



鳞翅目昆虫模式识别受体的研究进展*

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摘 要 昆虫虽然缺乏适应性免疫系统, 却进化出一套高效复杂的先天免疫系统来保护机体免受多种病原感染。昆虫先天免疫系统能够识别入侵病原体并迅速启动以消除潜在的威胁。免疫反应的第一步依赖于种系编码的模式识别受体, 模式识别受体可以检测并结合入侵病原体表面的病原相关分子模式, 进而激活下游免疫反应。到目前为止, 在鳞翅目昆虫中已经鉴定到多种模式识别受体, 如肽聚糖识别蛋白、 β -1,3-葡聚糖识别蛋白和 C 型凝集素等。多种模式识别受体分工协作, 共同参与复杂而精细的免疫应答。生物化学、遗传学和分子生物学等研究发现, 不同种类的模式识别受体具有结合特异性和适应性进化。本文将重点总结模式识别受体相关基因家族的起源、结构特征以及在鳞翅目模式昆虫的生理功能和进化等方面的研究进展, 为进一步研究其它鳞翅目昆虫的免疫系统, 探索免疫基因与宿主生态适应性的关系提供参考。

关键词 模式识别受体; 病原相关分子模式; 先天免疫; 免疫识别; 适应性进化

Advances in research on pattern recognition receptors in the Lepidoptera

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Abstract Though lacking an adaptive immune system, insects have evolved a highly efficient and complex innate immune system to protect them from multiple pathogens. This innate immune system can recognize invading pathogens, and rapidly activate an immune response to eliminate potential threats. The first step of the immune response is dependent on pattern recognition receptors (PRRs), which detect and bind certain pathogen-associated molecular patterns (PAMPs) on the surface of invading pathogens, thereby activating a downstream immune response. So far, a variety of PRRs have been identified in the Lepidoptera, such as peptidoglycan recognition protein (PGRP) and C-type lectin (CTL). Various types of PRRs work together to participate in complex and delicate immune responses. Research in biochemistry, genetics and molecular biology have found that different types of PRRs have unique binding specificity and adaptive evolution. This review focuses on summarizing the origin and structural characteristics of PRR related gene families, as well as progress in research on physiological functions and evolution of PRRs in Lepidopteran model insects. This information will assist further research on the immune system of other Lepidopteran insects and provide a reference for exploring the relationship between immune genes and mechanisms of host ecological adaptation.

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昆虫在长期的自然进化过程中,不断抵御外界损伤及病原物的侵袭,逐渐形成了一套高效复杂的先天免疫系统(Innate immune system)(Yu *et al.*, 2002; Hultmark, 2003; 胡启豪等, 2019),包括体液免疫和细胞免疫(图 1)。先天免疫并非完全是非特异的,通过有限数量的种系编码的模式识别受体(Pattern recognition receptors, PRRs)(Akira *et al.*, 2006)可以区别自身和各

种病原体,这一过程是通过模式识别受体特异性识别并结合病原相关分子模式(Pathogen associated molecular pattern, PAMP)来实现,PAMP指的是病原微生物内部或其细胞壁上的一些保守成分(Janeway, 1989; Lemaitre and Hoffmann, 2007)。病原体入侵的检测是防御反应成功的关键一步,是触发后续免疫反应的开关(Werner *et al.*, 2000; Wang *et al.*, 2005)。

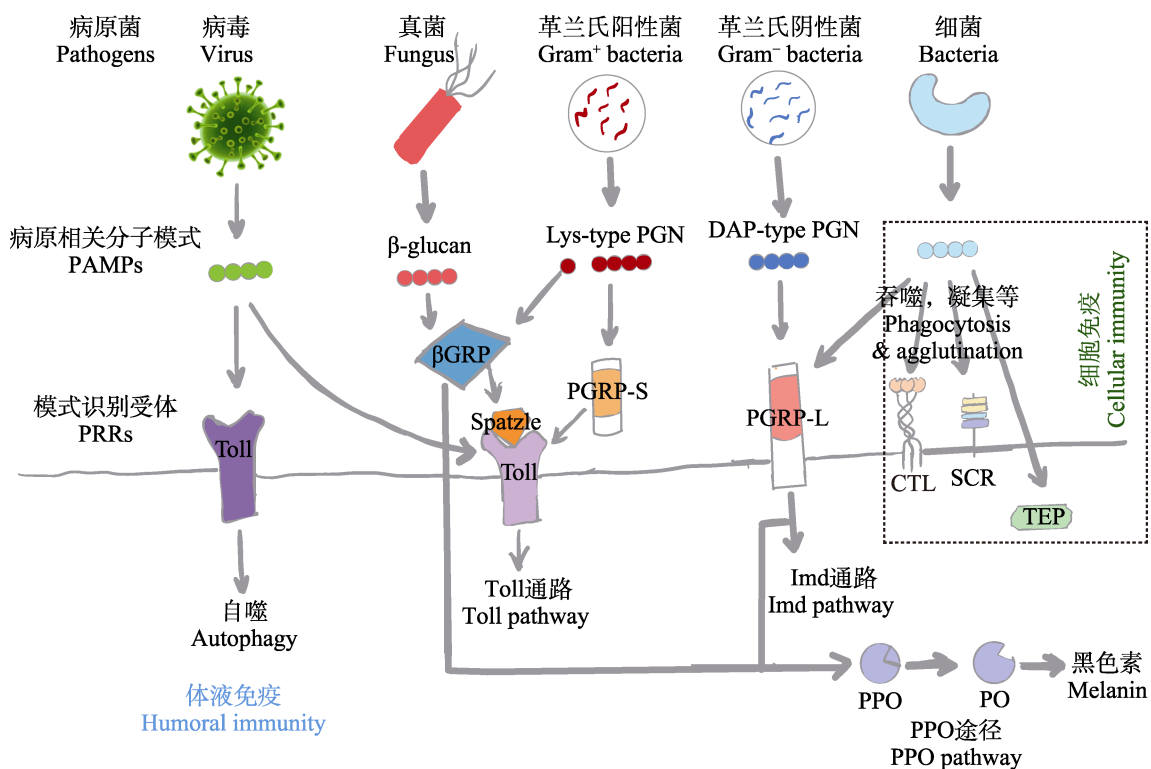


图 1 昆虫的先天免疫示意图 (Xia *et al.*, 2015; Hillyer, 2016; Lu *et al.*, 2020a)

Fig. 1 Schematic representation of innate immunity in insects (Xia *et al.*, 2015; Hillyer, 2016; Lu *et al.*, 2020a)

鳞翅目昆虫是最早应用于先天免疫研究的一类昆虫,如家蚕 *Bombyx mori*、烟草天蛾 *Manduca sexta* 等(Steiner, 2004)。随着基因组学和转录组学的发展,多种模式识别受体基因家族在鳞翅目昆虫中相继得以鉴定,同时比较基因组学分析也为探究昆虫的适应性分子进化模式的关系提供了契机(Viljakainen *et al.*, 2009; Bulmer *et al.*, 2010)。研究表明,免疫基因进化

速度比基因组平均进化速度更快,昆虫免疫基因的分化是由于基因家族的扩张与收缩,以及由宿主-病原体相互作用的正选择所驱动的,不同功能类别的免疫基因在选择和分化模式上存在较大差异(Sackton *et al.*, 2007; Viljakainen *et al.*, 2009)。

目前,已有相关文章综述了鳞翅目昆虫模式识别受体的研究进展(Lin *et al.*, 2020)。该综

述重点关注了 4 种鳞翅目昆虫中 C 型凝集素基因的进化与功能研究进展, 并简要介绍了肽聚糖识别蛋白、 β -1,3-葡聚糖识别蛋白和半乳糖凝集素 3 种模式识别受体的相关研究。本文主要对鳞翅目模式昆虫中模式识别受体的结构和功能的研究进展进行了综述, 并结合昆虫基因组学的发展, 对鳞翅目昆虫模式识别受体的比较基因组学研究、进化以及相关功能研究的发展方向进行了展望。

1 昆虫中模式识别受体的分类与基本特征

模式识别受体的概念是由美国免疫学家 Janeway 于 1989 年提出, 指的是一类种系编码的基因 (Janeway, 1989; Brubaker *et al.*, 2015), 可以检测入侵微生物表面的某些病原相关分子模式 (PAMP), 引起快速应答。PAMP 通常是病原微生物表面某些共有的高度保守的分子结构, 包括细菌的脂多糖 (Lipopolysaccharide, LPS)、肽聚糖 (Peptidoglycan, PGN) 和真菌的 β -1,3-葡聚糖, 以及病毒的核苷酸 (Pal and Wu, 2009; Wang *et al.*, 2017)。PAMP 是宿主免疫识别的理想靶标, 模式识别受体向着识别不同病原体的保守结构进化。在昆虫中鉴定到模式识别受体形式多样, 研究比较广泛的包括识别细菌的肽聚糖识别蛋白 (Peptidoglycan recognition protein, PGRP)、识别真菌的 β -1,3-葡聚糖识别蛋白 (β -1,3-glucan recognition protein, β GRP) 和既有识别功能又可以发挥凝集作用 (Agglutination) 的 C 型凝集素 (C-type lectin, CTL)。此外, 还存在一些其他的 PRR, 比如清道夫受体 (Scavenger receptor, SCR)、半乳糖凝集素 (Galectin, GALE)、含硫酯键蛋白 (Thioester protein, TEP)、纤维蛋白原相关蛋白 (Fibrinogen-related protein, FREP), 以及鳞翅目昆虫特有的 Hemolin 等 (Zhang *et al.*, 2015; Hillyer, 2016; Lu *et al.*, 2020a)。

PGRP 是从昆虫到哺乳动物保守的模式识别分子, 在 C 端都有一个保守的约 165 个氨基酸

的 PGRP 结构域 (Kang *et al.*, 1998; Xiong *et al.*, 2015), 能够特异性识别细菌及其特有的细胞壁成分肽聚糖 (Fabrick *et al.*, 2003; Dziarski, 2004)。昆虫 PGRP 具有四种特有的效应功能: 激活酚氧化酶原 (Prophenoloxidase, proPO) 级联反应、激活 Toll 受体、激活 IMD 通路和诱导吞噬 (Dziarski, 2004)。PGRP 结构域与噬菌体 T7 溶菌酶具有同源性 (Kang *et al.*, 1998; Kurata *et al.*, 2006), 部分 PGRP 保留了与 Zn^{2+} 配位的 5 个氨基酸残基, 具有酰胺酶活性, 能够直接裂解肽聚糖 (Kim *et al.*, 2003)。PGRP 家族根据分子量大小分为长型 (L 型) 和短型 (S 型) (Werner *et al.*, 2000)。短型 PGRP 主要是小于 20 ku 的分泌型胞外蛋白, 主要存在于血淋巴和表皮中, 在脂肪体中组成性表达或诱导合成, 也在肠道等其他组织器官中有表达 (Dziarski and Gupta, 2006), 能够识别革兰氏阳性菌表面的 DAP 肽聚糖, 激活 Toll 通路 (Lu *et al.*, 2020a)。长型 PGRP 的分子量通常是短型 PGRP 的两倍, N 端有一个可变序列, 根据信号肽和跨膜结构的有无, 可以分为胞外、跨膜或胞内蛋白, 主要在血细胞中表达, 在激活 IMD 通路中发挥免疫识别功能 (Valanne *et al.*, 2011; Wang *et al.*, 2019)。

β GRP 也被称为革兰氏阴性菌结合蛋白 (Gram-negative binding protein, GGBP), 可与真菌细胞壁的结构成分 β -1,3-葡聚糖和细菌表面的 LPS 结合 (Dai *et al.*, 2013)。典型的 β GRP 有一个 N 端的 β -1,3-葡聚糖识别结构域, 参与识别病原微生物, 并触发丝氨酸蛋白酶级联反应以激活 proPO 或 Toll 通路 (Ochiai and Ashida, 2000; Fabrick *et al.*, 2004; Jiang *et al.*, 2004; Takahasi *et al.*, 2009; Matskevich *et al.*, 2010; Sun *et al.*, 2011)。在 β GRP 的 C 端, 含有一个与 β -1,3-葡聚糖酶序列相似的糖基水解酶家族 16 (Glycosyl hydrolases family 16, GH16) 结构域, 但通常由于缺乏关键的氨基酸残基而不具有水解酶活性 (Lee *et al.*, 2000; Fabrick *et al.*, 2004; Jiang, 2008; Xiong *et al.*, 2015)。

CTL 是一类具有碳水化合物识别结构域 (Carbohydrate recognition domain, CRD) 的细

胞分泌蛋白或膜结合蛋白 (Xia *et al.*, 2018), 以 Ca^{2+} 依赖的方式与碳水化合物结合 (Kanost *et al.*, 2004; Drickamer and Taylor, 2015)。典型的 CRD 结构域由 110-130 个氨基酸残基组成, 这些氨基酸残基形成一个典型的折叠, 由 α 螺旋、 β 折叠和环组成, 两到三对二硫键维持其结构稳定 (Lu *et al.*, 2020b)。昆虫中的 C 型凝集素数量巨大, 种类繁多。根据 CRD 的数量, 可以分为单一 CRD 凝集素 (CTL-S)、双 CRD 凝集素 (Immulectins, IMLs) 和除了 CRD 还有其他功能域的凝集素 (CTL-X) (Rao *et al.*, 2015a, 2015b; Xia *et al.*, 2015, 2018; Song *et al.*, 2020)。

2 鳞翅目模式识别受体的功能研究

2.1 肽聚糖识别蛋白

第一个 PGRP 是从家蚕的血淋巴中分离出来, 作为一种模式识别受体, 专一性识别革兰氏阳性细菌细胞壁上的肽聚糖, 并触发酚氧化酶级联反应发生黑化 (Yoshida *et al.*, 1996)。目前, 已在多种鳞翅目昆虫中鉴定到 PGRP 基因 (表 1)。单从数目来看, PGRP 在鳞翅目中是比较保守的。在鳞翅目昆虫中, PGRP 参与免疫识别激

活下游免疫反应。例如, 烟草天蛾 PGRP1 识别肽聚糖来激活酚氧化酶级联反应 (Sumathipala and Jiang, 2010); 家蚕 PGRP-S1, 棉铃虫 *Helicoverpa armigera* PGRP-A 都能够激活酚氧化酶级联反应 (Yoshida *et al.*, 1996; Rao *et al.*, 2015a); 亚洲玉米螟 *Ostrinia furnacalis* PGRP-S 可结合金黄色葡萄球菌 *Staphylococcus aureus* 和苏云金芽孢杆菌 *Bacillus thuringiensis* 后激活酚氧化酶级联反应 (Sun *et al.*, 2014)。家蚕 PGRP-S5 与肽聚糖结合时, 不仅能激活 proPO 通路, 还能裂解肽聚糖表现出抗菌活性 (Chen *et al.*, 2014, 2016)。棉铃虫 PGRP-A 还可以起到凝集大肠杆菌 *Escherichia coli* 和金黄色葡萄球菌的作用 (Rao *et al.*, 2015a)。研究表明, 鳞翅目 PGRP 不仅可以作为免疫识别分子参与免疫信号转导, 而且一些 PGRP 具有酰胺酶活性或细菌凝集能力, 可以直接清除病原微生物 (Wang *et al.*, 2019)。果蝇 *Drosophila melanogaster* 中 PGRP-LC 作为受体可以作用于突触前调节突触囊泡的释放, 进而抑制突触后谷氨酸受体的功能 (Harris *et al.*, 2015)。鳞翅目昆虫中 PGRP 是否也参与神经系统的调节还需要今后的实验验证来回答。

表 1 几种代表性鳞翅目昆虫中的模式识别受体基因
Table 1 Pattern recognition receptor genes in several Lepidoptera insects

物种	PGRP	β GRP	CTL	GALE	TEP	SCR	FREP	参考文献 References
家蚕 <i>Bombyx mori</i>	12	4	23	4	3	18	3	Tanaka <i>et al.</i> , 2008; Rao <i>et al.</i> , 2015b
烟草天蛾 <i>Manduca sexta</i>	14	5	34	4	3	—	4	Rao <i>et al.</i> , 2015a; Zhang <i>et al.</i> , 2015
小菜蛾 <i>Plutella xylostella</i>	9	18	7	4	1	15	2	Xia <i>et al.</i> , 2015
棉铃虫 <i>Helicoverpa armigera</i>	9	5	26	3	4	10	2	Wang <i>et al.</i> , 2012; Xiong <i>et al.</i> , 2015
金蝙蝠蛾 <i>Hepialus xiaojinensis</i>	9	4	32	4	3	12	0	Meng <i>et al.</i> , 2015, 2019
亚洲玉米螟 <i>Ostrinia furnacalis</i>	10	4	36	2	—	9	—	Liu <i>et al.</i> , 2014; Shen <i>et al.</i> , 2018

PGRP 表示肽聚糖识别蛋白; β GRP 表示 β -1,3-葡聚糖识别蛋白; CTL 表示 C 型凝集素; GALE 表示半乳凝集素; TEP 表示含硫酯键蛋白; SCR 表示清道夫受体; FREP 表示纤维蛋白原相关蛋白。

PGRP represents peptidoglycan recognition protein; β GRP represents β -1,3-glucan recognition protein; CTL represents C-type lectin; GALE represents galectin; TEP represents thioester-containing protein; SCR represents scavenger receptor; FREP represents fibrinogen-related protein.

2.2 β -1,3 葡聚糖识别蛋白

GNBP 首次从家蚕的血淋巴中纯化得到, 与革兰氏阴性菌的细胞壁有强烈的结合能力 (Lee *et al.*, 1996)。进化分析表明, 这组蛋白质是由一个古老的葡聚糖酶基因的复制和功能分化而来 (Wang *et al.*, 2011)。鳞翅目昆虫的 β GRP 蛋白主要在脂肪体表达, 然后分泌到血淋巴。在家蚕, 金蝙蝠蛾 *Hepialus xiaojinensis* 和玉米螟中都鉴定到 4 个 β GRP 基因, 烟草天蛾和棉铃虫中有 5 个 β GRP 基因 (Ochiai and Ashida, 2000; Jiang *et al.*, 2004)。然而, 在小菜蛾 *Plutella xylostella* 中一共鉴定到 18 个 β GRP (表 1), 该家族在小菜蛾中发生了明显的基因扩增, 说明小菜蛾在生活环境中可能面临多种革兰氏阴性菌和真菌的威胁。

研究证实, 在几种鳞翅目昆虫中, β GRPs 都可以激活 proPO 级联反应 (Ma and Kanost, 2000; Fabrick *et al.*, 2003; Jiang *et al.*, 2004; Wu *et al.*, 2018)。烟草天蛾中的 MBP (Microbe binding protein), 是 β -1,3-葡聚糖酶相关蛋白超家族“GNBP”分支的成员, 通过与其他模式识别受体和丝氨酸蛋白酶共同作用, 触发 proPO 激活系统 (Wang *et al.*, 2011), 并且文章作者认为“GNBP”代表一种从“ β -葡聚糖酶”到“ β GRP”的过渡状态 (Wang *et al.*, 2011)。在烟草天蛾中, 血淋巴蛋白酶 14 (Hemolymph protease, HP14) 是酚氧化酶原激活系统的起始酶 (Ji *et al.*, 2004)。 β GRP1 和 β GRP2 可以与多糖结合导致 HP14 前体 (proHP14) 的激活并触发酚氧化酶原级联反应 (Wang and Jiang, 2006, 2010)。有趣的是, 烟草天蛾 β GRP2 与昆布多糖的比例会影响黑化反应的激活水平 (Takahashi *et al.*, 2014), 在摩尔比 [C]/[P] > 5 时形成可溶性复合物, 可以在较低水平下激活酚氧化酶; 而 [C]/[P] 在 1 左右时会形成不溶性复合物, 可强烈刺激原始血浆中的酚氧化酶原 (Rao *et al.*, 2018)。这种碳水化合物与蛋白质比例相关的调控方式可能正好解释了避免酚氧化酶原系统过度激活的机制。家蚕 β GRP4 具有类似按蚊和赤拟谷盗 *Tribolium*

castaneum β GRP 家族成员的活性催化残基 (Zou *et al.*, 2007), 但其葡聚糖催化活性还未得到证实。除了激活黑化反应, β GRP 还可以调控抗菌肽的表达, 如小菜蛾的 β GRP1 对抗菌肽 cecropins 的表达具有正调控作用 (Huang *et al.*, 2015)。

2.3 C 型凝集素

在鳞翅目昆虫中, CTL 数目在不同物种中差异较大。小菜蛾中仅有 7 个, 家蚕中有 23 个, 而在烟草天蛾中有 34 个 (表 1)。CTL 通过 CRD 域能够结合多种配体, 如甘露糖、半乳糖、N-乙酰氨基葡萄糖、N-乙酰半乳糖胺、 β 酰葡聚糖、非碳水化合物配体等 (Lu *et al.*, 2020b)。IML 的两个串联 CRD 有可能会显著增强其结合能力并扩大其识别谱 (Wang *et al.*, 2012)。MsIML-2 是一种多功能的模式识别受体: 具有促进吞噬的调节作用 (Ling and Yu, 2006); 与丝氨酸蛋白酶同源物 (SPH) 等形成复合物激活 proPO (Yu *et al.*, 2003); 其 CRD2 结构域可以结合秀丽隐杆线虫 *Caenorhabditis elegans* 增强其包被 (Encapsulation) 和黑化反应 (Yu and Kanost, 2004), 也参与了粘质沙雷氏菌 *Serratia marcescens* 的清除 (Yu and Kanost, 2003)。家蚕 CTL 家族成员包括 12 个 CTL-S, 6 个 IML 和 5 个 CTL-X。BmCTL5 只有一个 CRD 结构域, 在各组织中都有表达, 在 Ca^{2+} 参与下凝集金黄色葡萄球菌, 从而增强了对大肠杆菌和金黄色葡萄球菌的清除作用 (Rao *et al.*, 2015b; Zhan *et al.*, 2016)。BmCTL5 的表达是由白僵菌感染所诱导, 可作为模式识别受体参与 JAK/STAT 信号通路 (Geng *et al.*, 2016)。系统发育分析表明, 家蚕和烟草天蛾的 IML 基因可能由共同的祖先 CTL-S 基因通过复制产生的 (Xia *et al.*, 2018)。

根据免疫转录组研究, 在玉米螟中鉴定到 36 个 CTL (表 1), 包括 15 个 IML, 其中 IML-10 可以结合于血细胞表面促进其聚集, 并进一步提高其包被能力 (Song *et al.*, 2020)。在棉铃虫中, 一共鉴定到 26 个 CTL 基因, 包括 15 个 IML、9

个 CTL-S 和 2 个 CTL-X (Wang *et al.*, 2012; Xiong *et al.*, 2015)。棉铃虫 CTL3 是一个双 CRD 的 IML, 能识别大肠杆菌和金黄色葡萄球菌 (Wang *et al.*, 2012), 并促进棉铃虫的包被和黑化作用 (Wang *et al.*, 2017)。HaCTL7 主要依赖于 C 端有 4 个半胱氨酸残基的 CRD2 参与凝集、包被和黑化反应; 而 CRD1 与粒细胞结合, 可能参与吞噬等其他免疫过程 (Wang *et al.*, 2014)。研究表明, HaCTL14 可能是调控棉铃虫龄期依赖性抗真菌免疫的关键因素 (Wang *et al.*, 2017; Cheng *et al.*, 2018)。

在鳞翅目昆虫中, CTL 除了作为模式识别受体参与 proPO 的激活、血细胞介导的凝集、包被、调理作用 (Opsonification) 等重要的免疫防御机制 (Fujita *et al.*, 2004; Vasta, 2009), 还参与线虫感染防御反应。棉铃虫 HaCTL3 与线虫表面结合, 并且与 Ha β -整合素相互作用, 在包被反应中防御寄生线虫 (Wang *et al.*, 2017)。一些 CTL 还可以帮助寄生蜂实现定殖和入侵, 小菜蛾内寄生蜂的共生病毒 *Cotesia plutellae* bracovirus (CpBV) 编码的 CTLs 能帮助小菜蛾绒茧蜂 *Cotesia plutellae* 逃避宿主的免疫系统 (Madanagopal and Kim, 2007)。

在鳞翅目昆虫中, 大多数 CTL-S 和 CTL-X 的功能目前尚未阐明。与哺乳动物和双翅目不同的是, 鳞翅目昆虫 IML 家族在进化过程中发生了显著扩张 (Meng *et al.*, 2019)。全基因组分析显示: IML 家族基因存在于所有鳞翅目昆虫中, 而在其他目昆虫中, 目前只在赤拟谷盗中发现了 IML 家族成员 (Zou *et al.*, 2007; Lin *et al.*, 2020)。研究表明, CTL-X 中的其他结构域可以帮助和提高 CTL 的结合能力 (Xia *et al.*, 2018)。另外, 通过比较分析发现, 含有细胞外结构域 (Extracellular domain, CUB)、表皮生长因子样结构域 (Epidermal growth factor like domain, EGF) 和免疫球蛋白模块结构域 (Immunoglobulin modules, IG) 等非 CRD 结构域的 CTL-X 为鳞翅目昆虫所特有 (Xia *et al.*, 2018), 表明这些物种特异性的 CTL-X 可能是由于受到进化和选择的压力而产生并保留下来。

2.4 其他模式识别受体

Hemolin 属于免疫球蛋白超家族成员, 目前只在鳞翅目昆虫中发现, 而在其他目昆虫中均未发现 (Tanaka *et al.*, 2008; 宁媛媛等, 2009)。Hemolin 能够被微生物刺激后诱导高表达, 由 4 个免疫球蛋白 (Ig) 结构域组成, 常见于脊椎动物和无脊椎动物的细胞粘附蛋白中 (Su *et al.*, 1998; Jung *et al.*, 2019)。研究表明, Hemolin 可能参与宿主抗病毒免疫反应。例如, 在核型多角体病毒感染柞蚕 *Antheraea pernyi* 后, Hemolin 显著上调表达, 延缓柞蚕的发病时间 (Hirai *et al.*, 2004)。此外, Hemolin 还可以通过结合 LPS 或 LTA 来识别细菌, 并与血细胞结合, 诱导吞噬和结节的形成 (Eleftherianos *et al.*, 2007; Jiang *et al.*, 2010)。甜菜夜蛾 *Spodoptera exigua* Hemolin 在细菌和血细胞之间起调理作用, 保护甜菜夜蛾免受昆虫病原体的感染 (Jung *et al.*, 2019)。

清道夫受体是一个由细胞表面糖蛋白组成的多结构域受体超家族 (Gough and Gordon, 2000; Peiser *et al.*, 2002), 在生理或病理过程中执行多种功能, 如清除病原体、脂质运输和细胞内物质运输 (Canton *et al.*, 2013)。根据氨基酸序列的不同, 清道夫受体 (SCR) 家族分为 8 个亚家族 (A-H), 都属于跨膜糖蛋白。SCR 可识别多种配体, 并用于清除凋亡的细胞和细菌 (Peiser *et al.*, 2002)。在昆虫中, 主要存在 A、B、C 3 种类型的清道夫受体, 其中 C 型清道夫受体只在少数无脊椎动物中有发现 (Yang *et al.*, 2016)。B 型清道夫受体大部分都有一个 CD36 (Cluster of differentiation 36) 结构域, 成员众多, 主要参与对微生物的吞噬作用以及细胞凋亡等免疫反应 (Hawkes *et al.*, 2010; Canton *et al.*, 2013; Park, 2014; Jay *et al.*, 2015)。A 型和 C 型清道夫受体的研究还较少, 其中 SCRC 在草地贪夜蛾 *Spodoptera frugiperda* 中通过内吞作用介导苏云金芽孢杆菌杀虫蛋白 Vip3Aa 的内化 (Jiang *et al.*, 2018)。SCRC 在抗病毒免疫反应中所起到的作用, 我们知之甚少。2016 年在日本对虾 *Penaeus japonicus* 中首次确定了 SCRC 为

一种促进病毒吞噬的模式识别受体 (Yang *et al.*, 2016)。这为 C 型清道夫受体在抗病毒免疫研究方面提供了方向。

含硫酯键蛋白 (TEP) 是一种分泌蛋白, 是昆虫中与补体因子 C3/C4/C5 和大球蛋白家族序列相似的保守蛋白 (Blandin and Levashina, 2004; Shokal and Eleftherianos, 2017)。TEP 最初在果蝇 (Williams, 2007) 中发现, 包含一个序列基序 (motif) (GCGEQ)。在裂解激活后, 一些 TEP 利用半胱氨酸和谷氨酰胺残基之间的亚稳态硫酯键共价附着在病原体上, 并标记它们从而通过吞噬作用进行清除 (Lagueux *et al.*, 2000; Zou *et al.*, 2007)。TEP 不仅具有促进血细胞对革兰氏阴性细菌的吞噬作用 (Lagueux *et al.*, 2000; Oduol *et al.*, 2000; Boutros *et al.*, 2002), 还能直接或间接杀死一些入侵病原体 (Duval *et al.*, 2020)。TEP 家族在鳞翅目各物种中成员数目较少 (表 1)。

纤维蛋白原相关蛋白 (FREP) 是一种具有纤维蛋白原结构域的免疫相关蛋白, 参与识别无脊椎动物中的细菌和寄生虫 (Adema *et al.*, 1997; Gokudan *et al.*, 1999; Schroder *et al.*, 2003)。FREP 家族在冈比亚按蚊 *Anopheles gambiae* 中的数目很大, 有 59 个家族成员 (Dong and Dimopoulos, 2009), 在按蚊先天免疫系统中发挥核心作用, 不同成员间具有互补和协同功能 (Wang *et al.*, 2005; Waterhouse *et al.*, 2007; Dong and Dimopoulos, 2009)。果蝇、埃及伊蚊 *Aedes aegypti* 和赤拟谷盗 FREP 家族成员分别有 14、37 和 7 个 (Zou *et al.*, 2007; Dong and Dimopoulos, 2009)。然而, 鳞翅目昆虫中 FREP 家族成员数目较少: 家蚕中有 3 个, 小菜蛾和棉铃虫中仅有 2 个 (表 1)。

昆虫的半乳糖凝集素与胚胎发育或先天免疫有关 (Kamhawi *et al.*, 2004), 半乳糖凝集素是凝集素的一个 β -半乳糖苷结合蛋白家族, 具有 1-2 个保守碳水化合物识别域 (CRD) (Pace and Baum, 2002; Kamhawi *et al.*, 2004; 曾令瑜等, 2019)。与 C 型凝集素不同, 半乳糖凝集素能够特异性结合 β -半乳糖而不依赖 Ca^{2+} (Vasta,

2009)。半乳糖凝集素在细胞外或细胞内发挥作用, 尽管缺乏典型的信号肽, 但仍然可以通过非经典机制分泌, 影响细胞的生长和凋亡 (Vasta, 2009)。鳞翅目昆虫中半乳糖凝集素家族成员的数量变化不大 (表 1)。

3 总结与展望

生物防治方法大都涉及到天敌因子与害虫的相互作用, 很大程度上与昆虫免疫相关, 模式识别受体介导的免疫识别作用是昆虫免疫防御的关键环节, 因此模式识别受体的相关研究有助于生物制剂的设计与生产, 从而应用于害虫的生物防控实践。到目前为止, 一些鳞翅目昆虫的模式识别受体已经得到了较好的研究和表征, 例如烟草天蛾和家蚕的 PGRP 和 β GRP (Rao *et al.*, 2018; 徐平震, 2010), 但是在非模式的鳞翅目昆虫中仍鲜有研究。随着鳞翅目昆虫基因组及转录组计划的完成, 破译新发现模式识别受体基因的功能, 确定免疫信号通路之间的调控网络有望成为研究热点。

随着昆虫基因组测序的完成, 免疫研究也从传统的单基因功能研究向组学研究转变。大多数的免疫基因在昆虫中是共有的, 但在几个功能类别中, 特别是免疫识别和效应基因中, 已经发生了谱系特异性的分化 (Little and Cobbe, 2005; Evans *et al.*, 2006; Sackton *et al.*, 2007; Gerardo *et al.*, 2010; Tian *et al.*, 2010)。多种 PRR 蛋白家族在不同类群中扩张或收缩。例如, Hemolin 只存在于鳞翅目昆虫中 (Jung *et al.*, 2019), 其他目昆虫中未有发现; PGRP 在多数昆虫中都有发现, 但是半翅目豌豆蚜 *Acyrtosiphon pisum* 却没有编码 PGRP 的基因 (Gerardo *et al.*, 2010)。FREP 基因家族成员数目在不同昆虫中差异较大, 在冈比亚按蚊中数目极大, 而在鳞翅目昆虫中非常少, 家蚕中只有 2-4 个, 甚至在金蝙蝠蛾中未鉴定到 (表 1)。在鳞翅目昆虫中, 小菜蛾的 β GRP 家族由于基因复制发生了明显扩张, 而 CTL 家族却明显收缩 (表 1)。豌豆蚜、意大利蜜蜂 *Apis mellifera* 和榕小蜂 *Ceratosolen solmsi* 的一些免疫相关基因发生了退化甚至缺失

(Evans *et al.*, 2006; Gerardo *et al.*, 2010; Xiao *et al.*, 2013)。这些基因家族的扩张与收缩现象可能与昆虫的生活习性和所受环境压力因素有关。

免疫系统必须不断进化,以便在病原体变化时保持有效(Sackton *et al.*, 2007)。全基因组测序为研究先天免疫的进化提供了契机。研究发现,与其他基因相比,免疫基因进化速度更快,特别是与病原物识别有关的基因,可能是由宿主与病原物间相互作用的正选择所驱动(Bulmer *et al.*, 2010; Keehnen *et al.*, 2018)。在鳞翅目中, Tan 等(2021)及 Keehnen 等(2018)分别在帝王蝶 *Danaus plexippus* 和暗脉粉蝶 *Pieris napi* 的群体中,研究了全套免疫基因的进化。研究表明,不同功能类别的免疫基因在选择和分化模式上存在差异,其中信号转导基因最保守,效应基因主要受平衡选择,识别基因群体间分化程度较高。

最近有研究表明,免疫基因是鸟类和哺乳动物中正选择的共同“热点”(Shultz and Sackton, 2019)。受体基因或与病原体直接作用的基因往往是正选择的目标基因(Sackton *et al.*, 2007; Waterhouse *et al.*, 2007; Ellis *et al.*, 2012)。同病毒发生相互作用的蛋白质所经历的氨基酸变化是不发生相互作用的蛋白质的两倍(Enard *et al.*, 2016)。在果蝇中研究发现,识别蛋白的受选择位点主要位于微生物识别相关区域(Sackton *et al.*, 2007)。上述研究表明,宿主与病原体相互作用驱动免疫系统的适应性进化。从功能和进化的角度来看,受到强烈选择的基因是值得进一步研究的候选基因。通过对鳞翅目昆虫免疫基因的正选择分析,将有助于分析鳞翅目模式识别受体的起源和适应性进化,为功能研究提供新的靶标基因。

模式识别受体具有双重功效,不仅参与对抗入侵病原菌,也是共生菌与机体交流的接口,能够区分共生菌和致病菌,维持微生物和宿主的共生关系(Chu and Mazmanian, 2013)。模式识别受体与共生菌的互惠共生以及与致病菌之间的军备竞赛的作用机制是生命科学的热门研究方

向。另外,在鳞翅目昆虫中,免疫相关研究目前主要集中在细菌和真菌等方面,昆虫如何识别和抵御病毒侵染的研究仍较为缺乏。因此,模式识别受体在病毒与宿主互作中的作用及其分子机制的研究可以作为今后的重点研究方向。

参考文献 (References)

- Adema CM, Hertel LA, Miller RD, Loker ES, 1997. A family of fibrinogen-related proteins that precipitates parasite-derived molecules is produced by an invertebrate after infection. *Proceedings of the National Academy of Sciences of the United States of America*, 94(16): 8691–8696.
- Akira S, Uematsu S, Takeuchi O, 2006. Pathogen recognition and innate immunity. *Cell*, 124(4): 783–801.
- Blandin S, Levashina EA, 2004. Thioester-containing proteins and insect immunity. *Molecular Immunology*, 40(12): 903–908.
- Boutros M, Agaisse H, Perrimon N, 2002. Sequential activation of signaling pathways during innate immune responses in *Drosophila*. *Developmental Cell*, 3(5): 711–722.
- Brubaker SW, Bonham KS, Zanoni I, Kagan JC, 2015. Innate immune pattern recognition: A cell biological perspective. *Annual Review of Immunology*, 33: 257–290.
- Bulmer MS, Lay F, Hamilton C, 2010. Adaptive evolution in subterranean termite antifungal peptides. *Insect Molecular Biology*, 19(5): 669–674.
- Canton J, Neculai D, Grinstein S, 2013. Scavenger receptors in homeostasis and immunity. *Nature Reviews Immunology*, 13(9): 621–634.
- Chen K, Zhou L, Chen F, Peng Y, Lu Z, 2016. Peptidoglycan recognition protein-S5 functions as a negative regulator of the antimicrobial peptide pathway in the silkworm, *Bombyx mori*. *Developmental & Comparative Immunology*, 61: 126–135.
- Chen KK, Liu C, He Y, Jiang HB, Lu ZQ, 2014. A short-type peptidoglycan recognition protein from the silkworm: Expression, characterization and involvement in the prophenoloxidase activation pathway. *Developmental and Comparative Immunology*, 45(1): 1–9.
- Cheng Y, Lin Z, Wang JM, Xing LS, Xiong GH, Zou Z, 2018. CTL14, a recognition receptor induced in late stage larvae, modulates anti-fungal immunity in cotton bollworm *Helicoverpa armigera*. *Developmental and Comparative Immunology*, 84: 142–152.
- Chu H, Mazmanian SK, 2013. Innate immune recognition of the microbiota promotes host-microbial symbiosis. *Nat. Immunol.*,

- 14(7): 668–675.
- Dai HE, Hiromasa Y, Takahashi D, VanderVelde D, Fabrick JA, Kanost MR, Krishnamoorthi R, 2013. An initial event in the insect innate immune response: Structural and biological studies of interactions between beta-1,3-glucan and the N-terminal domain of beta-1,3-glucan recognition protein. *Biochemistry*, 52(1): 161–170.
- Dong Y, Dimopoulos G, 2009. Anopheles fibrinogen-related proteins provide expanded pattern recognition capacity against bacteria and malaria parasites. *Journal of Biological Chemistry*, 284(15): 9835–9844.
- Drickamer K, Taylor ME, 2015. Recent insights into structures and functions of C-type lectins in the immune system. *Current Opinion in Structural Biology*, 34: 26–34.
- Duval D, Pichon R, Lassalle D, Laffitte M, Gourbal B, Galinier R, 2020. A new assessment of thioester-containing proteins diversity of the freshwater snail *Biomphalaria glabrata*. *Genes*, 11(1): 69.
- Dziarski R, 2004. Peptidoglycan recognition proteins (PGRPs). *Molecular Immunology*, 40(12): 877–886.
- Dziarski R, Gupta D, 2006. The peptidoglycan recognition proteins (PGRPs). *Genome Biology*, 7(8): 232.
- Eleftherianos I, Gokcen F, Felfoldi G, Millichap PJ, Trenczek TE, ffrench-Constant RH, Reynolds SE, 2007. The immunoglobulin family protein Hemolin mediates cellular immune responses to bacteria in the insect *Manduca sexta*. *Cellular Microbiology*, 9(5): 1137–1147.
- Ellis JS, Turner LM, Knight ME, 2012. Patterns of selection and polymorphism of innate immunity genes in bumblebees (Hymenoptera: Apidae). *Genetica*, 140(4/6): 205–217.
- Enard D, Cai L, Gwennap C, Petrov DA, 2016. Viruses are a dominant driver of protein adaptation in mammals. *Elife*, 5: e12469.
- Evans JD, Aronstein K, Chen YP, Hetru C, Imler JL, Jiang H, Kanost M, Thompson GJ, Zou Z, Hultmark D, 2006. Immune pathways and defence mechanisms in honey bees *Apis mellifera*. *Insect Molecular Biology*, 15(5): 645–656.
- Fabrick JA, Baker JE, Kanost MR, 2003. cDNA cloning, purification, properties, and function of a beta-1,3-glucan recognition protein from a pyralid moth, *Plodia interpunctella*. *Insect Biochemistry and Molecular Biology*, 33(6): 579–594.
- Fabrick JA, Baker JE, Kanost MR, 2004. Innate immunity in a pyralid moth-functional evaluation of domains from a beta-1,3-glucan recognition protein. *Journal of Biological Chemistry*, 279(25): 26605–26611.
- Fujita T, Matsushita M, Endo Y, 2004. The lectin-complement pathway-its role in innate immunity and evolution. *Immunological Reviews*, 198: 185–202.
- Geng T, Lv DD, Huang YX, Hou CX, Qin GX, Guo XJ, 2016. JAK/STAT signaling pathway-mediated immune response in silkworm (*Bombyx mori*) challenged by *Beauveria bassiana*. *Gene*, 595(1): 69–76.
- Gerardo NM, Altincicek B, Anselme C, Atamian H, Barribeau SM, De Vos M, Duncan EJ, Evans JD, Gabaldon T, Ghanim M, Heddi A, Kaloshian I, Latorre A, Moya A, Nakabachi A, Parker BJ, Perez-Brocail V, Pignatelli M, Rahbe Y, Ramsey JS, Spragg CJ, Tamames J, Tamarit D, Tamborindeguy C, Vincent-Monegat C, Vilcinskis A, 2010. Immunity and other defenses in pea aphids, *Acyrtosiphon pisum*. *Genome Biology*, 11(2): R21.
- Gokudan S, Muta T, Tsuda R, Koori K, Kawahara T, Seki N, Mizunoe Y, Wai SN, Iwanaga S, Kawabata S, 1999. Horseshoe crab acetyl group-recognizing lectins involved in innate immunity are structurally related to fibrinogen. *Proceedings of the National Academy of Sciences of the United States of America*, 96(18): 10086–10091.
- Gough PJ, Gordon S, 2000. The role of scavenger receptors in the innate immune system. *Microbes and Infection*, 2(3): 305–311.
- Harris N, Braiser DJ, Dickman DK, Fetter RD, Tong A, Davis GW, 2015. The innate immune receptor PGRP-LC controls presynaptic homeostatic plasticity. *Neuron*, 88(6): 1157–1164.
- Hawkes M, Li X, Crockett M, Diassiti A, Finney C, Min-Oo G, Liles WC, Liu J, Kain KC, 2010. CD36 deficiency attenuates experimental mycobacterial infection. *BMC Infectious Diseases*, 10: 299–299.
- Hillyer JF, 2016. Insect immunology and hematopoiesis. *Developmental and Comparative Immunology*, 58: 102–118.
- Hirai M, Terenius O, Li W, Faye I, 2004. Baculovirus and dsRNA induce Hemolin, but no antibacterial activity, in *Antheraea pernyi*. *Insect Molecular Biology*, 13(4): 399–405.
- Hu QH, Lu YZ, Yu XQ, 2019. Advances in research on the innate immune system of *Drosophila melanogaster*. *Chinese Journal of Applied Entomology*, 56(5): 947–956. [胡启豪, 卢玉珍, 余小强, 2019. 黑腹果蝇先天免疫研究进展. *应用昆虫学报*, 56(5): 947–956.]
- Huang W, Xu X, Freed S, Zheng Z, Wang S, Ren S, Jin F, 2015. Molecular cloning and characterization of a β -1,3-glucan recognition protein from *Plutella xylostella* (L.). *New Biotechnology*, 32(2): 290–299.
- Hultmark D, 2003. *Drosophila* immunity: Paths and patterns. *Current Opinion in Immunology*, 15(1): 12–19.
- Janeway CAJ, 1989. Approaching the asymptote? Evolution and revolution in immunology. *Journal of Immunology*, 191(9): 147–154.

- 4475–4487.
- Jay AG, Chen AN, Paz MA, Hung JP, Hamilton JA, 2015. CD36 binds oxidized low density lipoprotein (LDL) in a mechanism dependent upon fatty acid binding. *Journal of Biological Chemistry*, 290(8): 4590–4603.
- Ji CY, Wang Y, Guo XP, Hartson S, Jiang HB, 2004. A pattern recognition serine proteinase triggers the prophenoloxidase activation cascade in the tobacco hornworm, *Manduca sexta*. *Journal of Biological Chemistry*, 279(33): 34101–34106.
- Jiang H, Ma C, Lu ZQ, Kanost MR, 2004. β -1,3-glucan recognition protein-2 (β GRP-2) from *Manduca sexta*: An acute-phase protein that binds β -1,3-glucan and lipoteichoic acid to aggregate fungi and bacteria and stimulate prophenoloxidase activation. *Insect Biochemistry and Molecular Biology*, 34(1): 89–100.
- Jiang H, 2008. The biochemical basis of antimicrobial responses in *Manduca sexta*. *Insect Science*, 15(1): 53–66.
- Jiang H, Vilcinskas A, Kanost MR, 2010. Immunity in lepidopteran insects//Soderhall K(ed.). *Invertebrate Immunity*. Vol. 708. New York: Springer Science+Business Media, LLC. 181–204.
- Jiang K, Hou XY, Tan TT, Cao ZL, Mei SQ, Yan B, Chang J, Han L, Zhao D, Cai J, 2018. Scavenger receptor-C acts as a receptor for *Bacillus thuringiensis* vegetative insecticidal protein Vip3Aa and mediates the internalization of Vip3Aa via endocytosis. *PLoS Pathogens*, 14(10): e1007347.
- Jung J, Sajjadian SM, Kim Y, 2019. Hemolin, an immunoglobulin-like peptide, opsonizes nonself targets for phagocytosis and encapsulation in *Spodoptera exigua*, a lepidopteran insect. *Journal of Asia-Pacific Entomology*, 22(3): 947–956.
- Kamhawi S, Ramalho-Ortigao M, Pham VM, Kumar S, Lawyer PG, Turco SJ, Barillas-Mury C, Sacks DL, Valenzuela JG, 2004. A role for insect galectins in parasite survival. *Cell*, 119(3): 329–341.
- Kang DW, Liu G, Lundstrom A, Gelius E, Steiner H, 1998. A peptidoglycan recognition protein in innate immunity conserved from insects to humans. *Proceedings of the National Academy of Sciences of the United States of America*, 95(17): 10078–10082.
- Kanost MR, Jiang HB, Yu XQ, 2004. Innate immune responses of a lepidopteran insect, *Manduca sexta*. *Immunological Reviews*, 198: 97–105.
- Keehnen NLP, Hill J, Nylin S, Wheat CW, 2018. Microevolutionary selection dynamics acting on immune genes of the green-veined white butterfly, *Pieris napi*. *Molecular Ecology*, 27(13): 2807–2822.
- Kim MS, Byun MJ, Oh BH, 2003. Crystal structure of peptidoglycan recognition protein LB from *Drosophila melanogaster*. *Nature Immunology*, 4(8): 787–793.
- Kurata S, Arikawa S, Kawabata S, 2006. Recognition of pathogens and activation of immune responses in *Drosophila* and horseshoe crab innate immunity. *Immunobiology*, 211(4): 237–249.
- Lagueux M, Perrodou E, Levashina EA, Capovilla M, Hoffmann JA, 2000. Constitutive expression of a complement-like protein in Toll and JAK gain-of-function mutants of *Drosophila*. *Proceedings of the National Academy of Sciences of the United States of America*, 97(21): 11427–11432.
- Lee SY, Wang RG, Soderhall K, 2000. A lipopolysaccharide- and beta-1,3-glucan-binding protein from hemocytes of the freshwater crayfish *Pacifastacus leniusculus* purification, characterization, and cDNA cloning. *Journal of Biological Chemistry*, 275(2): 1337–1343.
- Lee WJ, Lee JD, Kravchenko VV, Ulevitch RJ, Brey PT, 1996. Purification and molecular cloning of an inducible gram-negative bacteria-binding protein from the silkworm, *Bombyx mori*. *Proceedings of the National Academy of Sciences of the United States of America*, 93(15): 7888–7893.
- Lemaitre B, Hoffmann J, 2007. The host defense of *Drosophila melanogaster*. *Annual Review of Immunology*, 25: 697–743.
- Lin Z, Wang JL, Cheng Y, Wang JX, Zou Z, 2020. Pattern recognition receptors from lepidopteran insects and their biological functions. *Developmental and Comparative Immunology*, 108: 103688.
- Ling EJ, Yu XQ, 2006. Hemocytes from the tobacco hornworm *Manduca sexta* have distinct functions in phagocytosis of foreign particles and self dead cells. *Developmental and Comparative Immunology*, 30(3): 301–309.
- Little TJ, Cobbe N, 2005. The evolution of immune-related genes from disease carrying mosquitoes: Diversity in a peptidoglycan-and a thioester-recognizing protein. *Insect Molecular Biology*, 14(6): 599–605.
- Liu Y, Shen D, Zhou F, Wang G, An C, 2014. Identification of immunity-related genes in *Ostrinia furnacalis* against entomopathogenic fungi by RNA-seq analysis. *PLoS ONE*, 9(1): e86436.
- Lu Y, Su F, Li Q, Zhang J, Li Y, Tang T, Hu Q, Yu XQ, 2020a. Pattern recognition receptors in *Drosophila* immune responses. *Developmental and Comparative Immunology*, 102: 103468.
- Lu Y, Su F, Zhu K, Zhu M, Li Q, Hu Q, Zhang J, Zhang R, Yu XQ, 2020b. Comparative genomic analysis of C-type lectin-domain genes in seven holometabolous insect species. *Insect Biochemistry and Molecular Biology*, 126: 103451.
- Ma CC, Kanost MR, 2000. A beta 1,3-glucan recognition protein from an insect, *Manduca sexta*, agglutinates microorganisms and activates the phenoloxidase cascade. *Journal of Biological*

- Chemistry*, 275(11): 7505–7514.
- Madanagopal N, Kim Y, 2007. Biochemical characterization of C-type lectin in the diamondback moth, *Plutella xylostella* (Yponomeutidae: Lepidoptera). *Journal of Asia-Pacific Entomology*, 10(3): 229–237.
- Matskevich AA, Quintin J, Ferrandon D, 2010. The *Drosophila* PRR GGBP3 assembles effector complexes involved in antifungal defenses independently of its Toll-pathway activation function. *European Journal of Immunology*, 40(5): 1244–1254.
- Meng Q, Yu HY, Zhang H, Zhu W, Wang ML, Zhang JH, Zhou GL, Li X, Qin QL, Hu SN, Zou Z, 2015. Transcriptomic insight into the immune defenses in the ghost moth, *Hepialus xiaojinensis*, during an *Ophiocordyceps sinensis* fungal infection. *Insect Biochemistry and Molecular Biology*, 64: 1–15.
- Meng Q, Zhang JH, Zhang H, Zhou GL, Ni RY, Zhao YN, Qin QL, Zou Z, 2019. Comparative analysis of C-type lectin domain proteins in the ghost moth, *Thitarodes xiaojinensis* (Lepidoptera: Hepialidae). *Insect Science*, 26(3): 453–465.
- Ning YY, You MS, Wang CS, 2009. Advances in the mechanisms of insect immune recognition and pathogen immune escape. *Acta Entomologica Sinica*, 52(5): 567–575. [宁媛媛, 尤民生, 王成树, 2009. 昆虫免疫识别与病原体免疫逃避机理研究进展. 昆虫学报, 52(5): 567–575.]
- Ochiai M, Ashida M, 2000. A pattern-recognition protein for beta-1,3-glucan-The binding domain and the cDNA cloning of beta-1,3-glucan recognition protein from the silkworm, *Bombyx mori*. *Journal of Biological Chemistry*, 275(7): 4995–5002.
- Oduol F, Xu JN, Niare O, Natarajan R, Vernick KD, 2000. Genes identified by an expression screen of the vector mosquito *Anopheles gambiae* display differential molecular immune response to malaria parasites and bacteria. *Proceedings of the National Academy of Sciences of the United States of America*, 97(21): 11397–11402.
- Pace KE, Baum LG, 2002. Insect galectins: Roles in immunity and development. *Glycoconjugate Journal*, 19(7): 607–614.
- Pal S, Wu LP, 2009. Lessons from the fly: Pattern recognition in *Drosophila melanogaster*. *Advances in Experimental Medicine and Biology*, 653: 162–174.
- Park YM, 2014. CD36, a scavenger receptor implicated in atherosclerosis. *Experimental and Molecular Medicine*, 46(6): e99.
- Peiser L, Mukhopadhyay S, Gordon S, 2002. Scavenger receptors in innate immunity. *Current Opinion in Immunology*, 14(1): 123–128.
- Rao XJ, Cao X, He Y, Hu Y, Zhang X, Chen YR, Blissard G, Kanost MR, Yu XQ, Jiang H, 2015a. Structural features, evolutionary relationships, and transcriptional regulation of C-type lectin-domain proteins in *Manduca sexta*. *Insect Biochemistry and Molecular Biology*, 62: 75–85.
- Rao XJ, Shahzad T, Liu S, Wu P, He YT, Sun WJ, Fan XY, Yang YF, Shi Q, Yu XQ, 2015b. Identification of C-type lectin-domain proteins (CTLDPs) in silkworm *Bombyx mori*. *Developmental and Comparative Immunology*, 53(2): 328–338.
- Rao XJ, Zhan MY, Pan YM, Liu S, Yang PJ, Yang LL, Yu XQ, 2018. Immune functions of insect β GRPs and their potential application. *Developmental and Comparative Immunology*, 83: 80–88.
- Sackton TB, Lazzaro BP, Schlenke TA, Evans JD, Hultmark D, Clark AG, 2007. Dynamic evolution of the innate immune system in *Drosophila*. *Nature Genetics*, 39(12): 1461–1468.
- Schroder HC, Ushijima H, Krasko A, Gamulin V, Thakur NL, Diehl-Seifert B, Muller IM, Muller WEG, 2003. Emergence and disappearance of an immune molecule, an antimicrobial lectin, in basal Metazoa-A tachylectin-related protein in the sponge *Suberites domuncula*. *Journal of Biological Chemistry*, 278(35): 32810–32817.
- Shen D, Wang L, Ji J, Liu Q, An C, 2018. Identification and characterization of C-type lectins in *Ostrinia furnacalis* (Lepidoptera: Pyralidae). *Journal of Insect Science*, 18(2): 24.
- Shokal U, Eleftherianos I, 2017. Evolution and function of thioester-containing proteins and the complement system in the innate immune response. *Front Immunol.*, 8: 759.
- Shultz AJ, Sackton TB, 2019. Immune genes are hotspots of shared positive selection across birds and mammals. *Elife*, 8: e41815.
- Song ZK, Tian ML, Dong YP, Ren CB, Du Y, Hu J, 2020. The C-type lectin IML-10 promotes hemocytic encapsulation by enhancing aggregation of hemocytes in the Asian corn borer *Ostrinia furnacalis*. *Insect Biochemistry and Molecular Biology*, 118: 103314.
- Steiner H, 2004. Peptidoglycan recognition proteins: On and off switches for innate immunity. *Immunological Reviews*, 198: 83–96.
- Su XD, Gastinel LN, Vaughn DE, Faye I, Poon P, Bjorkman PJ, 1998. Crystal structure of hemolin: A horseshoe shape with implications for homophilic adhesion. *Science*, 281(5379): 991–995.
- Sumathipala N, Jiang H, 2010. Involvement of *Manduca sexta* peptidoglycan recognition protein-1 in the recognition of bacteria and activation of prophenoloxidase system. *Insect Biochemistry and Molecular Biology*, 40(6): 487–495.
- Sun Q, Xu XX, Freed S, Huang WJ, Zheng ZH, Wang S, Ren SX, Jin FL, 2014. Molecular characterization of a short peptidoglycan recognition protein (PGRP-S) from Asian corn borer (*Ostrinia furnacalis*) and its role in triggering proPO activity. *World Journal of Microbiology & Biotechnology*, 30(1): 1–10.

- 263–270.
- Sun ZX, Wu WJ, Zhang GR, 2011. Structure and expression of β -1,3-glucan recognition proteins from the ghost moth, *Thitarodes pui* (Hepialidae), and their response to *Beauveria bassiana* infection. *Journal of Insect Physiology*, 57(12): 1660–1669.
- Takahashi D, Dai H, Hiromasa Y, Krishnamoorthi R, Kanost MR, 2014. Self-association of an insect β -1,3-glucan recognition protein upon binding laminarin stimulates prophenoloxidase activation as an innate immune response. *Journal of Biological Chemistry*, 289(41): 28399–28410.
- Takahashi K, Ochiai M, Horiuchi M, Kumeta H, Ogura K, Ashida M, Inagaki F, 2009. Solution structure of the silkworm beta GRP/GNBP3 N-terminal domain reveals the mechanism for β -1,3-glucan-specific recognition. *Proceedings of the National Academy of Sciences of the United States of America*, 106(28): 11679–11684.
- Tan WH, Talla V, Mongue AJ, de Roode JC, Gerardo NM, Walters JR, 2021. Population genomics reveals variable patterns of immune gene evolution in monarch butterflies (*Danaus plexippus*). *Molecular Ecology*, 30(18): 4381–4391.
- Tanaka H, Ishibashi J, Fujita K, Nakajima Y, Sagisaka A, Tomimoto K, Suzuki N, Yoshiyama M, Kaneko Y, Iwasaki T, Sunagawa T, Yamaji K, Asaoka A, Mita K, Yamakawa M, 2008. A genome-wide analysis of genes and gene families involved in innate immunity of *Bombyx mori*. *Insect Biochemistry and Molecular Biology*, 38(12): 1087–1110.
- Tian CH, Gao B, Fang Q, Ye GY, Zhu SY, 2010. Antimicrobial peptide-like genes in *Nasonia vitripennis*: A genomic perspective. *BMC Genomics*, 11: 187.
- Valanne S, Wang JH, Ramet M, 2011. The *Drosophila* toll signaling pathway. *Journal of Immunology*, 186(2): 649–656.
- Vasta GR, 2009. Roles of galectins in infection. *Nature Reviews Microbiology*, 7(6): 424–438.
- Viljakainen L, Evans JD, Hasselmann M, Rueppell O, Tingek S, Pamilo P, 2009. Rapid evolution of immune proteins in social insects. *Molecular Biology and Evolution*, 26(8): 1791–1801.
- Wang JL, Liu XS, Zhang Q, Zhao HB, Wang YF, 2012. Expression profiles of six novel C-type lectins in response to bacterial and 20E injection in the cotton bollworm (*Helicoverpa armigera*). *Developmental and Comparative Immunology*, 37(2): 221–232.
- Wang JL, Zhang Q, Tang L, Chen L, Liu XS, Wang YF, 2014. Involvement of a pattern recognition receptor C-type lectin 7 in enhancing cellular encapsulation and melanization due to its carboxyl-terminal CRD domain in the cotton bollworm, *Helicoverpa armigera*. *Developmental & Comparative Immunology*, 44(1): 21–29.
- Wang P, Zhuo XR, Tang L, Liu XS, Wang YF, Wang GX, Yu XQ, Wang JL, 2017. C-type lectin interacting with beta-integrin enhances hemocytic encapsulation in the cotton bollworm, *Helicoverpa armigera*. *Insect Biochemistry and Molecular Biology*, 86: 29–40.
- Wang Q, Ren MJ, Liu XY, Xia HC, Chen KP, 2019. Peptidoglycan recognition proteins in insect immunity. *Molecular Immunology*, 106: 69–76.
- Wang XG, Zhao Q, Christensen BM, 2005. Identification and characterization of the fibrinogen-like domain of fibrinogen-related proteins in the mosquito, *Anopheles gambiae*, and the fruitfly, *Drosophila melanogaster*, genomes. *BMC Genomics*, 6: 114.
- Wang Y, Jiang HB, 2006. Interaction of β -1,3-glucan with its recognition protein activates hemolymph proteinase 14, an initiation enzyme of the prophenoloxidase activation system in *Manduca sexta*. *Journal of Biological Chemistry*, 281(14): 9271–9278.
- Wang Y, Jiang HB, 2010. Binding properties of the regulatory domains in *Manduca sexta* hemolymph proteinase-14, an initiation enzyme of the prophenoloxidase activation system. *Developmental and Comparative Immunology*, 34(3): 316–322.
- Wang Y, Sumathipala N, Rayaprolu S, Jiang H, 2011. Recognition of microbial molecular patterns and stimulation of prophenoloxidase activation by a β -1,3-glucanase-related protein in *Manduca sexta* larval plasma. *Insect Biochemistry and Molecular Biology*, 41(5): 322–331.
- Waterhouse RM, Kriventseva EV, Meister S, Xi Z, Alvarez KS, Bartholomay LC, Barillas-Mury C, Bian G, Blandin S, Christensen BM, Dong Y, Jiang H, Kanost MR, Koutsos AC, Levashina EA, Li J, Ligoxygakis P, MacCallum RM, Mayhew GF, Mendes A, Michel K, Osta MA, Paskewitz S, Shin SW, Vlachou D, Wang L, Wei W, Zheng L, Zou Z, Severson DW, Raikhel AS, Kafatos FC, Dimopoulos G, Zdobnov EM, Christophides GK, 2007. Evolutionary dynamics of immune-related genes and pathways in disease-vector mosquitoes. *Science*, 316(5832): 1738–1743.
- Werner T, Liu G, Kang D, Ekengren S, Steiner H, Hultmark D, 2000. A family of peptidoglycan recognition proteins in the fruit fly *Drosophila melanogaster*. *Proceedings of the National Academy of Sciences of the United States of America*, 97(25): 13772–13777.
- Williams MJ, 2007. *Drosophila* hemopoiesis and cellular immunity. *The Journal of Immunology*, 178(8): 4711–4716.
- Wu TY, Zhao Y, Wang ZY, Song QS, Wang ZX, Xu QW, Wang YJ, Wang LB, Zhang YQ, Feng CJ, 2018. β -1,3-Glucan recognition

- protein 3 activates the prophenoloxidase system in response to bacterial infection in *Ostrinia furnacalis* Guenee. *Developmental and Comparative Immunology*, 79: 31–43.
- Xia X, You M, Rao XJ, Yu XQ, 2018. Insect C-type lectins in innate immunity. *Developmental and Comparative Immunology*, 83: 70–79.
- Xia XF, Yu LY, Xue MQ, Yu XQ, Vasseur L, Gurr GM, Baxter SW, Lin HL, Lin JH, You MS, 2015. Genome-wide characterization and expression profiling of immune genes in the diamondback moth, *Plutella xylostella* (L.). *Scientific Reports*, 5: 9877.
- Xiao JH, Yue Z, Jia LY, Yang XH, Niu LH, Wang Z, Zhang P, Sun BF, He SM, Li Z, Xiong TL, Xin W, Gu HF, Wang B, Werren JH, Murphy RW, Wheeler D, Niu LM, Ma GC, Tang T, Bian SN, Wang NX, Yang CY, Wang N, Fu YG, Li WZ, Yi SV, Yang XY, Zhou Q, Lu CX, Xu CY, He LJ, Yu LL, Chen M, Zheng Y, Wang SW, Zhao S, Li YH, Yu YY, Qian XJ, Cai Y, Bian LL, Zhang S, Wang JY, Yin Y, Xiao H, Wang GH, Yu H, Wu WS, Cook JM, Wang J, Huang DW, 2013. Obligate mutualism within a host drives the extreme specialization of a fig wasp genome. *Genome Biology*, 14(12): R141.
- Xiong GH, Xing LS, Lin Z, Saha TT, Wang C, Jiang H, Zou Z, 2015. High throughput profiling of the cotton bollworm *Helicoverpa armigera* immunotranscriptome during the fungal and bacterial infections. *BMC Genomics*, 16(1): 321.
- Xu PZ, 2010. Study on the mechanism of innate immune pattern recognition, signal transduction and antimicrobial peptide genes expressional regulation in silkworm, *Bombyx mori*. Doctoral dissertation. Chongqing: Southwest University. [许平震, 2010. 家蚕先天免疫的模式识别信号转导和抗微生物肽基因表达调控机制研究. 博士学位论文. 重庆: 西南大学.]
- Yang MC, Shi XZ, Yang HT, Sun JJ, Xu L, Wang XW, Zhao XF, Wang JX, 2016. Scavenger receptor C mediates phagocytosis of white spot syndrome virus and restricts virus proliferation in shrimp. *PLoS Pathog*, 12(12): e1006127.
- Yoshida H, Kinoshita K, Ashida M, 1996. Purification of a peptidoglycan recognition protein from hemolymph of the silkworm, *Bombyx mori*. *Journal of Biological Chemistry*, 271(23): 13854–13860.
- Yu XQ, Kanost MR, 2003. *Manduca sexta* lipopolysaccharide-specific immulectin-2 protects larvae from bacterial infection. *Developmental & Comparative Immunology*, 27(3): 189–196.
- Yu XQ, Kanost MR, 2004. Immulectin-2, a pattern recognition receptor that stimulates hemocyte encapsulation and melanization in the tobacco hornworm, *Manduca sexta*. *Developmental and Comparative Immunology*, 28(9): 891–900.
- Yu XQ, Zhu YF, Ma C, Fabrick JA, Kanost MR, 2002. Pattern recognition proteins in *Manduca sexta* plasma. *Insect Biochemistry and Molecular Biology*, 32(10): 1287–1293.
- Yu XQ, Jiang HB, Wang Y, Kanost MR, 2003. Nonproteolytic serine proteinase homologs are involved in prophenoloxidase activation in the tobacco hornworm, *Manduca sexta*. *Insect Biochemistry and Molecular Biology*, 33(2): 197–208.
- Zeng LY, Li ZH, Liu LJ, 2019. Research progress in the immunity of insects and the immune mechanisms of five important invasive insects. *Acta Phytophylacica Sinica*, 46(1): 6–16. [曾令瑜, 李志红, 柳丽君, 2019. 昆虫免疫及五种重要入侵昆虫免疫机制研究进展. 植物保护学报, 46(1): 6–16.]
- Zhan MY, Shahzad T, Yang PJ, Liu S, Yu XQ, Rao XJ, 2016. A single-CRD C-type lectin is important for bacterial clearance in the silkworm. *Developmental and Comparative Immunology*, 65: 330–339.
- Zhang X, He Y, Cao X, Gunaratna RT, Chen YR, Blissard G, Kanost MR, Jiang H, 2015. Phylogenetic analysis and expression profiling of the pattern recognition receptors: Insights into molecular recognition of invading pathogens in *Manduca sexta*. *Insect Biochemistry and Molecular Biology*, 62: 38–50.
- Zou Z, Evans JD, Lu ZQ, Zhao PC, Williams M, Sumathipala N, Hetru C, Hultmark D, Jiang HB, 2007. Comparative genomic analysis of the *Tribolium* immune system. *Genome Biology*, 8(8): R177.