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12种杀菌剂制剂对半闭弯尾姬蜂的急性毒性及安全性评价

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摘要:采用试管药膜法测定了12种杀菌剂制剂对半闭弯尾姬蜂成蜂的急性接触毒性,结合安全性系数评价了供试药剂对半闭弯尾姬蜂的安全性。急性毒性测定结果表明,氟硅唑和啶菌噁唑对半闭弯尾姬蜂成蜂的接触毒性最高,LC₅₀值分别为220.022和223.115 mg·L⁻¹;其次为多抗霉素、丙森锌和戊唑醇,LC₅₀值分别为436.496、472.358和638.638 mg·L⁻¹;其余7种杀菌剂制剂嘧霉胺、啶酰菌胺、苯醚甲环唑、多菌灵、嘧菌酯、嘧菌环胺和异菌脲对半闭弯尾姬蜂成蜂的触杀毒性都较低,LC₅₀值均大于1 000 mg·L⁻¹。安全性评价结果表明,丙森锌和啶菌噁唑对半闭弯尾姬蜂成蜂具有高风险性,安全系数分别为0.16和0.42;嘧霉胺、多抗霉素、氟硅唑和戊唑醇为中等风险性,安全系数分别为1.43、1.56、2.20和3.23;其余6种杀菌剂制剂啶酰菌胺、苯醚甲环唑、多菌灵、嘧菌酯、嘧菌环胺和异菌脲对半闭弯尾姬蜂表现为低风险性,安全系数均大于5。结果显示:甾醇脱甲基抑制剂氟硅唑、戊唑醇和啶菌噁唑以及苯胺基嘧啶类杀菌剂嘧霉胺、有机硫杀菌剂丙森锌和抗菌素多抗霉素对半闭弯尾姬蜂成蜂具有急性毒性风险,在有害生物综合治理中应谨慎使用,特别是啶菌噁唑和丙森锌,以免对半闭弯尾姬蜂造成不良影响和危害。

关键词:杀菌剂;半闭弯尾姬蜂;急性毒性;安全性评价

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Acute Toxicity and Safety Evaluation of 12 Fungicide Formulations to *Diadegma semiclausum* (Hymenoptera: Ichneumonidae)

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Abstract: The acute contact toxicities of 12 fungicide formulations to adults of *Diadegma semiclausum* were tested with a test tube residue method. The obtained LC₅₀ values were used to calculate the safety coefficients with the formula of LC₅₀/recommended field concentration, which in turn were used to assess the risks of the 12 fungicides to the wasp. Contact toxicity bioassay showed that flusilazole and SYP-Z048 had the highest contact toxicity to adults of *D. semiclausum*, with a LC₅₀ of 220.022 and 223.115 mg·L⁻¹, respectively, followed by polyoxin, propineb and tebuconazole, with a LC₅₀ of 436.496, 472.358 and 638.638 mg·L⁻¹, respectively. The other 7 fungicides, inclu-

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ding pyrimethanil, boscalid, difenoconazole, carbendazim, azoxystrobin, ciprodinil and iprodione, had a LC_{50} of $> 1000 \text{ mg} \cdot \text{L}^{-1}$ and were the least toxic to adults of *D. semiclausum*. Safety evaluation revealed that propineb and SYP-Z048 had a safety coefficient of 0.16 and 0.42, respectively, and thus were of high risk to the wasp. Pyrimethanil, polyoxin, flusilazole and tebuconazole were of moderate risk to the wasp since their safety coefficients were 1.43, 1.56, 2.20 and 3.23, respectively. Boscalid, difenoconazole, carbendazim, azoxystrobin, ciprodinil and iprodione had a safety coefficient of > 5 and thus were of low risk to the wasp. Taken together, these results suggest that the sterol demethylation inhibitors flusilazole, tebuconazole and SYP-Z048, the aniline pyrimidine fungicide pyrimethanil, the organic sulfur fungicide propineb and the antibiotics polyoxin, especially SYP-Z048 and propineb, have an acute toxicity risk to adults of *D. semiclausum* and thus should be used with caution in integrated pest management programs in order to avoid their adverse effects on *D. semiclausum*.

Keywords: fungicides; *Diadegma semiclausum*; acute toxicity; safety assessment

半闭弯尾姬蜂(*Diadegma semiclausum* Hellen)属膜翅目姬蜂科,是小菜蛾(*Plutella xylostella*)幼虫期的一种重要的内寄生蜂,起源于欧洲,被许多国家和地区引进用于防治小菜蛾,对小菜蛾的种群数量和危害起到了很好的控制作用^[1-3]。

施用到田间的药剂作为环境因子被各种生物频繁接触,对非靶标生物特别是天敌构成威胁,长期暴露在药剂选择压力下会严重降低天敌的控害潜能,并且带来诸多生态问题^[4-5]。生物防治缓效、低效的缺点决定了目前化学药剂不可能被完全禁止,如何协调化学防治和生物防治是药剂对天敌安全性评价的主要目的,筛选高效低毒的化学药剂成为有害生物综合治理(IPM)研究的关键性问题^[6]。

在生产中,一般考虑较多的是杀虫剂对寄生蜂等天敌的毒性,杀菌剂对天敌的风险经常被忽略^[7]。有研究表明,一些杀菌剂对寄生蜂有害,如异稻·三环唑和苯甲·丙环唑对二化螟盘绒茧蜂雌成蜂具有较高毒性^[7],三唑酮、苯醚甲环唑、氟环唑、嘧菌酯、百菌清等杀菌剂对某些赤眼蜂品种具有较高毒性风险^[8-12]。有关杀虫剂对半闭弯尾姬蜂的毒性风险已有许多研究^[13-17],但杀菌剂对半闭弯尾姬蜂的毒性研究较少。为明确杀菌剂对半闭弯尾姬蜂的毒性,本研究评价了12种杀菌剂制剂对半闭弯尾姬蜂成蜂的安全性,以期为合理用药、保护和利用天敌以及协调化学防治和生物防治提供有益参考。

1 材料与方法(Materials and methods)

1.1 供试材料

1.1.1 供试昆虫

半闭弯尾姬蜂采集于山西农业大学农作站未施药甘蓝田,在室内以多年未接触药剂饲养的小菜蛾3龄幼虫供其寄生。饲养条件:温度(22±1)℃,相对

湿度(RH)70%,光周期14 h L:10 h D。

1.1.2 供试药剂

供试药剂相关信息见表1。

1.2 试验方法

参考贾变桃等^[17]的方法,采用试管药膜法测定药剂对半闭弯尾姬蜂成蜂的触杀毒性。在预备试验的基础上,将不同药剂分别配制5~7个试验浓度梯度,以清水作为对照。所有测定药液和对照均加入0.02%(体积分数)的Triton X-100。将配好的药液倒满2 cm×8 cm指形管,静置10 s后倒出,室温下倒置晾干形成药膜。每管接入羽化1~2 d的半闭弯尾姬蜂6~8头,管底放置10%(质量分数)蔗糖水棉花球供成蜂取食。用双层纱布将管口蒙住,橡皮筋扎紧。每处理重复4次。置于温度(22±1)℃,RH 70%,光周期14 h L:10 h D的光照培养箱中。24 h后观察成蜂死亡情况,以蜂不能正常爬行或飞行为死亡标准。

1.3 数据处理

采用PoloPlus 1.0软件进行统计分析,计算各药剂对半闭弯尾姬蜂成蜂的斜率(b)±标准误、 LC_{50} 值及95%置信限。

安全系数计算公式:安全系数= $LC_{50}(\text{mg} \cdot \text{L}^{-1}) \div \text{田间推荐使用剂量}(\text{mg} \cdot \text{L}^{-1})$ 。参考俞瑞鲜等^[18]的方法,农药对半闭弯尾姬蜂的风险性分级标准为:安全系数 > 5 为低风险性,0.5<安全系数≤5为中等风险性,0.05<安全系数≤0.5为高风险性,安全系数≤0.05为极高风险性。

2 结果(Results)

2.1 12种杀菌剂制剂对半闭弯尾姬蜂成蜂的触杀毒性

由表2可以看出,12种杀菌剂制剂中氟硅唑和

表 1 供试杀菌剂及田间推荐浓度

Table 1 Tested fungicides and their field recommended concentrations

杀菌剂 Fungicide	有效成分 Active ingredient	生产厂家 Manufacturer	田间推荐浓度/(mg·L ⁻¹) Recommended field concentration/(mg·L ⁻¹)
400 g·L ⁻¹ 福星乳油	氟硅唑	美国杜邦公司	100
400 g·L ⁻¹ Nustar EC	Flusilazole	DuPont	
10%世高水分散粒剂	苯醚甲环唑	先正达作物保护有限公司	50
10% Score WG	Difenoconazole	Syngenta	
430 g·L ⁻¹ 好克利悬浮剂	戊唑醇	标正作物科学有限公司	197.8
430 g·L ⁻¹ Haokeli SC	Tebuconazole	Biogen	
40% 恢典悬浮剂	嘧霉胺	标正作物科学有限公司	720
40% Huidian SC	Pyrimethanil	Biogen	
50% 瑞镇水分散粒剂	嘧菌环胺	先正达作物保护有限公司	960
50% Ruizhen WG	Cyprodinil	Syngenta	
10% 宝丽安可湿性粉剂	多抗霉素	日本科研制药株式会社	280
10% Baoli'an WP	Polyoxin	Kaken Pharmaceutical	
25% 菌思奇乳油	啶菌噁唑	沈阳科创化学品有限公司	535
25% Junsiqi EC	SYP-Z048	Shenyang Sciencreat Chemicals	
250 g·L ⁻¹ 阿米西达悬浮剂	嘧菌酯	先正达作物保护有限公司	500
250 g·L ⁻¹ Amistar SC	Azoxystrobin	Syngenta	
70% 安泰生可湿性粉剂	丙森锌	德国拜耳作物科学公司	2 996
70% Antracol WP	Propineb	Bayer	
25% 菌立安可湿性粉剂	多菌灵	陕西汤普森生物科技有限公司	1 000
25% Junli'an WP	Carbendazim	Shaanxi Tupsion	
50% 凯泽水分散粒剂	啶酰菌胺	德国巴斯夫植物保护有限公司	500
50% Cantus WG	Boscalid	Basf	
25.5% 统秀可湿性粉剂	异菌脲	标正作物科学有限公司	2 000
25.5% Tongxiu WP	Iprodione	Biogen	

表 2 12 种杀菌剂制剂对半闭弯尾姬蜂成蜂的急性毒性

Table 1 Acute toxicity of 12 fungicide preparations to adults of *D. semiclausum*

杀菌剂 Fungicide	斜率±标准误 Slope ± SE	LC ₅₀ (95% 置信限)/(mg·L ⁻¹) LC ₅₀ (95% fiducial limit)/(mg·L ⁻¹)	χ ²	自由度 Degree of freedom (Df)
氟硅唑 Flusilazole	1.762±0.406	220.022(137.200~336.287)	0.244	3
啶菌噁唑 SYP-Z048	1.222±0.173	223.115(141.001~350.961)	1.902	3
多抗霉素 Polyoxin	1.310±0.238	436.496(289.900~668.825)	1.592	4
丙森锌 Propineb	1.812±0.309	472.358(306.568~705.100)	0.733	2
戊唑醇 Tebuconazole	0.934±0.127	638.638(261.470~1 709.171)	5.1014	4
嘧霉胺 Pyrimethanil	0.969±0.154	1 031.298(310.230~5 878.122)	4.5065	3
啶酰菌胺 Boscalid	1.934±0.352	6 193.148(4 472.420~8 593.182)	0.335	3
苯醚甲环唑 Difenoconazole	-	>1 000	-	-
多菌灵 Carbendazim	-	>20 000	-	-
嘧菌酯 Azoxystrobin	-	>2 500	-	-
嘧菌环胺 Cyprodinil	-	>5 000	-	-
异菌脲 Iprodione	-	>20 400	-	-

啶菌噁唑对半闭弯尾姬蜂成蜂的接触毒性最高,LC₅₀值分别为220.022和223.115 mg·L⁻¹;其次为多抗霉素、丙森锌和戊唑醇,LC₅₀值分别为436.496、472.358和638.638 mg·L⁻¹;其余7种杀菌剂嘧霉胺、啶酰菌胺、苯醚甲环唑、多菌灵、嘧菌酯、嘧菌环胺和异菌脲对半闭弯尾姬蜂成蜂的触杀毒性都较低,LC₅₀值均大于1 000 mg·L⁻¹。

2.2 12种杀菌剂制剂的安全性等级

测定的12种杀菌剂制剂中,丙森锌和啶菌噁唑的安全系数最低,分别为0.16和0.42,对半闭弯尾姬蜂成蜂具有高风险;嘧霉胺、多抗霉素、氟硅唑和戊唑醇安全系数分别为1.43、1.56、2.20和3.23,这4种杀菌剂对半闭弯尾姬蜂具有中等风险;其余6种杀菌剂啶酰菌胺、苯醚甲环唑、多菌灵、嘧菌酯、嘧菌环胺和异菌脲安全系数均大于5,对半闭弯尾姬蜂成蜂表现为低风险(表3)。

3 讨论(Discussion)

半闭弯尾姬蜂是一种容性寄生蜂,其产卵到小菜蛾幼虫体内,幼虫在寄主体内发育,直到寄主做茧进入预蛹阶段才完成幼虫期发育,消耗完寄主的所有内含物,在寄主的丝茧内结茧化蛹^[19-20]。由于寄生蜂卵和幼虫在寄主体内完成发育,寄主表皮对其具有保护作用,蜂蛹被茧包围,避免与外界接触,只有成虫期暴露在外,在取食花蜜或寻找寄主等活动中食入或接触药剂,因此对药剂最敏感。一般来说,

表3 12种杀菌剂制剂对半闭弯尾姬蜂成蜂的安全性评价
Table 3 Safety evaluation of 12 fungicide preparations to adults of *D. semiclausum*

杀菌剂 Fungicide	安全系数 Safety coefficient	安全性等级 Safety grade
氟硅唑 Flusilazole	2.20	中等风险 Moderate risk
啶菌噁唑 SYP-Z048	0.42	高风险 High risk
多抗霉素 Polyoxin	1.56	中等风险 Moderate risk
丙森锌 Propineb	0.16	高风险 High risk
戊唑醇 Tebuconazole	3.23	中等风险 Moderate risk
嘧霉胺 Pyrimethanil	1.43	中等风险 Moderate risk
啶酰菌胺 Boscalid	12.39	低风险 Low risk
苯醚甲环唑 Difenconazole	>10.00	低风险 Low risk
多菌灵 Carbendazim	>5.00	低风险 Low risk
嘧菌酯 Azoxystrobin	>5.56	低风险 Low risk
嘧菌环胺 Cyprodinil	>5.20	低风险 Low risk
异菌脲 Iprodione	>10.20	低风险 Low risk

应选择天敌最敏感的生育期进行药剂的安全性评价^[11],因此我们选择对半闭弯尾姬蜂成蜂开展毒性测定及安全性评价研究。

在测定的12种杀菌剂中,氟硅唑、戊唑醇和苯醚甲环唑为三唑类脱甲基抑制剂,这一类杀菌剂某些品种对寄生性天敌毒性较高,如三唑酮对玉米螟赤眼蜂(*Trichogramma ostriniae*)具有极高风险性,对松毛虫赤眼蜂(*T. dendrolimi*)和稻螟赤眼蜂(*T. japonicum*)具有高风险性^[8-9];氟环唑对稻螟赤眼蜂、拟澳洲赤眼蜂(*T. confusum*)和亚洲玉米螟赤眼蜂均具有高风险性^[11]。尽管本研究中氟硅唑对半闭弯尾姬蜂成蜂为中等风险性,但这一杀菌剂对其他非靶标生物如大型溞(*Daphnia magna*)为剧毒^[21]、意大利蜜蜂(*Apis mellifera L.*)为高毒^[22]、斑马鱼(*Brachydanio rerio*)为中毒^[23]、家蚕(*Bombyx mori L.*)为中毒且具有高风险性^[24],表明这一杀菌剂对非靶标生物具有一定的风险,应谨慎使用。戊唑醇对半闭弯尾姬蜂成蜂为中等风险性,但其对稻螟赤眼蜂、拟澳洲赤眼蜂和亚洲玉米螟赤眼蜂为低风险^[11]。苯醚甲环唑对半闭弯尾姬蜂为低风险性,同样其对稻螟赤眼蜂和亚洲玉米螟赤眼蜂也为低风险性药剂,但对拟澳洲赤眼蜂具有中等风险性^[11]。可见,同种药剂对不同种类寄生蜂的毒性与安全性不同,同一种寄生蜂对不同药剂的反应也有差异。啶菌噁唑是由沈阳化工研究院自主研发的一种新型杀菌剂^[25],其作用机制与三唑类杀菌剂一样,抑制真菌麦角甾醇生物合成,作用靶标为C¹⁴- α 甾醇脱甲基酶^[26-27]。本研究表明啶菌噁唑对半闭弯尾姬蜂高风险,李肇丽等^[28]报道啶菌噁唑对松毛虫赤眼蜂中等风险,说明这一杀菌剂对寄生性天敌有一定的毒害作用。

多抗霉素是由金色链霉菌产生的肽嘧啶核苷类抗菌素,具有广谱杀菌活性,作用机理为干扰病菌细胞壁的生物合成,低毒低残留,对水生生物和蜜蜂低毒^[29],被认为是一种安全的绿色农药,但本研究测定其对半闭弯尾姬蜂成蜂为中等风险。

丙森锌属有机硫类保护性杀菌剂,李健宇等^[30]报道称其对环境生物鹌鹑(*Coturnix japonica*)、蜜蜂和蚯蚓(*Eisenia foetida*)低毒,但对家蚕剧毒,对斑马鱼和大型溞中毒。本研究也表明其对半闭弯尾姬蜂成蜂毒性较大,具有高风险性。

嘧霉胺和嘧菌环胺属苯胺基嘧啶类杀菌剂,作用机理是抑制氨基酸甲硫氨酸的合成,与其他类杀菌剂无交互抗性^[31]。据报道嘧霉胺对环境生物蜜

蜂、鹌鹑、家蚕、斑马鱼、泽蛙(*Rana limnocharis*)和蚯蚓均为低毒^[32],本研究表明该药对半闭弯尾姬蜂成蜂具有中等毒性风险,但嘧菌环胺为低风险。

杀菌剂对非靶标生物的作用机制尚不明确,可能与药剂影响了虫体内某些酶的活性有关。如金华超等^[33]研究认为三唑酮、肟菌脂、咪鲜胺和申嗪霉素明显激活玉米螟赤眼蜂酚氧化酶的活性。

本研究结果表明,12种杀菌剂中,啶菌噁唑和丙森锌对半闭弯尾姬蜂成虫具有高风险性,氟硅唑、戊唑醇、多抗霉素和嘧霉胺为中等风险性,其余6种杀菌剂为低风险性。因此,为保护半闭弯尾姬蜂,充分发挥其对小菜蛾的自然控制能力,在十字花科蔬菜田应严格禁止使用啶菌噁唑和丙森锌,谨慎使用戊唑醇、氟硅唑、多抗霉素和嘧霉胺。试管药膜法较好地模拟了施药后的蔬菜叶片,半闭弯尾姬蜂成蜂在寻找栖息场所或搜寻小菜蛾幼虫的过程中,在蔬菜叶片上爬行从而接触药剂,但并不能完全模拟药剂进入虫体的途径,如经口器吃入药剂等;此外,成蜂在田间感药后可以飞避施药区,而试管内空间相对比较封闭,因此本研究测得的毒性结果可能比田间实际结果要高。此外,本研究仅评价了12种杀菌剂制剂对半闭弯尾姬蜂成蜂的毒性及安全性,要全面详细评价这几种药剂对半闭弯尾姬蜂的安全性,还需测定它们对该寄生蜂其他虫态的毒性以及开展相关田间试验和生化测定等。

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