

• 综述 •

细胞外囊泡的中药药理作用及其与中医“气”的研究进展

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摘要: 细胞外囊泡 (extracellular vesicles, EVs) 是生物体内一类重要的活性微泡结构, EVs 包裹并传递功能物质如 miRNAs、转录因子、蛋白质等, 是介导细胞通讯和器官对话的重要载体。近年来研究表明, 相当多的中药具有调控 EVs 的药理作用, 在治疗疾病中发挥独特的跨器官、远程作用; 有些中药本身还含有植物源性 EVs, 可直接参与疾病的治疗。“气”是中医学的核心理论之一, 在人体生理病理及中药药理过程中发挥多种重要作用, 但其作用的科学内涵及潜在的物质载体尚不尽清晰。最新研究提示, EVs 的作用与“气”的作用有潜在相关性。为此, 本文综述了近年来益气中药调控 EVs 在治疗心血管疾病、神经系统疾病、肝脏疾病、肾性疾病、恶性肿瘤等疾病中的作用。EVs 可能作为中间媒介物质, 在部分中药治疗疾病的药理作用中扮演了重要角色。EVs 具有远程输送的特性, 与中医“气”的运动性相符合; EVs 携带多种功能性分子, 与“气”的功能性相符合。EVs 可能作为中医“气”的潜在物质基础, 其功能值得深入研究。

关键词: 细胞外囊泡; 外泌体; 中药; 中医“气”; 细胞通讯; 器官对话; 远程调控

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Research advances on extracellular vesicles-based regulation by traditional Chinese medicines and the relationship with TCM "Qi"

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Abstract: Extracellular vesicles (EVs) are an important type of active microvesicles. EVs encapsulate and transfer functional substances such as miRNAs, transcription factors and proteins, which are important vectors for cell communication and organ dialogue. In recent years, studies have shown that quite a number of Chinese medicinal herbs have the pharmacological effect of regulating EVs, and play a unique trans-organ and remote role in the treatment of diseases. Some Chinese medicinal herbs also contain plant-derived EVs themselves, which can be directly involved in the treatment of diseases. As one of the core theories of traditional Chinese medicines (TCM), Qi plays a variety of important roles in the physiological and pathological processes of human body and pharmacology. However, the scientific connotation of Qi's role and the potential material carrier are still unclear. The latest research suggests that the effect of EVs is potentially related to that of Qi. Therefore, this paper reviews the effect of Qi nourishing Chinese medicinal herbs in regulating EVs in the treatment of cardiovascular diseases, nervous system diseases, liver diseases, renal diseases, malignant tumors and other diseases in recent years. EVs may play an important role in the pharmacological effect of some Chinese medicinal herbs in the treatment of diseases as an intermediary substance. EVs have the characteristics of long-distance transportation, which is consistent with the movement of Qi in TCM. EVs carry a variety of functional molecules, which is consistent with

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the function of Qi. As the potential material basis of Qi in TCM, the function of EVs is worth further study.

Key words: extracellular vesicle; exosome; Chinese medicinal herb; Qi in TCM; intercellular communication; interorgan crosstalk; remote regulation

细胞外囊泡 (extracellular vesicles, EVs) 是生物体内一类重要的活性微泡结构, EVs 包裹并传递功能物质如 miRNAs、转录因子、蛋白质等, 是介导细胞通讯、器官对话的重要载体。EVs 在人体生理和病理过程中的重要作用正不断被研究和发现, EVs 的远程输送特性和功能调适能力, 使研究者逐渐更深刻地认识到人体作为一个巨系统的复杂性, 疾病的进展往往不仅有疾病靶细胞/靶器官的异常改变, 还可能受到其他细胞/器官的间接、远程调控, EVs 介导的细胞通讯、器官对话是当前生物医学研究的前沿领域之一。

近年来研究表明, 相当多的中药具有调控 EVs 的药理作用, 在治疗疾病中发挥独特的跨器官、远程作用; 以人参为代表的部分中药本身还含有植物源性 EVs, 可直接参与疾病的治疗。中药的作用机制不局限于病灶靶点的直接对抗作用, 还包括通过调控中间媒介物质发挥的跨器官 (脏腑)、跨系统、远程的作用^[1,2]。

“气”是中医学里一个独特的概念, 围绕“气”的概念和功能, 中医学有“正气内存, 邪不可干”等重要认识发病病机的独特思想, 由于气虚导致的虚证在治疗上以益气法为治疗原则。益气治法及益气中药的总体思想是以补益来治疗疾病, 是中医与西医对抗医学思想的重要区别之一。中医学认为“气”是人体基础物质, 具有转换和传输的性质; 这与 EVs 跨器官、远程作用相符合。为此, 本文以益气中药对 EVs 的调控作用为切入点, 综述近年来中药调控 EVs 在治疗心血管疾病、神经系统疾病、肝脏疾病、肾性疾病、恶性肿瘤等疾病中的作用, 进一步探讨 EVs 可能作为中间媒介物质在中药治疗疾病中所发挥的作用及规律, 以期为“气”的科学内涵找到突破口, 与同行进行交流探讨。

1 中医“气”的概念及内涵分析

中医学的“气”既是人体的重要组成部分, 又是激发和调控人体生命活动的动力源泉。以中医传统理论为基础, 结合近现代对中医“气”的含义阐释, “气”的科学内涵可能包括以下几个方面: 首先, “气”可能是能量的化生。“气”是人体内生命力很强、不断运动的极细微物质, 其与现代医学中 ATP、脂质、葡萄糖等高能物质或能量载体的作用有一定的相关性^[3]; 其次, “气”可能是具有生理功能的物质。“气”可以激发和调控人体生命活动, 其功能与分泌型细胞因子及免疫蛋白等现代

医学中物质相似^[4]; 第三, “气”具有传递属性。通过气的升降出入, 人体物质、能量、信息进行运动和转化, 人体的状态所出现的各种变化, 皆是气化的结果^[5]。

2 中医“气”与 EVs 之间的潜在关系分析

EVs 是由细胞分泌释放到细胞外基质的脂质双分子层膜结构的生物活性微泡, 主要分为外泌体、微囊泡和凋亡小体 3 类^[6]。EVs 可以介导细胞间的物质交换, 交换过程中细胞内多种遗传物质 (miRNA 和 mRNA) 及蛋白质被包裹入 EVs 中并向细胞外环境释放^[7]。不同细胞来源的 EVs 蛋白组成存在差异, 但它们有着共同的蛋白质组分, 包括跨膜蛋白超家族 (CD63、CD9 和 CD81 等)、膜转运和融合相关蛋白、整合素等^[8]。已有研究证明, EVs 通过转运遗传物质和蛋白质, 在心血管、神经系统、肝脏疾病、肾性疾病及恶性肿瘤等疾病的诊断和治疗中起到重要作用^[8-10]。

中医“气”与 EVs (以外泌体为代表) 之间的关系正逐渐受到关注。EVs 的特点与中医辨证的整体性、变化性极其相似, 可用来阐释中医脏腑相关理论的物质基础^[11]。EVs 作为一种生物标志物的载体, 可用于中医辨证论治中证型的判断与治疗^[12]。EVs 的源细胞性、分布广泛性、生物相容性和靶向运输性等特点均符合中医理论对物质基础的要求, EVs 中的 miRNA 比游离的 miRNA 更具有标志物的潜质^[13]。Chen 等^[14]提出 EVs 作用属于中医药超分子“气析”理论, 并建立了中药超分子“印迹模板”的研究方法。此外, EVs 的生物学特点及功能与中医学和针灸学中物质基础有契合之处, 因此 EVs 研究可能为中医及针灸作用机制的阐释提供实验依据, 同时可动态评估临床疗效, 以及通过 EVs 载药手段可拓宽中医及针灸的临床应用^[15]。然而, 关于中医“气”与 EVs 之间的相关性是否存在, 具体内涵是什么, 仍待进一步阐释。

3 以益气中药为代表的中药调控 EVs 作用的研究

益气治法和益气中药是中医临床治疗气虚证的治法和方药, 本课题组以益气中药对 EVs 的调控作用作为切入点, 探寻中医“气”与 EVs 之间的相关性。本文总结了益气中药通过调控 EVs 发挥对心血管疾病、神经系统疾病、肝脏疾病、肾性疾病和恶性肿瘤等疾病治疗作用的研究 (详见表 1^[16-28]), 以及益气中药植物源性 EVs 的最新研究, 拟从益气中药的作用探索中医“气”与 EVs 之间的联系。

Table 1 Qi-nourishing traditional Chinese medicines (TCM) exerts therapeutic effect on diseases by regulating extracellular vesicles (EVs). TAB2: TGF-beta activated kinase 1 binding protein 2; PI3K: Phosphatidylinositol 3-kinase; mTOR: Mechanistic target of rapamycin; GTPase: GTP-binding protein; UTX: Ubiquitously transcribed tetratricopeptide repeat, chromosome X; NRP-1: Neuropilin-1; VEGF: Vascular endothelial growth factor; SphK1: Sphingosine kinase 1; AKT: Protein kinase B; NF-κB: Nuclear factor kappa B; NLRP3: NOD-like receptor thermal protein domain associated protein 3; FN: Fibronectin; TGF-1: Transforming growth factor-1

Qi-nourishing TCM and its components	Disease	Subject of study	EVs type	Mechanism of action	Reference
Yiqi Huoxue Huatan Decoction (raw Astragalus root, Coptidis Rhizoma, Salvia Miltiorrhizae, Hirudo and whole Trichosanthis Fructus)	Atherosclerosis	Mouse	Macrophage exosomes	miRNA-let-7-5p/TAB2 signal pathway	[16]
Qishen Yiqi Dropping Pill (Astragalus root, Salvia Miltiorrhizae, Panax notoginseng, Dalbergia Odorifera)	Myocardial ischemia-reperfusion injury	Rat	Exosomes derived from mesenchymal stem cells	Regulating exocrine miR-155 targeting and mediating PI3K/mTOR signal pathway	[17]
Suxiao Jiuxin pill (Ligusticum Chuanxiong, Borneol)	Myocardial ischemia-reperfusion injury	Mouse	Exosomes from cardiac mesenchymal stem cells	Exosome secretion from cardiac mesenchymal stem cells <i>via</i> a GTPase-dependent pathway	[18]
Bu-Shen-Yi-Sui Capsule (Rehmanniae Radix, Rehmanniae Radix Praeparata, Polygoni Multiflori Radix, Rhei Radix et Rhizoma, Leonuri Herba, Fritillariae Thunbergii Bulbus, Hirudo, Scorpio, Gastrodiae Rhizoma, Forsythiae Fructus)	Multiple sclerosis	Mouse HL-1 cardiomyocyte line	Cardiac mesenchymal stem cell microvesicles	Decreasing H3K27 demethylase UTX expression in cardiomyocytes	[19]
Buyang Huanwu decoction (Astragalus root, Angelica Sinensis, Red Peony root, Ligusticum Chuanxiong, Flos Carthami, Peach seed, Lumbricus)	Acute coronary syndrome	Mice with autoimmune encephalomyelitis, oligodendrocyte progenitor cells	Serum exosomes	Regulation of the NRP-1 and GTX proteins and miRs in serum exosomes, which drive promyelination	[20]
Salidroside	Liver fibrosis	Rat	Exosomes from bone marrow stromal cells	Augmenting angiogenic miRNA and VEGF expression in exosomes secreted by bone marrow stromal cells and elevating angiogenesis in brain	[21]
Kaempferitrin	Diabetes	Mouse	Serum exosomes	Alleviating LX-2 cells migration and activation induced by exosomal SphK1 by inhibiting the AKT activation	[22]
Jian-Pi-Yi-Shen formula (Astragali Radix, Atractylodis Macrocephalae Rhizoma, Dioscoreae Rhizoma, Cistanches Herba, Amomi Fructus Rotundus, Salviae Miltiorrhizae Radix et Rhizoma, Rhei Radix et Rhizoma, Glycyrrhizae Radix et Rhizoma Praeparata cum Melle)	Chronic kidney disease	Liver cancer cell line HepG2	Liver cells secrete vesicles	Regulating extracellular vesicle secretions and lipid profile	[23]
Zhen-Wu-Tang (Aconitum Carmichaeli debx, Poria Cocos, Atractylodes Macrocephala Koidz, Paeonia Lactiflora Pall, Zingiber Officinale Roscoe)	Immunoglobulin A nephropathy	Rat	Serum exosomes	Modulation of exosomal miR-192-5p, miR-194-5p, miR-802-5p and miR-143-3p	[24]
Baoshen Tongluo recipe (Radix Rehmanniae Praeparata, Astragalus root, Semen Cuscutae, Salviae Miltiorrhizae Radix et Rhizoma, Hirudo)	Diabetic nephropathy	Rat	Exosomes from renal tubular epithelial cells	Regulating exosomes to inhibit NF-κB/NLRP3 pathway	[25]
Tongxinluo (Panax Ginseng, Hirudo, Scorpion, Red Peony root, Periostracum Cicadae, Ground Beettle, Centipede, Sanders, Dalbergia Odorifera, Olibanum, Semen Ziziphi Spinosae, Borneol)	Diabetic nephropathy	Type 2 diabetic KK-Ay mouse	Exosomes from glomerular mesangial cells	Reducing the expression of collagen IV, FN and miR-192 in exosomes	[26]
			Exosomes from endothelial cells	Preventing the transfer of TGF-β1 from glomerular endothelial cells to glomerular mesangial cells <i>via</i> exosomes	[27]

Qi-nourishing TCM and its components	Disease	Subject of study	EVs type	Mechanism of action	Reference
Yiqi Compound Recipe (Astragalus root, Radix Codonopsis)	Breast cancer	MDA-MB-231 breast cancer cell line	Exosomes released by triple negative breast cancer (MDA-MB-231) platinum resistant cells	Inhibiting exosomes transmitting drug resistance information	[28]

Continued

3.1 益气中药调控 EVs 治疗心血管疾病 中医认为心气不足多表现为心悸、神疲及气虚症状, 而气虚日久则多致心脉瘀血痹阻, 出现胸闷、心痛等症状。气虚血瘀是公认的心血管疾病主要中医病机之一, 益气中药在中医临床治疗心系病证中发挥重要的作用^[29]。近 5 年来, EVs 已被应用于心血管疾病的诊断及治疗中^[30], 益气活血类中药对 EVs 的调控作用逐步受到关注。研究表明, 益气活血化痰方可能调节巨噬细胞 EVs, 通过 miRNA-let-7-5p/转化生长因子 β 活化激酶 1 结合蛋白 2 (TGF- β activated kinase 1 binding protein 2, TAB2) 信号通路发挥治疗动脉粥样硬化作用^[16]。中药复方芪参益气滴丸联合 EVs 能显著改善心肌缺血再灌注损伤及调控巨噬细胞表型、改善炎症微环境^[17]。

研究发现, 速效救心丸通过 GTP 结合蛋白 (GTP-binding protein, GTPase) 依赖的途径促进小鼠心肌间充质干细胞分泌 EVs, 调节心脏内稳态, 使受体心肌细胞的表观遗传染色质重塑, 从而促进心肌细胞增殖^[18,19]。

3.2 益气中药调控 EVs 治疗神经系统疾病 EVs 的结构性质使其具备能穿透血脑屏障, 且无排异反应的特点。因此, EVs 在神经系统疾病的诊断和治疗中作用已逐渐受到重视^[31]。中医治疗脑相关疾病多采用益气补肾之品, 通过补益肾气, 进而生髓、充脑, 治疗脑病。补肾益髓胶囊能显著降低多发性硬化, 从而减少自身免疫性脑脊髓炎神经损伤后的复发率, 并促进再髓鞘化。该药物对再髓鞘化作用的机制可能是通过促进骨髓间充质干细胞分泌 EVs 入血, 血中 EVs 可进入中枢, 其所包裹的神经纤毛蛋白-1 (neuropilin-1, NRP-1)、GTX 蛋白和 miRNAs 驱动再髓鞘化, 从而改善多发性硬化^[20]。这是一种独特的、通过 EVs 介导的跨器官、远程的中药间接调控作用^[1]。

补阳还五汤联合间充质干细胞可以通过促进血管生成来减轻缺血性损伤, 该药物干预增加了大鼠骨髓间充质干细胞分泌的 EVs 中血管生成相关 miRNAs 和血管内皮生长因子 (vascular endothelial growth factor, VEGF) 的表达, 增加了大鼠脑内的血管生成^[21]。

3.3 益气中药调控 EVs 治疗肝脏疾病 EVs 在肝脏疾病的诊断及治疗中的作用, 主要体现在其调节肝脏

代谢、抑制肝纤维化、改善肝脏缺血/再灌注损伤等方面^[32-34]。对于肝失疏泄证的中医治法多以理气疏肝为主, 而对于肝不藏血证多以益气养阴为主。

如中药红景天具有益气活血通脉的作用, 红景天的活性成分红景天苷可能通过减少包裹鞘氨醇激酶-1 (sphingosine kinase 1, SphK1) 的肝窦内皮细胞 EVs 诱导的肝星状细胞活化和迁移, 发挥抑制四氯化碳诱导的小鼠肝纤维化的作用^[22]。肉桂具有温运阳气以鼓舞气血生长之效, 现代研究表明肉桂属中药中提取的山柰酚可调节 HepG2 细胞分泌 EVs, 从而发挥调控脂质代谢相关基因的作用^[23]。

3.4 益气中药调控 EVs 治疗肾性疾 EVs 介导的细胞间通讯及对损伤修复作用在肾性疾病的诊断和治疗中已受到关注^[35]。中医认为肾主藏精, 主水, 主纳气, 当肾精亏虚或肾气不足时, 中医治法多以补肾益气填精法为主。

研究发现, 健脾益肾方可显著降低慢性肾病大鼠血清肌酐和尿素氮水平, 减轻肾脏病理损伤。该方主要通过上调 EVs 中 miRNA (miR-192-5p、miR-194-5p、miR-802-5p 和 miR-143-3p) 表达, 保护肾脏免受腺嘌呤诱导形成慢性肾病^[24]。真武汤可通过调节肾小管上皮细胞 EVs 分泌, 抑制核因子 κ B (nuclear factor kappa B, NF- κ B)/NOD 样受体热蛋白结构域相关蛋白 3 (NOD-like receptor thermal protein domain associated protein 3, NLRP3) 通路的激活, 从而减轻肾功能不全, 发挥对免疫球蛋白 A 肾病的保护作用^[25]。

保肾通络方可以降低糖尿病肾病肾小球系膜细胞增殖活性, 抑制细胞外基质增生, 同时下调 EVs 中 miR-192 的表达, 从而减少胶原 IV 及纤维连接蛋白 (fibronectin, FN) 蛋白的表达, 起到对糖尿病肾病的治疗作用^[26]。中药复方通心络对糖尿病肾病患者有治疗作用, 能阻止 TGF- β 1 通过 EVs 通讯从肾小球内皮细胞转移到肾小球系膜细胞, 这可能是该方治疗糖尿病肾病的机制之一^[27]。

3.5 益气中药调控 EVs 治疗恶性肿瘤 EVs 的功能取决于其来源的细胞类型, 肿瘤来源的 EVs 参与肿瘤细胞与基底细胞的遗传信息交换、细胞迁移、细胞分化、肿瘤侵袭等方面^[36]。顺铂耐药细胞株分泌的 EVs 显著多于敏感细胞株, 而进一步将耐药细胞株分泌的

EVs传递给敏感细胞株后,后者的耐药性明显提高^[37]。肿瘤耐药信息在肿瘤细胞间的传递和通讯,是肿瘤耐药的重要共性机制之一^[38],在这一过程中,EVs发挥了重要的耐药信息传递的媒介作用^[39]。EVs中包裹的miRNAs(如miR-155^[40])可能是肿瘤耐药细胞向周围敏感肿瘤细胞传递耐药性信息的重要物质基础。除了miRNAs, EVs中包裹的P-糖蛋白也可以介导和传递肿瘤耐药性^[41]。

肿瘤属于中医癥瘕范畴,气虚血瘀是癥瘕主要的证型之一,其中医临床代表性治法为益气活血、扶正固本。临床观察和动物实验研究表明,以益气中药为主的扶正固本法治疗乳腺癌,可有效地降低乳腺癌复发、转移^[42,43],进一步的研究证实益气健脾药联合化疗药可有效降低耐药性^[44]。三阴性乳腺癌细胞顺铂耐药细胞株释放的EVs,可被敏感细胞株摄取并引起耐药,这与上述研究发现是类似的;但是,非常有意思的是,给予党参和黄芪(2:1)组成的益气小复方后,可有效地阻断EVs传递的耐药性^[28]。

上述结果提示,肿瘤细胞耐药株的EVs具有传递耐药信息的作用,而益气中药通过调控EVs,阻断耐药性的传递。耐药株肿瘤细胞EVs传递的信息,或可理解为“邪气”;益气中药对肿瘤耐药信息传递的抑制作用,或可理解为补益“正气”,以抑制肿瘤的耐药性。益气中药通过干预肿瘤相关EVs扶正祛邪,可能是一个非常独特且有重要前景的肿瘤治疗新策略。

3.6 益气中药植物源性EVs及纳米颗粒的治疗作用 除了益气中药干预EVs的作用,以人参为代表的益气中药植物自身分泌EVs或分离出的EVs样人参纳米颗粒的治疗作用也逐渐受到关注。人参源性EVs(G-Exos)可有效将miRNAs转移到骨髓间充质干细胞中,刺激其向神经细胞分化。G-Exos被证明可以在体外和体内将植物源性miRNAs转移到哺乳动物干细胞中促进神经分化,在神经再生医学中具有巨大潜力^[45]。EVs样人参纳米颗粒可以重新编程肿瘤相关巨噬细胞,增加趋化因子配体5和趋化因子配体9的分泌,将CD8⁺T细胞招募到肿瘤位置,对程序性细胞死亡蛋白-1(programmed cell death protein 1, PD-1)单抗治疗具有协同作用^[46]。

4 中医“气泡”的提出与思考

根据中医“气”的作用,比如“气”的推动作用、温煦作用、防御作用、营养作用和气化作用等,结合EVs的功能研究进行系统梳理,本课题组发现“气”与EVs在物质基础与功能作用上可能存在一定的相关性。基于益气中药通过调节EVs包裹蛋白和miRNAs表达,发挥改善疾病症状的作用;或含益气中药EVs及益气中

药植物源性EVs直接产生治疗作用,都提示EVs可能是中医“气”发挥功能的实质载体。EVs由双层脂质膜组成,其包裹的内容物包括蛋白质、脂质体、miRNAs、lncRNA等调控人体生命活动的物质。益气中药对EVs有调控作用,可能是中药发挥益气作用的一个可能的途径。此外,EVs由不同的细胞分泌,可运行于全身,实现跨脏腑远程传输。EVs通过转运遗传物质和蛋白质,影响甚至改善机体状态。

基于以上,本课题组提出一个有趣的名称或许可以代表两者之间的关系——中医“气泡”(图1)。在生理病理过程中,中医“气泡”发挥其自身的运输性,将其中包含的生物活性物质及基因转录等信息,精准地传递至目标部位。中药对EVs的分泌和传输的促进作用,提示“气泡”的表现形式可以是以EVs为代表的具有远程靶向运输功能的超微囊泡。

值得注意的是,EVs中同一类内容物在疾病过程中可能发挥不同的作用,如EVs中miRNAs介导了肿瘤成纤维细胞、正常纤维细胞和癌细胞之间的通讯,从而影响肿瘤侵袭和转移的过程。肿瘤成纤维细胞EVs-miRNAs中miRNA-21表达升高,会减缓癌细胞凋亡及增加细胞的耐药性;而miRNA-195表达升高,则可抑制癌细胞的侵袭作用^[47]。EVs-miRNAs的作用与其含量变化以及在疾病不同时期的作用密切相关,根据作用趋向的不同,或可归结到中医“正气”和“邪气”的作用上。

在中药调控EVs研究领域,现有的研究不仅针对常规给药后中药对EVs分泌及包裹内容物表达量的作用解析,也扩展到了中药干预后EVs作为治疗手段回输的作用研究。如通过尾静脉注射的方式将川芎嗪干预小鼠血清EVs回输至慢性肝损伤小鼠体内,发现川芎嗪干预后的血清EVs可显著延缓肝纤维化进程^[48]。同时,中药植物源性EVs的治疗作用也受到关注。如中药芦荟分泌EVs可以通过激活核因子E2相关因子2促进抗氧化防御机制和伤口愈合过程^[49]。以上研究方向的拓展,极大地丰富了中药的作用途径,同时对中医理论的科学内涵阐释也进行了一定补充。

中药干预EVs在基础研究中显示出明确的治疗作用,且具有容易吸收、穿透力强、无排异反应的优势。将中药干预EVs作为生物制剂药物用于治疗临床疾病可能成为未来治疗的一种新手段。因此,批量生产质量稳定、靶向明确的中药干预EVs是目前面临的技术挑战。中医“气泡”能否成为今后临床治疗的新趋势,一方面与技术优化相关,如将基因编辑靶向技术或生物工程技术与中药干预相结合,进一步提高中药EVs的高效性;另一方面,与中医药科学内涵的阐释密切相关

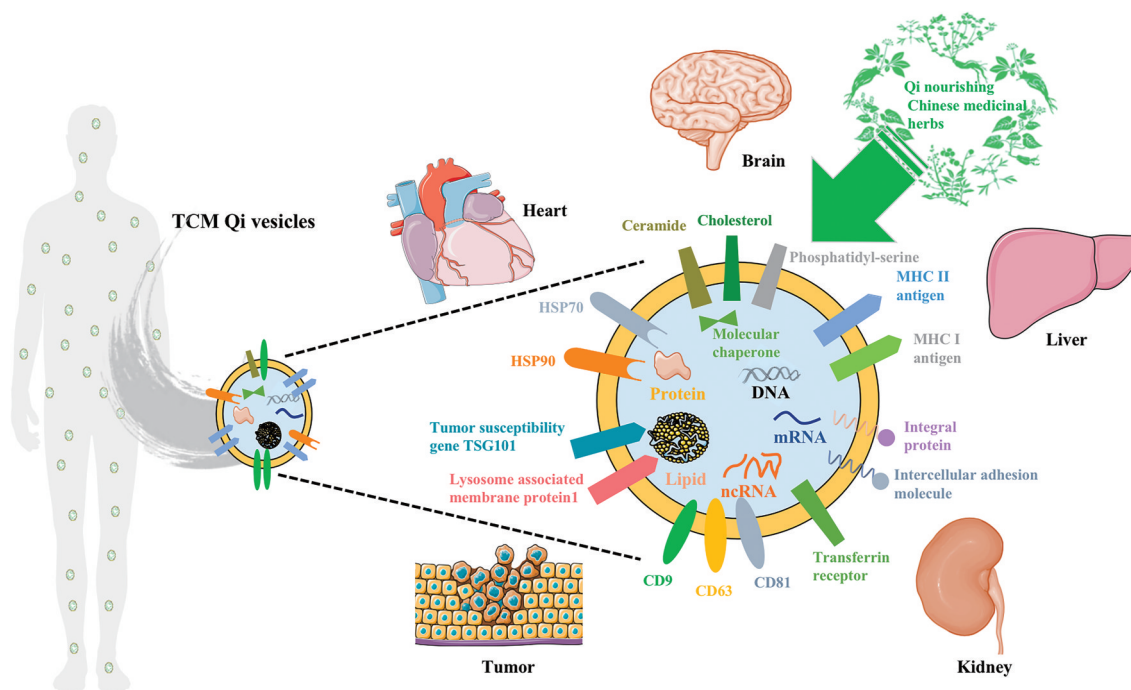


Figure 1 TCM "Qi vesicles" and Qi invigorating TCMs regulate EVs to play a therapeutic role. HSP70/90: Heat shock protein 70/90; TSG101: Tumor susceptibility gene 101 protein; MHC-I/II: Major histocompatibility complex-I/II

关, 中药干预EVs的作用是对中医功效科学内涵的丰富, 同时其进一步的应用拓展与技术提升应以中医理论为指导。

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References

- [1] Wang JB, Xiao XH. On indirect actions of Chinese medicines and innovation of indirect-acting Chinese medicines [J]. *China J Chin Mate Med (中国中药杂志)*, 2021, 46: 5443-5449.
- [2] Gao Y, Wang JB. Commentary: indirect action pattern: a remote and cross-organ pharmacological mechanism for drug innovation [J]. *Acta Pharm Sin B*, 2022, 12: 3448-3450.
- [3] Li LM, Ji YX. Relationship between decline and lack of Qi in traditional Chinese medicine and energy transformation of glycolipid 1 [J]. *Chin Arch Tradit Chin Med (中华中医药学刊)*, 2017, 35: 2865-2867.
- [4] Xi W, Song NN, Liang H, et al. Application of proteomics in the essential study of Qi in traditional Chinese medicine [J]. *China J Tradit Chin Med Pharm (中华中医药杂志)*, 2021, 36: 683-686.
- [5] Du WX, Zhu MD, Zhang F, et al. Theory of TCM gasification and mechanism of TCM healing [J]. *J Tradit Chin Med (中医杂志)*, 2013, 54: 1081-1084.
- [6] Mathieu M, Martin-Jaular L, Lavieu G, et al. Specificities of secretion and uptake of exosomes and other extracellular vesicles for cell-to-cell communication [J]. *Nat Cell Biol*, 2019, 21: 9-17.
- [7] van Niel G, D'Angelo G, Raposo G. Shedding light on the cell biology of extracellular vesicles [J]. *Nat Rev Mol Cell Biol*, 2018, 19: 213-228.
- [8] Urabe F, Kosaka N, Ito K, et al. Extracellular vesicles as biomarkers and therapeutic targets for cancer [J]. *Am J Physiol Cell Physiol*, 2020, 318: C29- C39.
- [9] Zhang Y, Bi JY, Huang JY, et al. Exosome: a review of its classification, isolation techniques, storage, diagnostic and targeted therapy applications [J]. *Int J Nanomedicine*, 2020, 15: 6917-6934.
- [10] Hill AF. Extracellular vesicles and neurodegenerative diseases [J]. *J Neurosci*, 2019, 39: 9269-9273.
- [11] Li YM, Tan XN, Ma RL, et al. A new interpretation of Zangfu related theory in traditional Chinese medicine -- a discussion on the relationship between exosomes and Zangfu related theory [J]. *Hunan J Tradit Chin Med (湖南中医杂志)*, 2017, 33: 1-4.
- [12] Jiao HY, Yan ZW, Liu YY, et al. The application of exosomes in the study of traditional Chinese medicine syndrome [J]. *Chin Med Mod Dist Edu China (中国中医药现代远程教育)*, 2018, 16: 145-148.
- [13] Ji KS, Yang MQ, Chen WN, et al. Relationship between exosome miRNA and TCM theory and research progress on its mechanism of coronary heart disease [J]. *Chin Arch Tradit Chin Med (中华中医药学刊)*, 2021, 39: 48-52.
- [14] Chen DF, Wu YF, Li HY, et al. Exploration of ideas and

- strategies for TCM modernization experimental research based on precise messenger targeting of exosomes and TCM supramolecular "Qi chromatography" theory [J]. *Chin J Exp Tradit Med Form* (中国实验方剂学杂志), 2022, 28: 198-206.
- [15] Shou Y, Ma YH, Hu L, et al. Application and prospect of exosome research in the fields of traditional Chinese medicine [J]. *Acad J Shanghai Univ Tradit Chin Med* (上海中医药大学学报), 2019, 33: 38-43.
- [16] Huang Y, Zhao HJ, Su LJ, et al. Effect of Yiqi Huoxue Huatan Decoction on antiatherosclerosis through macrophage exosome miRNA-let-7-5p/TAB2 signaling pathway [J]. *Chin J Integr Med Cardio/Cerebrovasc Dis* (中西医结合心脑血管病杂志), 2022, 20: 835-840.
- [17] He GX, Wei J, Qin WB, et al. Research progress on improving myocardial microenvironment by regulating PI3K/mTOR signal pathway mediated by exosomes miR-155 of Qishen Yiqi Dropping Pill [J]. *J Liaoning Univ Tradit Chin Med* (辽宁中医药大学学报), 2022, 24: 1-5.
- [18] Ruan XF, Ju CW, Shen Y, et al. Suxiao Jiuxin pill promotes exosome secretion from mouse cardiac mesenchymal stem cells *in vitro* [J]. *Acta Pharmacol Sin*, 2018, 39: 569-578.
- [19] Ruan XF, Li YJ, Ju CY, et al. Exosomes from Suxiao Jiuxin pill-treated cardiac mesenchymal stem cells decrease H3K27 demethylase UTX expression in mouse cardiomyocytes *in vitro* [J]. *Acta Pharmacol Sin*, 2018, 39: 579-586.
- [20] Zhao PY, Ji J, Liu XH, et al. Bu-Shen-Yi-Sui Capsule, an herbal medicine formula, promotes remyelination by modulating the molecular signals *via* exosomes in mice with experimental autoimmune encephalomyelitis [J]. *Oxid Med Cell Longev*, 2020, 2020: 7895293.
- [21] Yang JH, Gao F, Zhang YK, et al. Buyang Huanwu Decoction (BYHWD) enhances angiogenic effect of mesenchymal stem cell by upregulating VEGF expression after focal cerebral ischemia [J]. *J Mol Neurosci*, 2015, 56: 898-906.
- [22] Ye QN, Zhou Y, Zhao CQ, et al. Salidroside inhibits CCl₄-induced liver fibrosis in mice by reducing activation and migration of HSC induced by liver sinusoidal endothelial cell-derived exosomal SphK1 [J]. *Front Pharmacol*, 2021, 12: 677810.
- [23] Ku WC, Sridharan B, Chen JY, et al. Kaempferitrin-treated HepG2 differentially expressed exosomal markers and affect extracellular vesicle sizes in the secretome [J]. *Biomolecules*, 2021, 11: 187.
- [24] Liu XH, Liu SQ, Luo DG, et al. Involvement of circulating exosomal microRNAs in Jian-Pi-Yi-Shen Formula protection against adenine-induced chronic kidney disease [J]. *Front Pharmacol*, 2020, 11: 622658.
- [25] Li HL, Lu RR, Pang Y, et al. Zhen-Wu-Tang protects IgA nephropathy in rats by regulating exosomes to inhibit NF- κ B/NLRP3 pathway [J]. *Front Pharmacol*, 2020, 11: 1080.
- [26] Cui FQ, Wang YF, Gao YB. Effect of Baoshen Tongluo recipe on the proliferation of mesangial extracellular matrix and the expression of miR-192 in exosomes under high glucose culture [J]. *Hebei J Tradit Chin Med* (河北中医), 2020, 42: 253-257, 263.
- [27] Wu XM, Gao YB, Xu LP, et al. Tongxinluo inhibits renal fibrosis in diabetic nephropathy: involvement of the suppression of intercellular transfer of TGF- β 1-containing exosomes from GECs to GMCs [J]. *Am J Chin Med*, 2017, 45: 1075-1092.
- [28] Wang B, Zhang Yu, Chen HF. Mechanisms of Yiqi Compound Recipe on inhibiting exosomes transmitting drug resistance information in triple negative breast cancer cells [J]. *Acad J Shanghai Univ Tradit Chin Med* (上海中医药大学学报), 2018, 32: 50-55.
- [29] Han JY. Scientific connotation of cardiac Qi deficiency and stasis of blood, and the mechanism of Qishen Yiqi Dropping Pills in invigorating Qi and activating blood [J]. *Modern Tradit Chin Med Mater Med World Sci Technol* (世界科学技术-中医药现代化), 2019, 21: 139-147.
- [30] Sahoo S, Adamiak M, Mathiyalagan P, et al. Therapeutic and diagnostic translation of extracellular vesicles in cardiovascular diseases: roadmap to the clinic [J]. *Circulation*, 2021, 143: 1426-1449.
- [31] Wang RN, Wang XH, Zhang YT, et al. Emerging prospects of extracellular vesicles for brain disease theranostics [J]. *J Control Release*, 2022, 341: 844-868.
- [32] Zhao Y, Zhao MF, Jiang S, et al. Liver governs adipose remodelling *via* extracellular vesicles in response to lipid overload [J]. *Nat Commun*, 2020, 11: 719.
- [33] Wang HB, Lu ZM, Zhao XX. Tumorigenesis, diagnosis, and therapeutic potential of exosomes in liver cancer [J]. *J Hematol Oncol*, 2019, 12: 133.
- [34] Zheng J, Lu TY, Zhou CR, et al. Extracellular vesicles derived from human umbilical cord mesenchymal stem cells protect liver ischemia/reperfusion injury by reducing CD154 expression on CD4⁺ T cells *via* CCT2 [J]. *Adv Sci*, 2020, 7: 1903746.
- [35] Cao JY, Wang B, Tang TT, et al. Exosomal miR-125b-5p deriving from mesenchymal stem cells promotes tubular repair by suppression of p53 in ischemic acute kidney injury [J]. *Theranostics*, 2021, 11: 5248-5266.
- [36] He CJ, Zheng S, Luo Y, et al. Exosome theranostics: biology and translational medicine [J]. *Theranostics*, 2018, 8: 237-255.
- [37] Xiao X, Yu SR, Li SC, et al. Exosomes: decreased sensitivity of lung cancer A549 cells to cisplatin [J]. *PLoS One*, 2014, 9: e89534.
- [38] Zhao L, Liu WT, Xiao J, et al. The role of exosomes and "exosomal shuttle microRNA" in tumorigenesis and drug resistance [J]. *Cancer Lett*, 2015, 356: 339-346.
- [39] Jena BC, Mandal M. The emerging roles of exosomes in anti-cancer drug resistance and tumor progression: an insight towards tumor-microenvironment interaction [J]. *Biochim Biophys Acta Rev Cancer*, 2021, 1875: 188488.

- [40] Bayraktar R, Van Roosbroeck K. miR-155 in cancer drug resistance and as target for miRNA-based therapeutics [J]. *Cancer Metastasis Rev*, 2018, 37: 33-44.
- [41] Lv MM, Zhu XY, Chen WX, et al. Exosomes mediate drug resistance transfer in MCF-7 breast cancer cells and a probable mechanism is delivery of P-glycoprotein [J]. *Tumour Biol*, 2014, 35: 10773-10779.
- [42] Hu SF, Gu HP, Chen HF, et al. Professor LU Deming's experience of treating breast cancer by strengthening healthy Qi to eliminate pathogens [J]. *Chin Arch Tradit Chin Med (中华中医药学刊)*, 2013, 31: 2732-2734.
- [43] Liao MJ, Ye MN, Zhou RJ, et al. Yiqi formula enhances the antitumor effects of erlotinib for treatment of triple-negative breast cancer xenografts [J]. *Evid Based Complement Alternat Med*, 2014, 2014: 628712.
- [44] Zhang Yu, Chen HF. Evaluation of reversing effects of Yiqi Formula on cisplatin resistance in triple negative breast cancer based on NF- κ B signaling pathway [J]. *Acad J Shanghai Univ Tradit Chin Med (上海中医药大学学报)*, 2018, 32: 49-54.
- [45] Xu XH, Yuan TJ, Dad HA, et al. Plant exosomes as novel nanoplatforms for microRNA transfer stimulate neural differentiation of stem cells *in vitro* and *in vivo* [J]. *Nano Lett*, 2021, 21: 8151-8159.
- [46] Han X, Wei Q, Lv Y, et al. Ginseng-derived nanoparticles potentiate immune checkpoint antibody efficacy by reprogramming the cold tumor microenvironment [J]. *Mol Ther*, 2022, 30: 327-340.
- [47] Yang FM, Ning ZQ, Ma L, et al. Exosomal miRNAs and miRNA dysregulation in cancer-associated fibroblasts [J]. *Mol Cancer*, 2017, 16: 148.
- [48] Li YJ, Liu RP, Ding MN, et al. Tetramethylpyrazine prevents liver fibrotic injury in mice by targeting hepatocyte-derived and mitochondrial DNA-enriched extracellular vesicles [J]. *Acta Pharmacol Sin*, 2022, 43: 2026-2041.
- [49] Kim MK, Choi YC, Cho SH, et al. The antioxidant effect of small extracellular vesicles derived from *Aloe vera* Peels for wound healing [J]. *Tissue Eng Regen Med*, 2021, 18: 561-571.