906.

13 He M L, Wu Y, Zhao J M, *et al*. PIK3CA and AKT gene polymorphisms in susceptibility to osteosarcoma in a Chinese population. *Asian Pac J Cancer Prev*, 2013, 14(9):5117-5122.

14 Zhang X, Zhang Y, Mao Y, *et al*. The lncRNA PCAT1 is correlated with poor prognosis and promotes cell proliferation, invasion, migration and EMT in osteosarcoma. *Onco Targets Ther*, 2018, 11:629-638.

15 Chen Y, Li J, Xiao J K, *et al*. The lncRNA NEAT1 promotes the epithelial-mesenchymal transition and metastasis of osteosarcoma cells by sponging miR-483 to upregulate STAT3 expression. *Cancer Cell Int*, 2021, 21(1):90.

16 Brabletz T. EMT and MET in metastasis: where are the cancer stem cells?. *Cancer Cell*, 2012, 22(6):699-701.

17 De Craene B, Berx G. Regulatory networks defining EMT during cancer initiation and progression. *Nat Rev Cancer*, 2013, 13(2):97-110.

18 Wang L, Zhang Z G, Zhang R L, *et al*. Matrix metalloproteinase 2 (MMP2) and MMP9 secreted by erythropoietin-activated endothelial cells promote neural progenitor cell migration. *J Neurosci*, 2006, 26 (22):5996-6003.

19 Yang J S, Lin C W, Hsieh Y S, *et al*. Selaginella tamariscina (Beauv.) possesses antimetastatic effects on human osteosarcoma cells by decreasing MMP-2 and MMP-9 secretions via p38 and Akt signaling pathways. *Food Chem Toxicol*, 2013, 59:801-807.

20 Chen S, Jin Z, Dai L, *et al*. Aloperine induces apoptosis and inhibits invasion in MG-63 and U2OS human osteosarcoma cells. *Biomed Pharmacother*, 2018, 97:45-52.

21 Schiavone K, Garnier D, Heymann M-F, *et al*. The heterogeneity of osteosarcoma: the role played by cancer stem cells. *Adv Exp Med Biol*, 2019, 1139:187-200.

22 Zhang Y, Cheng H, Li W, *et al*. Highly - expressed P2X7 receptor promotes growth and metastasis of human HOS/MNNG osteosarcoma cells via PI3K/Akt/GSK3β/β - catenin and mTOR/HIF1α/VEGF signaling. *Int J Cancer*, 2019, 145(4):1068-1082.

23 Ma L, Zhang L, Guo A, *et al*. Overexpression of FER1L4 promotes the apoptosis and suppresses epithelial-mesenchymal transition and stemness markers via activating PI3K/AKT signaling pathway in osteosarcoma cells. *Pathol Res Pract*, 2019, 215(6):152412.

24 Ma K, Zhang C, Li W. Gamabufotalin suppressed osteosarcoma stem cells through the TGF-beta/periostin/PI3K/AKT pathway. *Chem Biol Interact*, 2020, 331:109275.

25 Yu X, Wang Q, Zhou X, *et al*. Celastrol negatively regulates cell invasion and migration ability of human osteosarcoma via downregulation of the PI3K/Akt/NF-kappaB signaling pathway in vitro. *Oncol Lett*, 2016, 12(5):3423-3428.

26 Li C, Zhou X, Sun C, *et al*. Isoliquiritigenin inhibits the proliferation, apoptosis and migration of osteosarcoma cells. *Oncol Rep*, 2019, 41(4): 2502-2510.

27 Hsu M J, Peng S F, Chueh F S, *et al*. Lupeol suppresses migration and invasion via p38/MAPK and PI3K/Akt signaling pathways in human osteosarcoma U-2 OS cells. *Biosci Biotechnol Biochem*, 2019, 83(9): 1729-1739.

28 Blagodatski A, Klimenko A, Jia L, *et al*. Small molecule wnt pathway modulators from natural sources: history, state of the art and perspectives. *Cells*, 2020, 9(3):589.

29 Liu W, Zhao Z, Wang Y, *et al*. Dioscin inhibits stem-cell-like properties and tumor growth of osteosarcoma through Akt/GSK3/beta-catenin signaling pathway. *Cell Death Dis*, 2018, 9(3):343.

30 Cai Y, Mohseny A B, Karperien M, *et al*. Inactive Wnt/beta-catenin pathway in conventional high-grade osteosarcoma. *J Pathol*, 2010, 220 (1):24-33.

31 Jin H, Luo S, Wang Y, *et al*. MiR-135b stimulates osteosarcoma recurrence and lung metastasis via notch and wnt/beta-catenin signaling. *Mol Ther Nucleic Acids*, 2017, 8:111-122.

32 Vega O A, Lucero C M J, Araya H F, *et al*. Wnt/β-catenin signaling activates expression of the bone-related transcription factor RUNX2 in select human osteosarcoma cell types. *J Cell Biochem*, 2017, 118(11): 3662-3674.

33 Nomura M, Rainusso N, Lee Y C, *et al*. Tegavivint and the beta-Catenin/ALDH axis in chemotherapy-resistant and metastatic osteosarcoma. *J Natl Cancer Inst*, 2019, 111(11):1216-1227.

34 Tian H, Zhou T, Chen H, *et al*. Bone morphogenetic protein-2 promotes osteosarcoma growth by promoting epithelial-mesenchymal transition (EMT) through the Wnt/beta-catenin signaling pathway. *J Orthop Res*, 2019, 37(7):1638-1648.

35 Wang S, Zhang D, Han S, *et al*. Fibulin-3 promotes osteosarcoma invasion and metastasis by inducing epithelial to mesenchymal transition and activating the Wnt/β -catenin signaling pathway. *Sci Rep*, 2017, 7(1):6215.

36 Fang F, VanCleave A, Helmuth R, *et al*. Targeting the Wnt/β-catenin pathway in human osteosarcoma cells. *Oncotarget*, 2018, 9(95):36780-36792.

37 Liu D, Chen L, Zhao H, *et al*. Small molecules from natural products targeting the Wnt/β-catenin pathway as a therapeutic strategy. *Biomed Pharmacother*, 2019, 117:108990.

38 Chang J, Li Y, Wang X, *et al*. Polyphyllin I suppresses human osteosarcoma growth by inactivation of Wnt/beta-catenin pathway in vitro and in vivo. *Sci Rep*, 2017, 7(1):7605.

39 Dai G, Zheng D, Wang Q, *et al*. Baicalein inhibits progression of osteosarcoma cells through inactivation of the Wnt/β-catenin signaling pathway. *Oncotarget*, 2017, 8(49):86098-86116.

40 Yang S, Chen J, Tan T, *et al*. Evodiamine exerts anticancer effects against 143B and MG63 cells through the wnt/beta-catenin signaling pathway. *Cancer Manag Res*, 2020, 12:2875-2888.

41 Tan T, Chen J, Hu Y, *et al*. Dihydrotanshinone I inhibits the growth of osteosarcoma through the Wnt/beta-catenin signaling pathway. *Onco Targets Ther*, 2019, 12:5111-5122.

42 Javaid N, Choi S. Toll-like receptors from the perspective of cancer treatment. *Cancers (Basel)*, 2020, 12(2):297.

43 Zhou J, Liu Q, Qian R, *et al*. Paeonol antagonizes oncogenesis of osteosarcoma by inhibiting the function of TLR4/MAPK/NF- κB pathway. *Acta Histochem*, 2020, 122(1):151455.

44 Min C, Eddy S F, Sherr D H, *et al*. NF-kappaB and epithelial to mesenchymal transition of cancer. *J Cell Biochem*, 2008, 104(3): 733-744.

45 Basseres D S, Baldwin A S. Nuclear factor-kappaB and inhibitor of kappaB kinase pathways in oncogenic initiation and progression. *Oncogene*, 2006, 25(51):6817-6830.

46 Nishimura A, Akeda K, Matsubara T, *et al*. Transfection of NF-kappaB decoy oligodeoxynucleotide suppresses pulmonary metastasis by murine osteosarcoma. *Cancer Gene Ther*, 2011, 18(4):250-259.

47 Kishida Y, Yoshikawa H, Myoui A. Parthenolide, a natural inhibitor of

Nuclear Factor-kappaB, inhibits lung colonization of murine osteosarcoma cells. *Clin Cancer Res*, 2007, 13(1):59-67.

48 Li Y, Zhang Z N, Zhao H M, *et al*. Matrine inhibits the invasive properties of human osteosarcoma cells by downregulating the ERK-NF-kappaB pathway. *Anticancer Drugs*, 2014, 25(9):1035-1043.

49 Zhang G, Jin C, Zhu Y, *et al*. Sulforaphene inhibits the progression of osteosarcoma via regulating FSTL1/NF-kappaB pathway. *Life Sci*, 2020, 263:118485.

50 Chang J, Wang H, Wang X, *et al*. Molecular mechanisms of Polyphyllin I-induced apoptosis and reversal of the epithelial-mesenchymal transition in human osteosarcoma cells. *J Ethnopharmacol*, 2015, 170: 117-127.

51 Kumar S, Singh S K, Rana B, *et al*. The regulatory function of mixed lineage kinase 3 in tumor and host immunity. *Pharmacol Ther*, 2021, 219:107704.

52 González-Rubio G, Fernández-Acero T, Martín H, *et al*. Mitogen-activated protein kinase phosphatases (MKPs) in fungal signaling: conservation, function, and regulation. *Int J Mol Sci*, 2019, 20(7):1709.

53 Rajoria S, Suriano R, Wilson Y L, *et al*. 3, 3′-diindolylmethane inhibits migration and invasion of human cancer cells through combined suppression of ERK and AKT pathways. *Oncol Rep*, 2011, 25 (2):491-497.

54 Dai F, Luo F, Zhou R, *et al*. Calponin 3 is associated with poor prognosis and regulates proliferation and metastasis in osteosarcoma. *Aging (Albany NY)*, 2020, 12(14):14037-14049.

55 Lu K H, Yang H W, Su C W, *et al*. Phyllanthus urinaria suppresses human osteosarcoma cell invasion and migration by transcriptionally inhibiting u-PA via ERK and Akt signaling pathways. *Food Chem Toxicol*, 2013, 52:193-199.

56 Hsieh Y S, Chu S C, Yang S F, *et al*. Silibinin suppresses human osteosarcoma MG-63 cell invasion by inhibiting the ERK-dependent c-Jun/AP-1 induction of MMP-2. *Carcinogenesis*, 2007, 28(5): 977-987.

57 Cheng G, Gao F, Sun X, *et al*. Paris saponin VII suppresses osteosarcoma cell migration and invasion by inhibiting MMP2/9 production via the p38 MAPK signaling pathway. *Mol Med Rep*, 2016, 14(4):3199-3205.

58 Kang H M, Park B S, Kang H K, *et al*. Delphinidin induces apoptosis and inhibits epithelial-to-mesenchymal transition via the ERK/p38 MAPK-signaling pathway in human osteosarcoma cell lines. *Environ Toxicol*, 2018, 33(6):640-649.

59 Cheng H L, Hsieh M J, Yang J S, *et al*. Nobiletin inhibits human osteosarcoma cells metastasis by blocking ERK and JNK-mediated MMPs expression. *Oncotarget*, 2016, 7(23):35208-35223.

60 Tingting R, Wei G, Changliang P, *et al*. Arsenic trioxide inhibits osteosarcoma cell invasiveness via MAPK signaling pathway. *Cancer Biol Ther*, 2014, 10(3):251-257.

61 Jie Z, Xie Z, Zhao X, *et al*. Glabridin inhibits osteosarcoma migration and invasion via blocking the p38- and JNK-mediated CREB-AP1 complexes formation. *J Cell Physiol*, 2019, 234(4):4167-4178.

62 Chen H J, Lin C M, Lee C Y, *et al*. Kaempferol suppresses cell metastasis via inhibition of the ERK-p38-JNK and AP-1 signaling pathways in U-2 OS human osteosarcoma cells. *Oncol Rep*, 2013, 30 (2):925-932.

63 Zhang P, Yang Y, Zweidler-McKay P A, *et al*. Critical role of notch signaling in osteosarcoma invasion and metastasis. *Clin Cancer Res*, 2008, 14(10):2962-2969.

64 Yu H, Pardoll D, Jove R. STATs in cancer inflammation and immunity: a leading role for STAT3. *Nat Rev Cancer*, 2009, 9(11):798-809.

65 Li Y, Zhang J, Ma D, *et al*. Curcumin inhibits proliferation and invasion of osteosarcoma cells through inactivation of Notch-1 signaling. *FEBS J*, 2012, 279(12):2247-2259.

66 Li Y, Zhang J, Zhang L, *et al*. Diallyl trisulfide inhibits proliferation, invasion and angiogenesis of osteosarcoma cells by switching on suppressor microRNAs and inactivating of Notch-1 signaling. *Carcinogenesis*, 2013, 34(7):1601-1610.

67 Jin X, Wang C, Wang L. Costunolide inhibits osteosarcoma growth and metastasis via suppressing STAT3 signal pathway. *Biomed Pharmacother*, 2020, 121:109659.

68 Xie T, Ren H Y, Lin H Q, *et al*. Sinomenine prevents metastasis of human osteosarcoma cells via S phase arrest and suppression of tumor-related neovascularization and osteolysis through the CXCR4-STAT3 pathway. *Int J Oncol*, 2016, 48(5):2098-2112.

Research Progress on the Effect and Mechanism of Effective Components of Traditional Chinese Medicine in Preventing and Treating Osteosarcoma Metastasis

Zhao Fulai[1,2], Chang Junli[1,2], Zhi Wenlan[1,2], Wang Xiaobo[1,2], Sun Xingyuan[1,2], Ma Xiaoping[1,2], Shi Qi[1,2], Wang Yongjun[1,2], Yang Yanping[1,2]

(1. Longhua Hospital, Shanghai University of Traditional Chinese Medicine, Shanghai 200032, China; 2.Key Laboratory of Musculoskeletal Theory and Treatment Principle, Ministry of Education, Shanghai 200032, China)

Abstract: Osteosarcoma is a primary and aggressive malignant tumor, which is prone to early metastasis and lacks effective therapeutic targets. Surgery, chemoradiotherapy and the combination of the two are the main treatment methods at present, but the overall survival rate of patients with metastatic osteosarcoma has not been significantly improved. Osteosarcoma belongs to the category of "cancer disease" in traditional Chinese medicine. Traditional Chinese medicine has a long history in the treatment of osteosarcoma and can play an active role in the prevention and treatment of osteosarcoma metastasis, but its mechanism is not clear yet. Signaling pathway plays an important role in the pathogenesis of osteosarcoma metastasis and is one of the targets for the study of pathologic mechanism of osteosarcoma metastasis and therapeutic drugs. In recent years, large number of studies have been conducted on the signaling pathways related to the prevention and treatment of osteosarcoma metastasis by effective components of traditional Chinese medicine. This review is expected to provide reference for the prevention and treatment of osteosarcoma metastasis by traditional Chinese medicine, new drug development and clinical diagnosis and treatment.

Keywords: Active components of traditional Chinese medicine, Osteosarcoma, Metastasis, Mechanism of action, Research progress

（责任编辑：周阿剑、郭思宇，责任译审：周阿剑，审稿人：王瑀、张志华）