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不同干预因素建立小鼠动脉粥样硬化模型的研究进展

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【摘要】 动脉粥样硬化(atherosclerosis,AS)是常见心脑血管疾病的潜在危险因素,严重威胁着人类的健康和生命。AS 的防治和抗 AS 新药的研发是医药学界一直关注的热点。选择理想的 AS 动物模型是研究 AS 防治措施及新药研发的关键。理想的 AS 动物模型应是与人类 AS 发病机理相似,病理及生化反应一致,且 AS 模型复制的重复性高、操作简便、易于推广。小鼠具有繁殖性强、遗传性高、可塑性好、AS 成模性快等特点,成为了 AS 研究的理想动物。本文就不同干预因素建立小鼠 AS 模型的复制方法进行了比较分析,以期为开展 AS 防治和中药新药研发提供参考和思路。

【关键词】 动脉粥样硬化;干预因素;小鼠;动物模型
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Research progress on establishment of a mouse atherosclerosis model by various intervention factors

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【Abstract】 Atherosclerosis (AS) is a potential risk factor for common cardiovascular and cerebrovascular diseases. Prevention and treatment of AS and research and development of anti-AS drugs have been a focus in the field of medicine. Selection of the ideal AS animal model is important to study prevention and control measures of AS and to research and develop traditional Chinese medicine for AS. The ideal animal model of AS should have similar pathogenesis to human AS, consistent pathological and biochemical reactions, high repeatability, simple operation, and easy adoption. Mice have the characteristics of strong reproduction, high heritability, good plasticity, and fast modeling of AS, and they have become ideal animals for AS research. In this article, the replication method of the mouse AS model established by various

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intervention factors were compared and analyzed to provide a reference and ideas for prevention and treatment of AS and research and development of traditional Chinese medicine for AS.

【Keywords】 atherosclerosis; intervention factors; mice; animal model

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动脉粥样硬化(atherosclerosis, AS)是心脑血管疾病的潜在危险因素,是全球死亡和发病的主要原因[1]。AS 被认为是由多种因素引起的,包括遗传和环境因素,其特征是动脉腔硬化和狭窄,因在动脉内壁上形成了称为斑块的脂肪沉积物[2]。人类 AS 的起始机制虽未被完全阐明,但从 AS 病变标本和动物模型角度来看,AS 已被定义为一种慢性炎症性疾病,涉及内皮细胞功能障碍、脂质浸润、巨噬细胞募集和血管平滑肌细胞迁移[3]。

研究表明,高水平的血浆低密度脂蛋白(low density lipoprotein, LDL-C)是 AS 最主要的危险因素之一[4]。LDL-C 倾向于积聚在动脉壁的内皮下空间,通过逐步氧化过程,产生最小修饰的低密度脂蛋白(minimally modified low density lipoprotein, mm-LDL),其中只有脂质成分被氧化,然后是完全氧化的低密度脂蛋白(oxidized low density lipoprotein, ox-LDL)[5]。ox-LDL 通过氧化低密度脂蛋白受体-1(oxidized low-density lipoprotein receptor-1, LOX-1)作用于内皮细胞,从而诱发内皮功能障碍,细胞表面粘附分子过表达,包括血管细胞粘附分子 1(vascular cell adhesion molecule 1, VCAM-1)、细胞内粘附分子 1(intracellular adhesion molecule 1, ICAM-1)和内皮白细胞粘附分子(endothelial leukocyte adhesion molecule, E-selectin),最终诱导动脉壁的炎症反应[6]。炎症反应促使循环单核细胞附着在受损的内皮上,将单核细胞迁移到脉管系统内皮下基质下方的内膜层,浸润的单核细胞进一步分化为巨噬细胞,巨噬细胞通过清道夫受体识别和摄取 ox-LDL 形成泡沫细胞,这种泡沫细胞内皮下的积聚也被称为“脂肪条纹形成”,这就是 AS 形成的早期阶段[7-8]。炎症反应还能促进循环单核细胞和 T 细胞的募集,刺激血管平滑肌细胞(vascular smooth muscle cell, VSMC)从血管壁内侧层迁移到内膜层,表现出异常高的增殖并分泌细胞外基质蛋白,在 AS 晚期形成纤维帽以稳定脆弱的斑块[9-10]。在 AS 晚期,随着斑块的进化,泡沫细胞和 VSMC 发生死亡,细胞外脂质从垂死或死细胞的碎片中积聚并积累脂蛋白,在许多斑块中形成脂质核心。纤维帽通常在脂质核心上方形成,由于合成减少和细胞外基质大分子(如胶原蛋白)的分解增加,使得这种纤维帽变薄,形成薄帽 AS[11]。这种高风险脆弱斑块的纤维帽一旦破裂,叠加的血栓形成可能会阻塞动脉腔并破坏血流,最终导致心肌梗死、中风或猝死,这也是 AS 发病率和死亡率最高的原因之一[12]。

AS 动物模型是提高对 AS 斑块形成与斑块破裂及其相关心血管事件发生认识的重要工具。因此,选择合适的 AS 动物模型至关重要。理想的 AS 动物模型应尽可能与人类临床发病机理相似,并可用于医学与药物研究,且成本合理、重复性高。小鼠是 AS 研究中使用最广泛的动物模型,具有繁殖性强、遗传性高、转基因的可塑性好、以及 AS 病变部位形成快等优势[13]。本文简要介绍饮食、转基因、新技术、遗传和环境等不同干预因素对小鼠 AS 模型的研究进展并进行比较分析,以期为 AS 的基础研究提供新的参考和思路。

1　饮食与小鼠 AS 模型

AS 是一种慢性的炎症过程,饮食对高脂血症促进 AS 斑块形成的影响颇大[14]。长期高胆固醇饮食会引起小鼠血清胆固醇(total cholesterol, TC)水平升高,其与主动脉病变的不断加剧呈正相关[15]。自 20 世纪 90 年代以来,21%脂肪结合 0.15%胆固醇的西方饮食是 AS 动物模型常用的饲料配方[16]。

早在 20 世纪 60 年代末,学者们就开始探究小鼠 AS 模型建立的方法。Vesselinovitch 等[17]通过高脂饮食、辐射诱导 CF1 小鼠 AS 病变。但是,在高脂饮食和辐射及环境因素的共同作用下,使得这项研究个体差异极大,甚至伴随着较高死亡率。随后,Thompson[18]率先采用高胆固醇高脂肪(high-fat/high-cholesterol, HFHC)的饲料喂养近交系小鼠(C57BL/6)诱导 AS,25 周后,小鼠的存活率高达 100%,并且发现 AS 模型小鼠泡沫细胞普遍出现在主动脉瓣区域。从此,近交系小鼠逐渐成为 AS 模型的研究基础[19]。1985 年,Paigen 等[20]以(15%脂肪 + 1.25%胆固醇 + 0.5%胆酸)饮食诱导 10 种近交系 C57BL/6,结果发现小鼠 14 周后就形成高胆固醇血症,且主动脉瓣大量细胞内脂肪沉积,并含有大量泡沫细胞,同时病变已开始侵入下层介质形成

脂肪条纹。以半合成高脂肪饮食诱导 9 种近交系小鼠 18 周,同样发现 C57BL/6 是最容易发生 AS 病变的小鼠[21]。可见,C57BL/6 小鼠是 AS 模型基础研究的常用研究对象。

2 基因敲除小鼠 AS 模型

2.1 经典 ApoE$^{-/-}$、LDLR$^{-/-}$AS 模型

载脂蛋白 E(ApoE)是一种双域抗 AS 脂质转运蛋白,作为低密度脂蛋白(low density lipoprotein, LDL)受体的配体,可以介导含载脂蛋白 B(apolipoprotein B, ApoB)的致 AS 脂蛋白的清除,包括极低密度脂蛋白(very low density lipoprotein, VLDL)、中密度脂蛋白(intermediate-density lipoprotein, IDL)的受体,从而降低血浆脂质水平[22-23]。Plump 等[24]基于 C57BJ/6 菌株制备了 ApoE$^{-/-}$小鼠模型。该模型身体健康,在外形上与 C57BJ/6 小鼠并无差异。采用普通饮食分别喂养 ApoE$^{-/-}$和 C57BJ/6 小鼠 16 周后,ApoE$^{-/-}$小鼠的 LDL、TC 水平较 C57BJ/6 小鼠升高数倍,同时 ApoE$^{-/-}$小鼠主动脉血管伴随脂质沉积[25]。这表明在没有环境刺激的情况下,ApoE 的缺乏能引起脂蛋白代谢的巨大变化。研究发现,ApoE$^{-/-}$小鼠在正常饮食下,10 周后出现了动脉僵硬,15 周后 TC 水平提高了 50%,20 周后斑块出现在胸主动脉及其主要的分叉节点处;而在西方饮食下则会加快 ApoE$^{-/-}$小鼠 AS 病变的进程,如大量胶原沉积、主动脉僵硬程度成倍增加、钙化严重,40 周后主动脉完全受损[26-27]。

低密度脂蛋白受体敲除(LDLR$^{-/-}$)小鼠是另一种常用的 AS 研究的动物模型。LDLR 是一种膜受体,可介导含有载脂蛋白 ApoB100 或 ApoE 的血浆脂蛋白的肝清除率,从而维持血浆 LDL 的水平[28]。1994 年,Ishibashi 等[29]率先建立了 LDLR$^{-/-}$小鼠模型,与 ApoE$^{-/-}$小鼠不同,LDLR$^{-/-}$小鼠不会在普通饮食的基础上发生 AS 病变,但 TC 水平通常保持在 250 mg/dl 左右[30-31]。而采用西方饮食(21%的脂肪、0.15%的胆固醇)喂养 LDLR$^{-/-}$小鼠 12 周后,小鼠血浆 TC、甘油三脂(triglyceride, TG)、LDL-C 的水平远远高于普通饮食喂养的 LDLR$^{-/-}$小鼠,并且其主动脉根部有大量脂质沉积[32]。

2.2 复杂 ApoE$^{-/-}$、LDLR$^{-/-}$AS 病变模型

研究表明,采用高脂饲料喂养 ApoE$^{-/-}$与 LDLR$^{-/-}$双敲除小鼠(dKO),结果发现小鼠溶栓活性因素减少,增加了 AS 的进展;采用普通饲料喂养 ApoE$^{-/-}$与 LDLR$^{-/-}$受体双敲除小鼠,其 AS 的形成或病变比单敲除 ApoE$^{-/-}$小鼠更明显,说明 ApoE$^{-/-}$与 LDLR$^{-/-}$受体双敲除小鼠比 ApoE$^{-/-}$单敲除小鼠更易形成 AS[33-34]。亦有学者研究发现,IL-19/LDLR dKO 小鼠和 LDLR$^{-/-}$小鼠高脂饲料共同喂养 14 周后,IL-19/LDLR dKO 小鼠巨噬细胞和 VSMC 的炎症因子表达增加,油红 O 染色发现 IL-19/LDLR dKO 小鼠主动脉根部病变的面积是 LDLR$^{-/-}$小鼠的 2 倍,这表明在 LDLR$^{-/-}$的基础上缺乏 IL-19 更易形成或加重 AS 的病变[35]。ApoE$^{-/-}$小鼠与含有原纤维蛋白-(Fbn1)基因的突变(C1039G+/-)的小鼠杂交,成为 ApoEFbn1$^{-/-C1039G+/-}$小鼠,其中原纤维蛋白-1 的 Fbn1 基因突变导致马凡综合征(MFS),突变的 FBN1 蛋白可能导致弹性结构不稳定,从而扰乱 TGF-β 信号通路,导致主动脉表型受累更严重,主动脉根部直径更大,扩张速度更快,死亡和夹层风险往往增加[36-39],同时加剧了 ApoE$^{-/-}$小鼠 AS 高度不稳定斑块表型形成。在西方饮食的影响下,ApoEFbn1$^{-/-C1039G+/-}$小鼠核心扩大,纤维帽薄,胶原纤维大量损失,在头臂动脉、升主动脉均发生了斑块破裂,表现出临床终末期 AS 患者的许多特征。随着西方饮食的饲养周期延长,ApoEFbn1$^{-/-C1039G+/-}$小鼠不可避免地出现了 1/5 的死亡[39]。Wang 等[40]研究发现,将 Fbn1$^{-/-C1039G+/-}$小鼠与 LDLR$^{-/-}$小鼠杂交以获得 AS 易感斑块的新模型,相较于 ApoEFbn1$^{-/-C1039G+/-}$小鼠在整个高脂肪饮食喂养期间,所有小鼠都处于良好状态,并且没有记录到神经系统并发症或急性心血管事件,避免了在 ApoE$^{-/-}$小鼠中观察到的高死亡率问题。但是 LDLRFbn1$^{-/-C1039G+/-}$实验研究仅使用少量小鼠用于斑块染色实验,缺乏有力的证据。

2.3 SR-BI$^{-/-}$AS 模型

清道夫受体 B 类Ⅰ型(SR-BI/SCARB1)最初被发现是一种清除蛋白,可与一系列天然和修饰的脂蛋白结合,同时 SR-BⅠ是一种高亲和力高密度脂蛋白(high density lipoprotein, HDL)生理膜蛋白受体,其通过肝细胞介导 HDL 颗粒中胆固醇酯(cholesteryl ester, CEs)的选择性摄取[41]。SR-BⅠ是促进细胞和 HDL 之间游离胆固醇(free cholesterol, FC)的双向通量[42]。除肝外,SR-BⅠ在巨噬细胞和内皮细胞中大量表达[43]。研究表明,SR-BⅠ和 ATP 结合盒转运蛋白 A1(ATP binding

cassette transporter A1, ABCA1) 负责胆固醇从单核细胞衍生巨噬细胞向 HDL 外排[44]。采用高胆固醇饲料喂养 SR-B Ⅰ敲除(SR-B Ⅰ$^{-/-}$)小鼠,结果发现小鼠血浆 VLDL 大小范围内富含 FC 和 ApoE 的 HDL 颗粒显著积累,血浆胆固醇接近 1000 mg/dl,促进了 AS 的发展[45]。因此,SR-B Ⅰ$^{-/-}$高脂血症背景的小鼠能促进 AS 的形成,但其更容易导致冠状动脉闭塞、心肌梗死甚至死亡[46]。

2.4　载脂蛋白(Apo)E3-Leiden AS 模型

突变的 ApoE 亚型 ApoE3-Leiden 与人类显性遗传形式的 β-脂蛋白血症有关,过表达 ApoE3-Leiden 的转基因小鼠极易发生饮食诱导的高脂血症和 AS,还能表现出从胸主动脉和腹主动脉的早期脂肪条纹到主动脉弓的晚期病变[47-50]。因此,ApoE3-Leiden 转基因小鼠也可以用作 AS 的模型。转基因 ApoE3-Leiden.CETP 小鼠是人类脂蛋白代谢的成熟模型,具有人源化的脂蛋白代谢和对 AS 病变的异质反应,含 ApoB 脂蛋白中的胆固醇更多,HDL 胆固醇水平相对较低,类似于人类,能导致肝对循环中富含甘油三酯的脂蛋白残留物的摄取减少[49]。与 ApoE3-Leiden 小鼠相比,高脂饲料喂养 19 周后,ApoE3-Leiden.CETP 小鼠体内 VLDL 和 LDL 水平明显升高,主动脉根部 AS 病变面积增加,是评估 AS 药物治疗的成熟模型[50]。

2.5　一种 MicroRNA AS 模型的新探索

MicroRNA(miRs)是一类短的非编码 RNA,与 RNA 诱导的沉默复合物(RNA-induced silencing complex, RISC)结合,并与靶 mRNA 的互补序列(通常在 3'UTR 中)结合,通过抑制转录和转录后编码基因的表达来影响许多生物学途径,许多 miRNA 已被证明参与生物途径并影响发病机制[51]。miRNA 参与调节脂质代谢、炎症反应、细胞周期、氧化应激、血小板活化、内皮及 VSMC 功能,从而影响 AS 的发生与发展[52]。研究表明,miR-33 在体内的拮抗作用会增加循环 HDL 和 RCT,从而抑制 AS 的发生与发展,并增强其消退[53]。采用高脂饲料喂养 C57BL/6 小鼠和 miR-33(miR-33$^{-/-}$)敲除小鼠 12 周,结果发现 miR-33$^{-/-}$小鼠体内 TG 水平和肝中的脂质积累增多,引发了中度肝脂肪变性和高甘油三酯血症[54]。此外,miR-27b、miR-144 也代表了 miRNA 作为胆固醇稳态的关键介质,其可能作为 AS 的潜在生物标志物[55]。研究显示高脂肪饮食诱导 miR-144 基因敲除小鼠(miR-144$^{-/-}$)的 AS 病变是正常小鼠的 1.5 倍[56]。

3　新技术 AAV-PCSK9 的 AS 模型

作为脂蛋白代谢的关键调节因子,蛋白转化酶枯草溶菌素 9(proprotein convertase subtilisin kexin 9, PCSK9)可以增强肝 LDL 受体的降解,从而导致 LDL 水平升高[57]。腺相关病毒(adeno-associated virus, AAV)载体可以有效地、特异性地将转基因递送到肝,且能够持续表达[58]。AAV-PCSK9 小鼠是 10 年前开发的一种无种系基因工程的致 AS 小鼠模型新品系[59],高脂喂养同时注射 AAV-PCSK9 的 C57BL/6N 小鼠 3 个月,显示 TC 浓度升高至 1000 mg/dl 以上,远远高于普通饮食组小鼠 TC 浓度,油红 O 染色结果显示接受 AAV-PCSK9 注射的小鼠主动脉根部斑块和脂肪明显增加;同时确定,在西方饮食的基础上注射 AAV-PCSK9 病毒载量为 1.0×10^{11} VG 是诱发 AS 病变的最佳剂量[60]。与 ApoE$^{-/-}$或 LDLR$^{-/-}$小鼠的常规杂交相比,单次 AAV 注射足以在更快的时间内产生新的 AS 小鼠模型。

4　遗传和环境因素

4.1　母体遗传导致后代 AS 病变发展

长期以来,人们一直认为胎儿发育期间的致病事件会影响 AS 相关的疾病[61]。研究发现,不论在人类还是动物体内,AS 病变在胎儿期间开始形成,并且这一病变过程因母亲高胆固醇血症而加速[62]。通过以 C57BL/6J 雌性小鼠为研究对象,在其整个交配、妊娠期与哺乳期之间,补充外源性胆固醇,观察其雄性后代,结果发现雄性后代血清 VLDL 颗粒数量与大小均增加,肝脂谱的早期变化(单不饱和脂肪酸浓度较低,多不饱和脂肪酸浓度较高),并持续到成年期[63]。研究人员给 8 周龄 ApoE$^{-/-}$雌鼠在妊娠与哺乳期间喂养高脂饲料,6 个月后其生产的雄性后代中,整个主动脉 AS 病变是普通饲料喂养的雌鼠组的 2 ~ 3 倍[64]。

4.2　慢性应激加速 AS 的发展

慢性应激是指当身体长时间受到各种内部和外部的负面因素刺激下发生的非特异性全身反应。长期以来,在慢性应激下暴露的生理反应一直被认为与 AS 发生有着密切联系[65]。研究表明,慢性应激能增加血管疾病的发展及冠状动脉疾病的发病率和死亡率,其机制可能是由于慢性应激引起内皮损伤,直接激活巨噬细胞,促进泡沫细胞形成并产

生 AS 斑块[66]。Figueiro 等[67]给予 LDLR$^{-/-}$小鼠高脂饮食的同时,每周连续 4 d 的逆转明暗、3 d 的常规明暗(rotating shift schedules, RSS)模式,持续 11 周,结果发现 RSS 模式下的小鼠出现较大的 AS 病变,同时,原纤维胶原减少,并增加了内质网应激、细胞凋亡和病变内的坏死区域,加速了 LDLR$^{-/-}$小鼠 AS 的发展。此外,ApoE$^{-/-}$在轻度慢性应激暴露 12 周后,与未暴露 ApoE$^{-/-}$小鼠相比,暴露的 ApoE$^{-/-}$小鼠的 AS 斑块面积增加了一倍[68]。说明慢性应激和高脂饮食复合因素对基因编辑小鼠 AS 病变发展具有很强的协同作用。

5 结语

综上,实验性 AS 小鼠模型复制方法种类较多,且不同干预因素建立小鼠 AS 模型各有优劣(表 1)。ApoE$^{-/-}$小鼠可以自发性的产生 AS 病变,是建立 AS 的经典模型。LDLR 受体的缺失更加符合高脂血症诱导的 AS,但其需要高脂喂养或物理操作才能较好地实现 AS 病变。ApoE3-Leiden. CETP 小鼠克服了经典小鼠模型中非人类脂蛋白代谢的缺陷,但仍需要高脂喂养。SR-BI$^{-/-}$、ApoE/LDLR dKO、IL-19/LDLR dKO 小鼠 TC 水平升高,加速 AS 斑块形成,是实现 AS 快速造模的方法,但易造成严重管腔闭塞,导致过早死亡。而 ApoEFbn1$^{-/-C1039G+/-}$、LDLRFbn1$^{-/-C1039G+/-}$小鼠模型能够自发性 AS 斑块破裂,给晚期病变和自发性斑块破裂在经典小鼠中的发生提供了可用模型,但复杂病变的证据较少,尚待进一步深入研究。MicroRNA 敲除小鼠能加快 AS 病变

表 1 不同 AS 模型小鼠的优势与弊端

Table 1 Advantages and disadvantages of each type of atherosclerosis mice

干预因素 Intervention factors	优势 Advantage	弊端 Disadvantage
ApoE$^{-/-}$小鼠 ApoE$^{-/-}$ mice	无需高脂饲养自发性形成 AS 病变,高脂饲养下加重 No need for high-fat feeding to spontaneously form AS lesions, which worsens under high-fat feeding	ApoE 的多重功能、不能自发性形成斑块破裂和心肌梗死[69] ApoE multiple functions, inability to spontaneously form plaque rupture, and myocardial infarction[69]
LDLR$^{-/-}$小鼠 LDLR$^{-/-}$ mice	更加符合人类的脂质谱,LDL 受体的缺失对炎症没有影响 More in line with the human lipid profile, the absence of LDL receptors has no effect on inflammation	病变过程需要高脂喂养或物理干预 Pathological process requires high-fat feeding or physical intervention
dKO(ApoE$^{-/-}$、LDLR$^{-/-}$、IL-19$^{-/-}$、SR-B1$^{-/-}$小鼠) dKO(ApoE$^{-/-}$、LDLR$^{-/-}$、IL-19$^{-/-}$、SR-B1$^{-/-}$ mice)	AS 的严重程度增加,能自发性形成斑块破裂和心肌梗死 Severity of AS increases and can spontaneously form plaque rupture and myocardial infarction	易发生严重管腔闭塞,导致过早死亡[70] Prone to severe lumen occlusion, leading to premature death[70]
AAV-PCSK9 小鼠 AAV-PCSK9 mice	无需基因操作,经济性高 No genetic manipulation required, high economy	病变需要饮食干预,可能存在抗病毒宿主免疫反应[71] Pathological changes require dietary intervention, and there may be an antiviral host immune response[71]
ApoE3-Leiden 小鼠 ApoE3-Leiden mice	CETP 表达脂质代谢更加贴近“人源化” CETP expression of lipid metabolism is closer to “humanization”	病变过程需要高脂喂养,并且缺乏复杂病变的证据 Pathological process requires high-fat feeding and lacks evidence of complex lesions
MicroRNA 小鼠 MicroRNA mice	更加符合 AS 病变的过程 More in line with the process of AS lesions	缺乏复杂病变的证据[72] Lack of evidence for complex lesions[72]
SR-BI$^{-/-}$小鼠 SR-BI$^{-/-}$ mice	加速 AS 的形成 Accelerate the formation of AS	存活率低 Low survival rate
ApoEFbn1$^{-/-C1039G+/-}$小鼠 ApoEFbn1$^{-/-C1039G+/-}$ mice	自发性形成斑块破裂和心肌梗死 Spontaneous formation of plaque rupture and myocardial infarction	随着饲养周期延长,易出现死亡 As the feeding cycle prolongs, death is prone to occur
LDLRFbn1$^{-/-C1039G+/-}$小鼠 LDLRFbn1$^{-/-C1039G+/-}$ mice	自发性形成斑块破裂和心肌梗死 Spontaneous formation of plaque rupture and myocardial infarction	缺乏有力的证据 Lack of strong evidence
母体遗传后代至 AS 小鼠 Maternal inheritance of offspring to AS mice	模拟胎儿发育期间的致病过程,后代病变率高 Simulate the pathogenic process during fetal development, resulting in a high incidence of offspring lesions	操作复杂,不确定因素多 Complex operation with many uncertain factors
慢性应激小鼠 Chronic stress mice	符合当下人们的工作方式 In line with the current way people work	单独使用压力或高脂喂养无法成功诱导 AS 病变 Use of pressure or high-fat feeding alone cannot successfully induce AS lesions

的进程,但目前并未见到 MicroRNA 在小鼠体内动态影响 AS 病变的报道,仍需进一步研究。除基因缺失,以 AAV 为载体注射 PCSK9,导致体内 LDL 积聚,从而建立 AS 小鼠模型,该模型操作简便,但可能存在抗病毒宿主免疫反应。从现有的小鼠 AS 模型来看,用于评价或筛选防治 AS 中药新药研发的模型动物是经典的 ApoE$^{-/-}$小鼠,同时结合高脂饲料长期喂养,则更易形成 AS 斑块破裂或心肌梗死。

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