·综 述·

动物模型在细菌生物被膜研究中的应用与展望

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摘 要：细菌生物被膜的形成与其致病性、耐药性密切相关，在许多由细菌导致的慢性、亚慢性感染中发挥着重要作用。动物模型广泛应用于细菌生物被膜相关感染的研究中，为其致病机理和控制策略的探究提供了强有力的科学工具。因此，本文系统阐述了哺乳类 (鼠、兔、猪等) 和非哺乳类 (黑腹果蝇、斑马鱼、秀丽隐杆线虫等) 动物模型在细菌生物被膜相关研究中的应用，并对动物模型在细菌生物被膜研究中的应用前景进行了展望，以期为研究由生物被膜导致的相关感染而选择理想动物模型提供理论支撑，从而对生物被膜感染导致的潜在危害进行防控。

关键词：细菌；生物被膜；致病机理；动物模型

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Animal models in bacterial biofilm research: a review

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Abstract: Biofilm formation is closely related to pathogenicity and antibiotic resistance of bacteria, and plays important roles in a number of chronic and subchronic infections. Animal models are widely used in the research of bacterial biofilm-associated infections, and provide a powerful scientific tool for investigating its pathogenesis and control strategies. This review summarized the application of mammalian models (e.g. mouse, rabbit, and pig) and non-mammalian models (e.g. *Drosophila melanogaster*, *Zebrafish*, and *Caenorhabditis elegans*) in bacterial biofilm studies, and prospects the application of animal models in biofilm. This review may facilitate the selection of suitable animal models in the study of biofilm-associated infections, so as to prevent and control the potential adverse effects.

Keywords: bacteria; biofilm; pathogenic mechanism; animal models

生物被膜 (bacterial biofilm, BF) 是指附着在生物或非生物表面、由微生物及其分泌物组成的复杂微生物群落，自然界中有高达 90% 以上的细菌以生物被膜的形式进行生存和繁殖[1-2]。数据表明，约 80%的微生物感染与生物被膜有关[3]，20 世纪 80 年代，由加拿大微生物学家 John Costerton 将此概念引入医学微生物学领域[4]，此后关于生物被膜的性质、产生机理及潜在根除方法被大量研究[5]。与浮游生物相比，被膜态细菌表现出不同的生存性状和代谢类型，显著增强对宿主免疫防御机制和抗生素的耐受性[6-7]，从而导致超级细菌、无药可用等严峻的卫生安全问题，对人类健康造成巨大威胁[8-10]。

实验动物模型是指研究人员利用生物、化学或物理等致病因子，作用于小鼠、兔子、斑马鱼、秀丽隐杆线虫等动物，从而构建具有组织病变、器官损伤、免疫应激等人类疾病模拟表现的动物实验对象[11]。近年来，动物模型已经成为研究生物被膜的重要工具，可用于探究生物被膜在微生物感染中的致病与耐受机制，并有助于评价生物被膜清除及治疗方案的安全性与有效性[12-13]。例如笔者所在实验室已系统总结体内、体外胃肠道模型在食源性致病菌研究应用中的优势及缺陷，与体外模型相比，动物模型不仅可对致病菌的耐受性及致病力进行研究，还可进一步研究其致病机理及疫苗的开发[14]，具有十分重要的科研价值和实际意义，但至今为止，关于此类研究的系统综述尚未开展。

因此，本文系统地总结了近年来用于生物被膜研究的哺乳类 (鼠、兔、猪等) 和非哺乳类 (黑腹果蝇、斑马鱼、秀丽隐杆线虫等) 动物模型 (表 1)，重点概述了其在细菌生物被膜

表 1 生物被膜相关感染动物模型汇总

Table 1 Summary of animal models used for biofilm-related infections

Animal	Infected tissues or organs	Microorganisms	Type of biofilm-related infection	References
Mice	Eyes	*Staphylococcus aureus*	Endophthalmitis	[15]
	Eyes	*Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Fusarium falciforme*	Keratitis	[16-17]
	Lungs	*Staphylococcus aureus*, *Haemophilus influenza*, *Pseudomonas aeruginosa*	Cystic fibrosis, obstructive pulmonary emphysema, bronchitis	[18-21]
	Bladder	*Escherichia coli*, *Klebsiella pneumoniae*	Glandular cyslitis	[22-24]
	Prostatitis	*Proteus mirabilis*, *Escherichia coli*	Chronic bacterial prostatitis	[25-27]
	Vaginiti	*Candida albicans*, *Gardnerella vaginalis*	Vaginitis	[28-29]
	Skull	*Streptococcus suis*	Meningitis	[30-31]
	Skin	*Pseudomonas aeruginosa*, *Escherichia coli*, *Acinetobacter baumannii*, *Klebsiella pneumoniae*, *Enterobacter cloacae*	Chronic abscess infections	[32]
	Cochlearimplant	*Streptococcus pneumoniae*	Chronic otitis media	[33]
	Optimus Neuro System	*Staphylococcus aureus*	Implant-associated infections	[34]
	Central nerve duct	*Staphylococcus aureus*	Implant-associated infections	[35]
Rabbit	Skin	*Pseudomonas aeruginosa*	Chronic wounds infection	[36]
	Cardiac catheter	*Staphylococcus aureus*	Infective endocarditis	[37]
	Cavum nasi	*Staphylococcus aureus*, *Pseudomonas aeruginosa*	Chronic rhinosinusitis	[38-40]
	Bone marrow	*Staphylococcus aureus*	Osteomyelitis	[41-44]
	Urinary tract catheters	*Pseudomonas aeruginosa*, *Proteus mirabilis*	Urinary tract infections	[45]
	Arthriti	*Staphylococcus aureus*, *Escherichia coli*	Arthritis	[46-47]
	Spinal implant	*Staphylococcus aureus*	Implant-associated Infections	[48]
Pig	Tracheal catheter	Methicillin-resistant *Staphylococcus aureus*	Pneumonia	[49]
	Aortic implant	*Staphylococcus aureus*	Implant-associated Infections	[50]
	Urinary tract catheters	*Pseudomonas aeruginosa*	Urinary tract infections	[51]
Caenorhabditis elegans	Skin	*Candida albicans*, *Pseudomonas aeruginosa*, *Vibrio parahemolyticus*, *Yersinia pestis*	Chronic wounds infection	[52-55]
Danio rerio	Embryo	*Salmonella*, *Vibrio parahemolyticus*, *Candida albicans*		[56-59]
Drosophila melanogaster	Stomach, intestinal tract, esophagus	*Pseudomonas aeruginosa*		[60-62]

慢性感染研究中的应用，并对动物模型在生物被膜研究中的未来研究方向进行了展望，以期为生物被膜相关性感染研究中适合动物模型的选择提供理论支撑，从而针对生物被膜感染期间的宿主与致病菌相互作用开发新的防控策略。

1 哺乳类动物模型

动物模型用于生物被膜研究的最早例子可追溯到 20 世纪 40 年代[63]，随着科学技术的发展，越来越多的动物模型可应用于生物被膜相关的组织感染、设备感染和系统感染等[64-65]。在这些动物模型中，鼠 (小鼠、大鼠)、兔子和猪等哺乳动物模型的应用最为普遍，因为这些动物可实现与人体结构相似的解剖、愈合和免疫反应等过程，呈现具有代表性的病理反应[66]，有助于更好地揭示生物被膜相关感染的作用机理。

1.1 鼠类动物感染模型

鼠类是科学研究中重要的模式动物，与人类基因组具有较高同源性[67]，如图 1 所示，被广泛应用于耳部、眼部、肺部、伤口等部位及相关植入物的被膜态细菌感染。

1.1.1 鼠类动物眼、耳感染模型

被膜态的铜绿假单胞菌[68-69]、金黄色葡萄球菌[70]等细菌常感染人类的眼、耳等部位，引起严重的中耳炎、结膜炎等慢性疾病，鼠类模型是研究此类被膜态慢性感染的重要技术手段。Yadav 等在大鼠中耳腔接种了耐甲氧西林金黄色葡萄球菌、铜绿假单胞菌或二者混合液，从而构建了中耳炎模型，结果表明，金黄色葡萄球菌和铜绿假单胞菌可在小鼠中耳部位有效定殖，以生物被膜形式共存，引发了大量的炎

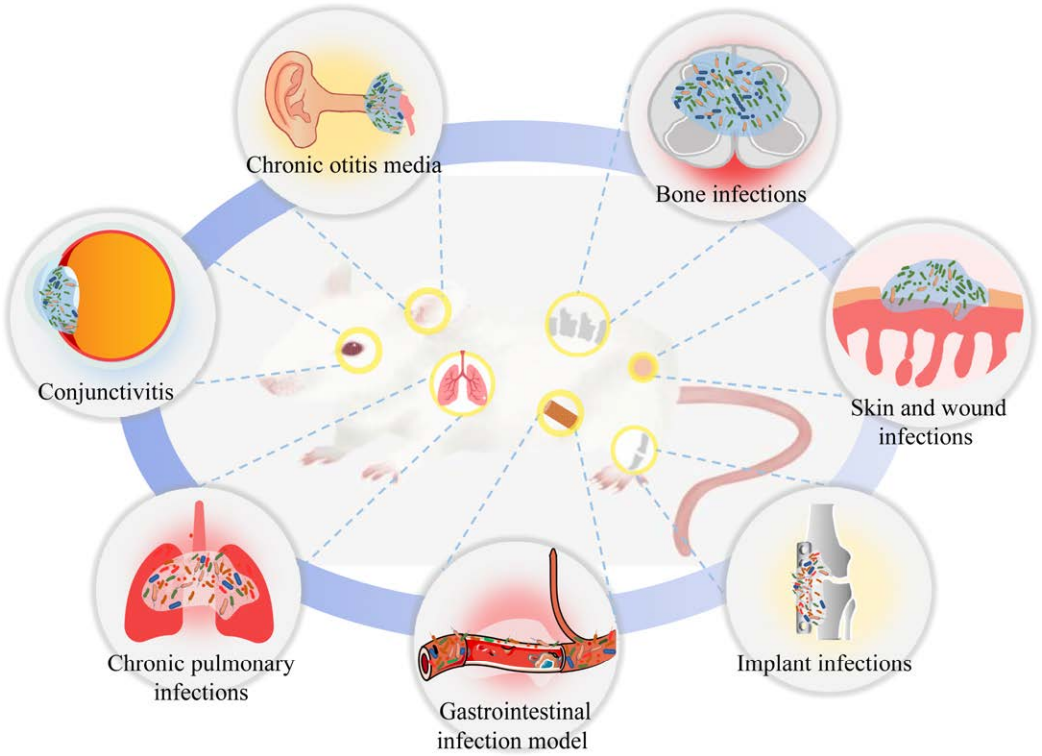

图 1 生物被膜感染的鼠类动物模型应用场景

Figure 1 Application of biofilm-infected rodent animal models.

症及免疫反应[71]。Saraswathi 和 Beuerman 通过小鼠眼角膜的浅层擦伤处理，在伤口处接种铜绿假单胞菌构建了角膜炎模型，并使用显微镜跟踪了眼角膜伤口处铜绿假单胞菌生物被膜的形成过程，透射电镜结果表明，在接种第 5 天后，小鼠角膜表面能够形成成熟的铜绿假单胞菌生物被膜，该模型的构建有助于被膜态细菌性角膜炎的研究[16]。

1.1.2 鼠类动物慢性肺部感染模型

囊性纤维化 (cystic fibrosi, CF) 是一种可侵犯多脏器的遗传性疾病，临床表现多样，并伴有不良的治疗结果，可导致胰腺功能衰竭、黏膜分泌物改变和肺部感染等[72]。以往数据表明，由于缺乏合适的慢性感染的动物模型，需要对囊性纤维化发病机制和宿主免疫反应进行研究。铜绿假单胞菌生物被膜可将多形核中性粒细胞 (polymorphonuclear leukocyte, PMN) 包围，从而加剧肺部感染，使得该疾病呈现较高的发病率和死亡率[73]。因此，Hoffmann 等从临床肺部粘液样本中分离了一株被膜态铜绿假单胞菌 NH57388A，能够稳定表达群体感应 (quorum sensing, QS) 基因，进而构建了一个稳定的被膜态铜绿假单胞菌慢性感染 CF 小鼠模型，结果表明，该菌可导致严重的肺部炎症及较高的死亡率[74]。Brao 等利用基因编辑技术构建了 *Scnn1b*-transgenic (Tg) BALB/c 小鼠，具有与 CF 相似的症状，并通过鼻腔感染接种野生和临床分离的铜绿假单胞菌，通过对比发现，接种临床株的 *Scnn1b*-Tg 小鼠具有更高的细菌负担及强烈的免疫应答，为探究铜绿假单胞菌在 CF 肺中早期定殖提供了高效的动物模型[75]。

1.1.3 鼠类动物胃肠道感染模型

细菌是导致人体肠道感染及相关疾病的重要原因，生物被膜能够加剧此类疾病的顽固性，造成慢性疾病及医疗负担[76]。胃肠道内复杂的营养环境为致病菌生物被膜的形成提供了天然场所，而体外模拟并不能成功复制这种复杂的胃肠道环境，因此动物模型有助于研究由生物被膜导致的相关胃肠道感染。Barnes 等通过以经口灌胃的方式在无菌小鼠胃肠道内接种粪肠球菌，构建了胃肠道无菌小鼠模型，通过显微成像技术表明，粪肠球菌在无菌小鼠肠道内定殖与生物被膜的形成有关[77]。Gallego-Hernandez 等开发了可定量分析生物被膜空间分布的软件 BiofilmQ，借助小鼠模型对比了浮游态和被膜态霍乱弧菌的肠道感染差异，结果表明两种状态的霍乱弧菌呈现迥异的定殖情况与空间分布，被膜态细菌在肠道中定殖使相关毒力因子的表达显著上调，说明了生物被膜能够增强细菌的致病力[78]。

1.1.4 鼠类动物皮肤 (伤口) 感染模型

皮肤是人体重要的屏障器官，皮肤损伤引起的细菌性感染可能会使宿主面临较大的感染风险[79]，其中危害最大的是被膜态的金黄色葡萄球菌[80]和铜绿假单胞菌[81]。以往研究中，许多模型被开发来模拟不同的皮肤损伤，包括皮肤擦伤、烧伤、外科和切除伤口等，小鼠模型是最常用的动物模型之一[82]。

由被膜态细菌引发烧伤感染不是单一的病理过程，可导致多种器官、系统的结构和功能缺陷，甚至败血症的发生，呈现出较高的死亡率[83]。Dai 等构建了耐甲氧西林金黄色葡萄球菌感染的小鼠皮肤擦伤模型，通过该模型来研究生物被膜形成和光动力疗法对皮肤伤口感染的治疗效果[84]。Brandenburg 等通过在大鼠皮肤表面接种铜绿假单胞菌，构建了大鼠烫伤模型，经过为时 11 d 的动态监测，在小鼠伤口处检测到铜绿假单胞菌生物被膜的定殖，并呈现强烈的炎症反应，与对照组相比，铜绿假单胞菌生物被膜的存在加剧了炎症反应，使得大

量中性粒细胞涌入炎症部位，导致更严重的组织损伤[85]。

1.1.5 鼠类动物植入物感染模型

鼠模型除应用于上述组织相关的感染中，还被用于研究与医疗器械相关的生物被膜感染，其中包括人工耳蜗[33]、手术螺钉[34]、中枢神经系统导管[35]及尿导管等。器械相关生物被膜感染最早发现于一个复发性金黄色葡萄球菌感染患者的植入起搏器中[86]。2015 年，Cevizci 等将肺炎双球菌浸泡的人工耳蜗设备植入豚鼠的耳后，用于研究生物被膜介导的植入式耳蜗设备感染，并测试了一种新型群体感应抑制剂的疗效[33]。Glage 等通过在大鼠的颅骨中植入钛螺钉，模拟术中金黄色葡萄球菌感染，开发了生物被膜及相关炎症反应的大鼠模型[34]。Snowden 等开发了一个中枢神经导管内生物被膜引起炎症反应的小鼠模型，与无菌导管相比，植入带菌导管的小鼠表现出强烈的免疫细胞渗透和炎症反应[35]。Witso 等开发了一个用于研究肌肉骨骼系统中慢性植入物感染的小鼠模型，结果显示，在所有植入物上都存在生物被膜[87]。Brandenburg 等采用膀胱切开术植入带有热带念珠菌的导管，与浮游态菌株相比，被膜态的热带念珠菌呈现出更高更持久的感染效率，并能更有效地逃避宿主反应[88]。

1.2 兔类动物感染模型

实验兔是由遗传背景明确、来源清楚的家兔经人工饲养、繁育，并对其携带的微生物及寄生虫进行控制培育而成，具有易饲养、抗病力强、繁殖率高等优点[89]，且其生理代谢、组织结构及病理反应与人类高度相似，被广泛应用于生殖生理学、心血管疾病、免疫学、皮肤反应等实验研究中[90]，在伤口、关节炎及植入物介导的骨髓炎等细菌生物被膜感染研究中也得到了广泛应用 (图 2)。

1.2.1 兔类动物伤口感染模型

小鼠的伤口主要表现为挛缩愈合[91]，而大多数人类伤口的愈合是通过上皮化和肉芽形成的，兔则可精确地模拟了人类慢性伤口中出现的真皮缺失[89]，因此常被应用于致病菌生物被膜导致的皮肤伤口感染的研究中[90]。Hermans 等建立了兔皮肤感染模型，探究了高毒力和低

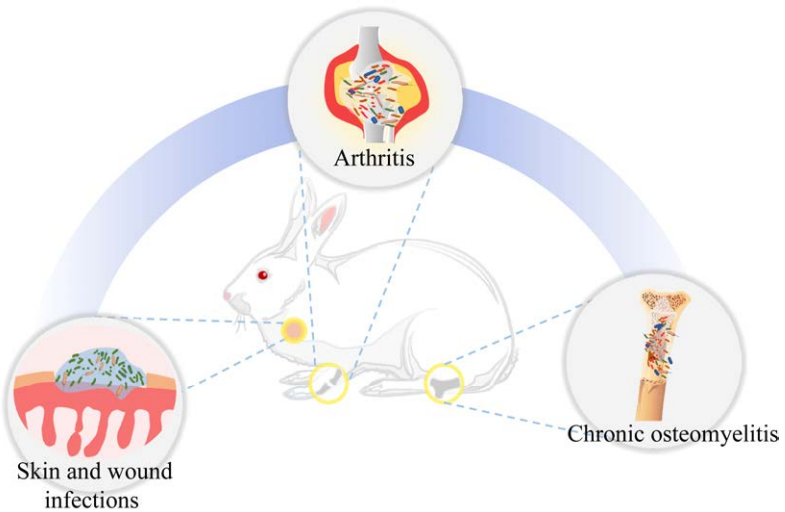

图 2 生物被膜感染的兔类动物模型应用场景

Figure 2 Application of biofilm-infected rabbit animal models.

毒力金黄色葡萄球菌对皮肤造成的损伤差异。结果表明，高毒力和低毒力菌株在脓肿的大小和严重程度上存在显著差异，与低毒株相比，接种高毒株的实验兔发病更急、脓肿更大、消退更慢，为进一步探究金黄色葡萄球菌毒力因子在感染过程中发挥作用奠定了良好的模型基础[91]。

此外，兔模型还被广泛应用于生物被膜感染治疗方案的开发，其中，最具代表性的是兔耳生物被膜模型，许多研究人员基于该模型展开了一系列的研究[92-94]。Hong 等运用兔耳生物被膜模型，评价了一种特异性噬菌体对金黄色葡萄球菌生物被膜感染的治疗效果，发现在生物被膜结构破坏的情况下，噬菌体可有效治疗局部金黄色葡萄球菌引起的伤口感染[93]。D'Arpa 等采用兔耳生物被膜模型，证明了负压伤口治疗 (negative pressure wound therapy, NPWT) 可有效降低伤口中的细菌数量、毒力因子和生物被膜的形成[94]。

1.2.2 兔类动物关节炎感染模型

脓毒性关节炎是一种侵袭性疾病，可导致严重的关节软骨病或骨缺损，造成关节功能的不可逆损害[95-96]，并可引发严重的败血症，常见的致病菌为金黄色葡萄球菌[97]、铜绿假单胞菌[98]等，最常见的患病部位是膝关节[99]。Olney 等借助兔子模型，从患有败血症的兔子身上抽取血液，并直接注射到未患病兔子的关节中，成功开发了一个可用于研究脓毒性关节炎的动物模型[100]。Sinha 等[101]、Marcheix 等[102]和 Oner 等[103]选用新西兰白兔构建了金黄色葡萄球菌脓毒性关节炎的兔子模型，并利用此模型评估了抗生素对关节感染的治疗效果。Wei 等将铜绿假单胞菌注射到兔子的膝关节腔中，结果显示，首次观察到铜绿假单胞菌可在关节腔内形成生物被膜，接种 7 d 后膝关节腔内有大量炎性渗出物，并借助此模型探明了细胞内环鸟苷二磷酸浓度对生物膜形成具有重大影响[104]。

1.2.3 兔类动物骨髓炎感染模型

骨髓炎是指化脓性细菌感染骨髓、骨皮质和骨膜而引起的炎症性疾病[105]，植入物介导的骨髓炎是骨科创伤患者接受骨折固定术后的重大并发症，迄今仍是骨科临床一大难题[106]。植入物介导的骨髓炎通常由金黄色葡萄球菌引起，生物被膜增强了该菌在植入物表面的附着能力，并对抗生素和免疫因子产生耐受性，从而加剧了细菌的慢性感染[107]。此类技术存在一定的局限性，即缺乏种植稳定性和髓内固定稳定性。在此基础上 Zhang 等对原有模型进一步改进，将被膜态金黄色葡萄球菌接种于钢板固定的股骨骨折处，实验兔出现脓液、骨膜反应、皮质破坏及吸收等症状，并在钢板上观察到生物被膜的形成，成功构建了一种骨折固定术后感染的兔类模型，为植入物相关骨髓炎的研究提供了一种新工具[108]。Hovis 等在兔胫骨手术部位植入携带耐甲氧西林金黄色葡萄球菌的固定植入物，并应用万古霉素进行治疗，结果显示，万古霉素可显著降低兔胫骨植入物的细菌感染和生物被膜的形成，从而治愈植入物相关骨髓炎[109]。

1.3 猪类动物模型

猪作为实验常用的哺乳动物之一，其皮肤系统、心血管系统、免疫系统等解剖学和免疫系统方面都与人体极为相似[110-111]，并拥有重量相近的器官，是研究人畜共患性疾病、动物烈性传染病、生物被膜慢性感染等疾病的理想模式生物[112-113]。细菌生物被膜感染性关节炎会引起大量的关节积液，是判断该病的重要病理特征，但由于鼠和兔等啮齿动物体型较小，很难从这些物种中获得大量积液以用于疾病的研究[114]，而猪模型可以很好地弥补这一缺陷 (图 3)。

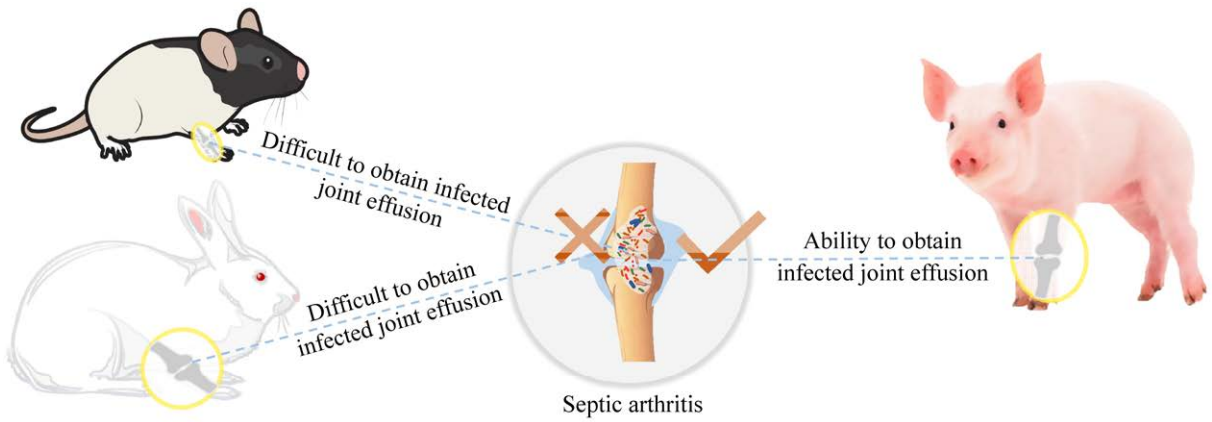

图 3　猪类动物模型用于细菌生物被膜感染性关节炎的优势

Figure 3　Advantages of using pig animal models for bacterial biofilm infectious arthritis.

Harrison 等使用猪模型研究了关节积液中金黄色葡萄球菌等致病菌的生长情况，结果表明这些致病菌能够在积液中聚集并形成生物被膜，同时对抗生素表现出较强的耐受性[47]。Johansen 等运用猪模型探究了金黄色葡萄球菌及生物被膜导致骨髓炎的病理，在猪的右股中接种金黄色葡萄球菌，通过肽核酸荧光原位杂交证明细菌聚集并形成生物被膜，在骨髓炎的发展中起着一定作用[115]。

2　非哺乳类动物模型

近年来，一些非哺乳动物模型已被开发应用于细菌生物被膜的感染研究中，包括秀丽隐杆线虫[116]、斑马鱼[117]和果蝇[118]等 (图 4)。与哺乳动物模型相比，非哺乳动物无法表现出复杂的免疫反应，限制了它们在某些致病菌感染研究中的适用性，但由于非哺乳动物模型通常具备体型较小、生长周期较短、操作简便、价格便宜、重复性强等优势，可用于细菌生物被膜体内感染研究的初探或病理反应的大规模筛选，可作为哺乳动物模型实验的良好替代或补充[119-120]。

2.1　黑腹果蝇 (*Drosophila melanogaster*)

果蝇模型已被广泛应用于预测致病菌在哺乳动物宿主体内的毒力变化[121-122]，特别是在铜绿假单胞菌生物被膜感染中得到了较好的验证[123]，具体原因如下：首先，果蝇是一种复杂的无脊椎动物[121]，具有得天独厚的免疫和遗传优势，与哺乳动物先天免疫系统有高度的相似性[124]；其次，在 2000 年果蝇的全基因组测序已经完成，其与 75%的人类致病相关基因具有同源性[125]，是研究宿主对生物被膜感染的有力模型；第三，果蝇模型操作简便，且成本低廉适合高通量筛选。

Mulcahy 等使用针头蘸取铜绿假单胞菌并刺入果蝇腹部，构建了一种生物被膜感染的果蝇模型，用于研究生物被膜中铜绿假单胞菌和宿主的相互作用，显微镜分析显示，在感染果蝇过程中，铜绿假单胞菌主要以生物被膜形式存在，并且细菌耐药性显著增强[60]。Tufenkji 等运用黑腹果蝇模拟了铜绿假单胞菌慢性感染的体内试验，该实验通过连续 5 d 的冻结-解冻实验 (free and thaw, FT)，对两种极端的条件进行了测试，结果发现，FT 暴露显著增加铜绿假

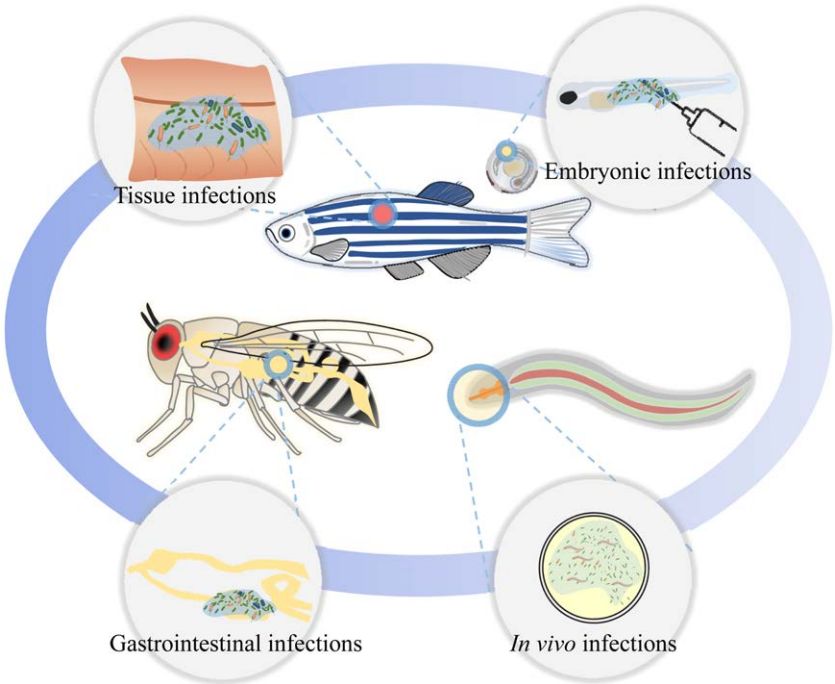

图 4 生物被膜感染的非哺乳类动物模型应用场景

Figure 4 Application of biofilm-infected non-mammalian animal models.

单胞菌生物被膜的形成能力和毒力[126]。而 Alexeyev 等建立了一种新的适合研究丙酸杆菌生物被膜的活体果蝇模型，该模型既可用于丙酸杆菌生物被膜的机理研究，也可用于生物被膜靶向治疗模式的研究[127]。

2.2 斑马鱼 (*Danio rerio*)

斑马鱼的基因与人类基因相似度达到 87%[128]，近年来被广泛应用在分子发育生物学、环境毒理学等研究中。斑马鱼胚胎的外部发育和光学清晰度有助于可视化操作，并能够对细胞结构和行为进行活体成像观测[129]，有效弥补了鼠、兔、猪等较大型动物无法开展活体成像研究的空缺。与其他已建立的脊椎动物感染模型 (如小鼠和大鼠) 相比，斑马鱼模型的优势包括体型小、生长快、生命周期相对较短、繁殖方便，在基因组分析、活体成像和高通量小分子筛选等新技术的加持下，斑马鱼已然成为生物医学研究中的重要工具[130-131]。

目前，许多研究已经使用肌肉或腹腔注射等方式，感染成年或刚孵化的斑马鱼，以分析细菌的致病性及其与宿主的相互作用[129]。Subramaniyan 等采用肌肉注射的方式接种鼠伤寒沙门氏菌以感染健康的斑马鱼，并通过该模型证实了铂纳米颗粒 (platinum nanoparticles, PtNCs) 的治疗功效，发现 PtNCs 可以消除鼠伤寒沙门氏菌的生物被膜，进而抑制组织中鼠伤寒沙门氏菌的感染[56]。Lu 等利用斑马鱼胚胎的光学通透性，并借助成像技术检测白色念珠菌的感染过程，证明了基因型为 DST659 的白色念珠菌具有较强的生物被膜形成能力[57]。Milivojevic 等通过体外毒力数据预测与体内斑马鱼胚胎感染实验，证实了铜绿假单胞菌生物被膜对于其发挥细胞毒性具有重要作用[59]。

2.3 秀丽隐杆线虫 (*Caenorhabditis elegans*)

秀丽隐杆线虫是一种食菌动物，以生长在腐烂水果或植物上的细菌及其生物被膜为食[132]，

作为一种适用于发育生物学、感染行为研究等众多研究领域的模式生物，具有体积小、生长迅速、发育过程简单、身体透明、易于显微观测等优点[133]，可用于探析秀丽隐杆线虫与被膜态细菌作为捕食者-猎物间的复杂作用，还可作为开发抗菌药物的高通量、低成本体内感染模型[134]。

研究表明，假结核耶尔森氏菌、鼠疫耶尔森氏菌和嗜线虫病菌可在秀丽隐杆线虫头部周围形成生物被膜，通过堵塞口腔并阻止线虫吸收细菌，导致线虫因饥饿而死亡[135-137]。生物被膜还可一定程度上改变线虫的运动能力，Atkinson 等首次报道了在自然环境中，生物被膜可以阻止秀丽隐杆线虫的移动，并作为陷阱减少其对生物被膜的进一步损害，从而提高细菌的整体存活率[138]。Wang 等利用秀丽隐杆线虫模型研究发现，绿原酸 (chlorogenic acid, CA) 能抑制铜绿假单胞菌中生物被膜的形成，可用于提高铜绿假单胞菌感染的治疗效率[54]。Lee 等利用全反式维甲酸的活性提取物，治疗铜绿假单胞菌感染的秀丽隐杆线虫，与对照组相比，该活性提取物具有抗铜绿假单胞菌感染的特性，并能够减弱细菌毒素和生物被膜的形成，有效提高了秀丽隐杆线虫的存活能力[55]。张日丽运用秀丽隐杆线虫-白色念珠菌感染模型，开展了抗白色念珠菌感染药物的高通量筛选，发现龙血素 A 能够有效地抑制白色念珠菌的生物被膜[139]。

3 总结与展望

动物模型在细菌生物被膜研究中的广泛应用，可获得大量生物被膜相关感染的理论与实践知识，有助于生物被膜感染类疾病的研究及控制。这些模型的复杂程度各不相同，从简单的导管接种到复杂的整形外科手术，几乎所有生物被膜组织相关感染或器械相关感染都有相对应的模型，展现了此类动物模型的广阔应用前景。然而，由于细菌生物被膜自身复杂的特性，现阶段的动物模型尚不足以完全揭示生物被膜在人体内的形成机制，亟需新型动物模型的研究与开发。因此，本文针对目前动物模型在细菌生物被膜研究中的不足，对其未来发展方向提出以下 3 点展望。

3.1 建立标准化的生物被膜感染动物模型

基于动物模型探究生物被膜感染及治疗已有较深厚的研究基础，但目前动物模型的构建方式相对比较个性化。现行动物模型虽能解决一些生物被膜产生导致的实际问题，但在基础研究向实际应用的转化过程中，缺少相应的标准、法规或指南进行约束，造成了动物模型构建的困难以及人们对模型有效性的质疑[140]。因此，依托科研院所开发系统完备的生物被膜感染动物模型，并依托有关部门进行相关标准法规的建立，可为探究生物被膜相关疾病的感染机制研究提供可靠的科研工具，为生物被膜治疗方案的探索提供规范的流程。本文对生物被膜感染动物模型研究中的关键步骤进行汇总，绘制了一个模型构建流程图 (图 5)，以期为后续相关研究奠定理论基础。

3.2 运用人源化动物模型模拟生物被膜真实的感染情况

尽管普通动物模型被视为致病菌研究的可靠工具，然而动物和人之间的种属差异是客观存在的[14]，所以利用动物模型得到的实验结果有时不能适用到人体上。因此，在探究人体相关生物被膜感染疾病的形成机制的同时，需开发更接近人体实际情况的动物模型。目前使用的大多数动物模型，与患者的实际情况存在差距，因此构建具有人体相似免疫环境的动物模型显得尤为重要，例如目前国内外的研究热点：

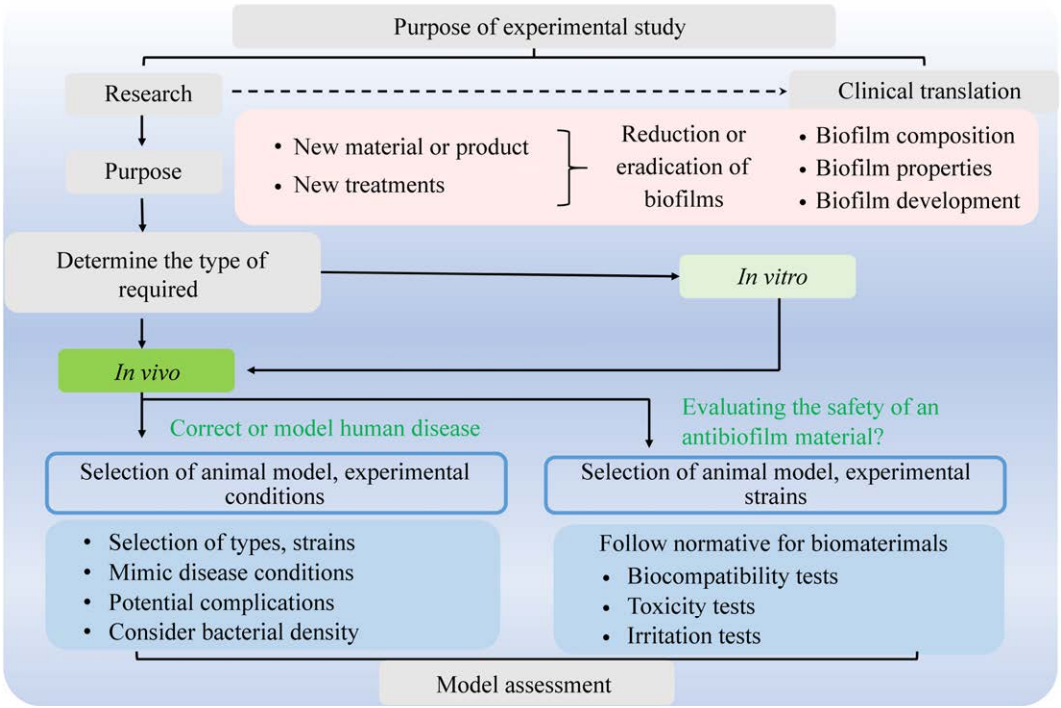

图 5　构建生物被膜感染动物模型的建议指南

Figure 5　Suggested guidelines for constructing biofilm-infected animal models.

人源化小鼠模型[141-142]，本课题组前期通过综述研究介绍了两种人源化动物模型：人源化菌群动物模型和人源化免疫系统动物模型，此类模型可对实验动物的肠道菌群或免疫系统实现“人源化”模拟，可较好地重现生物被膜在人体内感染的真实情况[143]。

3.3　运用动物模型评价并开发生物被膜抑制剂

生物被膜相关研究的最终目的是减轻或消除生物被膜所引起的疾病，迄今为止，借助不同动物模型对被膜态细菌的定殖情况、感染机制及宿主反应展开了一定的研究，但关于生物被膜的治疗或控制依旧是全世界范围内的焦点、难点问题。未来研究应继续开发细菌生物被膜相关新型动物模型，探究宿主与致病菌相互作用的分子机制，研发新型抗生物被膜的治疗或控制策略。目前，相关研究已陆续开展，例如，Hoque 等开发了一种抗生物被膜的水凝胶，通过细菌急性皮肤感染的大鼠模型、豚鼠模型和兔模型，证明了该水凝胶能够根除金黄色葡萄球菌和大肠杆菌生物被膜[144]。

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