

杀虫剂亚致死浓度对普通大蓟马繁殖的影响*

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摘 要 【目的】在中国海南省, 普通大蓟马 *Megalurothrips usitatus* (Bagrall) 是豇豆 *Vigna unguiculata* (L.) Walp. 上的重要害虫, 化学防治还是重要的控制手段。本研究旨在了解 3 种杀虫剂亚致死浓度对该虫繁殖的影响。【方法】在 $(26 \pm 1)^\circ\text{C}$ 、相对湿度 $60\% \pm 5\%$ 、光周期 $L:D = 14:10$ 条件下, 采用叶盘浸药法测定溴氰虫酰胺、双甲脒和茚虫威 48 h 内对普通大蓟马 1 日龄已交配雌虫的毒杀效果。经杀虫剂 LC_{20} 处理的幼嫩豇豆叶叶盘供已交配雌虫和未交配雌虫取食 48 h 后, 改用未浸药叶盘继续饲养。每天观察雌虫所产子代的卵数和成虫数, 计算已交配雌虫的子代成虫性比和卵性比。【结果】3 种杀虫剂 LC_{20} 处理均使亲代雌虫的存活时间显著缩短。溴氰虫酰胺和双甲脒 LC_{20} 处理还能显著降低已交配亲代雌虫在胁迫处理后所产子代的成虫数量。亲代雌虫经溴氰虫酰胺、双甲脒和茚虫威 LC_{20} 处理后的子代成虫性比分别为 0.60、0.47 和 0.41, 与清水对照 (0.52) 之间的差异均未达显著水平; 卵性比在各处理之间都没有显著差异。【结论】溴氰虫酰胺、双甲脒和茚虫威 LC_{20} 处理对普通大蓟马亲代雌虫的存活时间和子代成虫数量有不同程度的负面影响, 但对子代性别分配均没有显著的干扰作用。

关键词 普通大蓟马; 溴氰虫酰胺; 双甲脒; 茚虫威; 性比; 豇豆

Sublethal effect of insecticides on reproduction of *Megalurothrips usitatus* (Bagrall) (Thysanoptera: Thripidae)

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Abstract [Objectives] The aim of this paper is to explore the sublethal effects of three insecticides on reproduction of bean flower thrips [*Megalurothrips usitatus* (Bagrall)], one of the most economically important insect pests of cowpea [*Vigna unguiculata* (L.) Walp.] in Hainan Island, China. [Methods] Under laboratory conditions of $(26 \pm 1)^\circ\text{C}$, $60\% \pm 5\%$ R.H. and a photoperiod of 14L:10D, with young leaves of cowpea (var. Chunfeng-changjiangdou) as food of the bean flower thrips, the lethal and sublethal concentrations of cyantraniliprole, amitraz, and indoxacarb for the 1-d-old mated adult females were determined using a leaf-disc dipping bioassay method. After 48 h exposure to LC_{20} concentration of each insecticide, the surviving mated or unmated adult females were provided with non-treated leaf discs. The number of eggs and subsequent adult offspring were counted daily until the parental females died. Sex ratios of adult offspring produced by mated females were calculated as proportion of males, and then sex ratios of eggs laid by them were estimated using survival rates of the offspring produced by both mated females and unmated females. [Results] Exposure to three insecticides at LC_{20} concentration all significantly decreased the survival duration of either mated or unmated females. The number of adult offspring produced by mated females exposed to cyantraniliprole and amitraz were also significantly reduced. Sex ratios of adult offspring produced by mated females following sublethal exposure to cyantraniliprole, amitraz, and indoxacarb were 0.60, 0.47, and 0.41, respectively, which all were not significantly different from that of control. There was no significant difference in sex ratio of eggs laid by mated females among insecticide treatments and control. [Conclusion] Sublethal exposure to cyantraniliprole,

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amitraz, and indoxacarb decreases to some extent the survival duration of the parental females of the bean flower thrips, and the number of adult offspring produced by them, but does not significantly change their sex allocation.

Key words *Megalurothrips usitatus*; cyantraniliprole; amitraz; indoxacarb; sex ratio; cowpea

普通大蓟马 *Megalurothrips usitatus* (Bagnall) 又称豆花蓟马、豆大蓟马, 隶属缨翅目 Thysanoptera、蓟马科 Thripidae、大蓟马属 *Megalurothrips*, 是豆类植物上的一种重要害虫 (Chang, 1988; 邱海燕等, 2014; 谭珂等, 2015a; Tang *et al.*, 2015)。其若虫和成虫为害嫩芽、花朵和嫩荚, 导致植株生长停滞、叶片萎缩与畸形、落花落荚等, 严重影响豆类的产量和品质 (范咏梅等, 2013; 谭珂等, 2015a, 2015b)。在我国海南, 该虫 1 年可发生 24-26 代 (邱海燕等, 2014), 现已成为豇豆 *Vigna unguiculata* (L.) Walp. 的头号害虫, 其治理高度依赖化学防治 (唐良德等, 2016)。

在田间, 杀虫剂按推荐剂量施用后, 其浓度会逐步下降到亚致死水平。杀虫剂亚致死浓度对靶标害虫和非靶标生物的生长发育、存活、繁殖和行为仍有不同程度的影响, 可呈现出抑制作用、促进作用或无影响。杀虫剂的亚致死效应与昆虫种类和杀虫剂类型密切相关 (Desneux *et al.*, 2007; 韩文素等, 2011; de França *et al.*, 2017; Bantz *et al.*, 2018; Müller, 2018)。例如, 不同杀虫剂对西花蓟马 *Frankliniella occidentalis* (Pergande) 的亚致死效应是有差异的 (Bielza and Guillen, 2015; Gong *et al.*, 2015; Zhang *et al.*, 2015; 杨广明等, 2016; Cao *et al.*, 2019)。迄今, 有关杀虫剂亚致死浓度对普通大蓟马的影响尚未见报道。本文拟评价 3 种不同作用机制的杀虫剂对普通大蓟马雌虫的亚致死效应, 以明确亲代雌虫的存活时间、性别分配及子代数量与存活是否因杀虫剂的不同而发生变化。

1 材料与方法

1.1 供试虫源

在海南大学海甸校区植物保护学院教学实验基地, 采集取食豇豆花朵的普通大蓟马成虫, 并带回室内以幼嫩豇豆豆荚 (品种: 纯丰长豇豆)

饲养, 饲养条件设定为 (26 ± 1) °C、相对湿度 $60\% \pm 5\%$ 、光周期 L : D = 14 : 10。待化蛹后, 挑取健康的蛹, 单头饲养于玻璃试管 (12 mm × 100 mm) 内。成虫羽化后, 立即按性比 1 : 1 进行配对饲养。部分刚羽化的雌虫仍继续单头饲养。最后, 挑选已交配或未交配的 1 日龄雌虫作为试虫。

1.2 供试植物

在网室大棚内, 盆栽种植豇豆 (品种: 纯丰长豇豆), 按常规栽培措施进行管理。在苗龄 15 d 的植株上选取已展开的健康嫩叶作为供试叶片。

1.3 供试杀虫剂

选用豇豆害虫防治中常用的 3 种杀虫剂: 美国杜邦公司出产的 10% 溴氰虫酰胺乳油 (杜邦-倍内威); 爱利思达生命科学株式会社出产的 200 g/L 双甲脒可分散油悬浮剂 (满柯-双甲脒) 和 150 g/L 茚虫威乳油 (杜邦-凯恩)。

1.4 试验方法

1.4.1 生物测定 采用叶盘浸药法测定 3 种杀虫剂对已交配和普通大蓟马雌虫的毒杀效果。每种杀虫剂设 6 个水平, 以清水为对照, 每水平重复 4 次。用直径为 20 mm 的打孔器切取叶盘, 将叶盘浸入已配好的药液中 10 s, 取出晾干后, 转移到垫有湿润滤纸的培养皿 (直径 90 mm) 内, 叶盘背面朝上, 每皿放置 2 个叶盘。然后接入已交配雌虫 (30 头/皿), 并用扎有小孔的保鲜膜封口。所有培养皿置于 (26 ± 1) °C、相对湿度 $60\% \pm 5\%$ 、光周期 L : D = 14 : 10 条件下。48 h 后, 检查雌虫的存活状况。以毛笔笔尖触碰后, 不动者即视为死亡。建立毒力回归方程, 计算 LC_{50} 和 LC_{20} 。

1.4.2 杀虫剂 LC_{20} 胁迫试验 每种杀虫剂按 LC_{20} 配制药液。将小叶盘 (直径 5 mm) 浸入已

配好的药液中 10 s, 取出晾干备用。用清水处理的小叶盘作为对照。试验条件与方法参照上述生物测定, 不同之处在于: 小培养皿(直径 35 mm)内放置 1 个小叶盘, 每皿接入 1 头试虫。每个处理的已交配雌虫和未交配雌虫均为 45 头。48 h 后, 将仍存活的雌虫移到可透气的产卵盒(直径 35 mm×高 10 mm)中进行单头饲养, 每笼放置 1 个未经药液处理的小叶盘, 每 24 h 更换一次小叶盘, 直至雌虫死亡。将每次更换出来的小叶盘放在体视显微镜下检查卵粒数, 然后再移入垫有湿润滤纸和未药液处理的叶盘(直径 12 mm)的培养皿中继续饲养。每天检查羽化出来的雌虫和雄虫数量, 并适时补充未经药液处理的叶盘。计算每头亲代雌虫在 LC₂₀ 胁迫后的存活时间、总卵量、子代成虫数、子代从卵至羽化时的存活率, 以及已交配亲代雌虫所产子代成虫的性比(即雄性占成虫比例)。已交配的普通大蓟马雌虫产受精卵和未受精卵, 分别发育成雌性和雄性后代; 未交配雌虫仅产未受精卵, 全为雄性后代。假设已交配雌虫所产雄性后代的存活率与未交配雌虫的后代存活率相等, 可利用已交配亲代雌虫所产子代成虫性比来估算子代卵性比, 即文献中所指的初级性比(Primary sex ratio), 可用来表征性别分配(Jarošík *et al.*, 2003; Bondy and Hunter,

2019)。卵性比的估算公式为:

$$R_e = R_a \times S_m / S_u$$

其中, R_e 为已交配雌虫所产子代卵性比; R_a 为已交配雌虫所产子代成虫性比; S_m 为已交配雌虫所产子代从卵至羽化时的存活率; S_u 为未交配雌虫所产子代从卵至羽化时的存活率平均值。

1.5 数据分析

利用 Excel 2010 进行试验数据处理和绘图; 利用 SPSS 19.0 软件进行试验数据的统计分析和毒力方程参数的估算。存活率和性比数据经反正弦转换后, 进行方差分析(ANOVA)和多重比较(Duncan's 多重比较法, $P<0.05$)。

2 结果与分析

2.1 杀虫剂 LC₅₀ 和 LC₂₀

采用叶盘浸药法测定了溴氰虫酰胺、双甲脒和茚虫威 48 h 内对已交配的 1 日龄普通大蓟马雌虫的毒杀效果(表 1), 其 LC₅₀ 分别为 13 504.267、7 196.336、2 663.735 mg/L, 而 LC₂₀ 分别为 4 868.716、3 903.815、962.476 mg/L。3 种杀虫剂对普通大蓟雌成虫的毒力依次为: 茚虫威>双甲脒>溴氰虫酰胺。

表 1 杀虫剂对已交配的普通大蓟马雌虫的毒力
Table 1 Toxicity of insecticides to the mated adult females of *Megalurothrips usitatus*

杀虫剂 Insecticides	回归方程 Regression equation	LC ₅₀ (mg/L) (95% CI)	LC ₂₀ (mg/L) (95% CI)
溴氰虫酰胺 Cyantraniliprole	$y = -7.846 + 1.900x$	13 504.267 (10 392.269 – 18 507.601)	4 868.716 (3 017.772 – 6 591.165)
双甲脒 Amitraz	$y = -12.221 + 3.168x$	7 196.336 (5 388.816 – 8 825.297)	3 903.815 (2 216.502 – 5 249.207)
茚虫威 Indoxacarb	$y = -6.521 + 1.904x$	2 663.735 (2 117.826 – 3 285.814)	962.476 (621.042 – 1 294.962)

2.2 杀虫剂 LC₂₀ 处理后亲代雌虫的存活时间

3 种杀虫剂 LC₂₀ 胁迫处理 48 h 对已交配和未交配的普通大蓟马雌虫的存活时间均有显著($P<0.05$)的影响(表 2)。溴氰虫酰胺、双甲脒和茚虫威 LC₂₀ 处理后的已交配雌虫可存活

12.37 - 13.13 d, 彼此之间无显著差异, 但均显著($P<0.05$)短于对照(18.26 d)。同样地, 3 种杀虫剂处理也导致未交配雌虫的存活时间显著缩短($P<0.05$)。经溴氰虫酰胺、双甲脒和茚虫威 LC₂₀ 处理后未交配雌虫的存活时间分别为 8.03、11.30 和 9.88 d, 其中溴氰虫酰胺处理和双

甲胺处理之间的差异达到了显著水平 ($P<0.05$)。

表 2 杀虫剂亚致死浓度对普通大蓟马亲代雌成虫存活时间的影响
Table 2 Survival time of adult females of *Megalurothrips usitatus* following 48 h sublethal exposure to insecticides

处理 Treatments	存活时间 (d) Survival time (d)	
	已交配雌虫 Mated females	未交配雌虫 Unmated females
对照 Control	18.26 ± 0.88a	17.63 ± 0.82a
溴氰虫酰胺 Cyantraniliprole	12.37 ± 0.75b	8.03 ± 0.46c
双甲脒 Amitraz	13.43 ± 1.17b	11.30 ± 0.86b
茚虫威 Indoxacarb	13.13 ± 0.94b	9.88 ± 0.83bc

表中数值为平均数 ± 标准误, 同一列数据后标有不同字母表示在 $P<0.05$ 差异显著 (Duncan's 多重比较法)。下表同。
Data in the table are mean ± SE, and followed by different letters within the same column are significantly different at 0.05 level by Duncan's multiple range test. The same below.

2.3 杀虫剂 LC₂₀ 处理后所产子代的成虫数量和存活率

3 种杀虫剂 LC₂₀ 胁迫处理 48 h 对普通大蓟

马雌虫在存活期间所产子代的成虫数量有不同程度的抑制作用 (表 3)。溴氰虫酰胺、双甲脒和茚虫威 LC₂₀ 处理后已交配雌虫的子代成虫数分别为 3.95、3.83 和 5.25 头/雌, 彼此之间的差异未达显著水平, 但溴氰虫酰胺和双甲脒处理与对照 (5.94 头/雌) 均有显著差异 ($P<0.05$)。未交配雌虫经溴氰虫酰胺、双甲脒和茚虫威 LC₂₀ 处理后, 子代成虫数也相差无几, 介于 2.21-2.85 头/雌, 但双甲脒处理与对照 (3.66 头/雌) 之间没有显著差异。

3 种杀虫剂 LC₂₀ 胁迫处理对普通大蓟马雌虫在存活期间所产子代的存活率也有不同程度的影响 (表 3)。溴氰虫酰胺、双甲脒和茚虫威 LC₂₀ 处理后已交配雌虫所产子代的存活率分别为 54.18%、47.53% 和 58.16%, 其中溴氰虫酰胺和茚虫威处理与对照 (40.84%) 之间的差异均达到显著 ($P<0.05$) 水平。未交配雌虫经 3 种杀虫剂 LC₂₀ 胁迫处理后所产子代的存活率介于 59.33%-65.41%, 彼此之间没有显著差异, 但溴氰虫酰胺处理的子代存活率显著 ($P<0.05$) 高于对照 (47.63%)。

表 3 杀虫剂亚致死浓度处理普通大蓟马亲代雌虫后所产子代的成虫数量和存活率
Table 3 Number of adult offspring and survival of offspring produced by adult females of *Megalurothrips usitatus* following 48 h sublethal exposure to insecticides

处理 Treatments	子代成虫数 (头/雌) Number of adult offspring per female		子代存活率 (%) Offspring survival until adulthood (%)	
	已交配雌虫 Mated females	未交配雌虫 Unmated females	已交配雌虫 Mated females	未交配雌虫 Unmated females
对照 Control	5.94 ± 0.76a	3.66 ± 0.45a	40.84 ± 2.88b	47.63 ± 3.75b
溴氰虫酰胺 Cyantraniliprole	3.95 ± 0.34b	2.21 ± 0.31b	54.18 ± 4.61a	65.41 ± 5.06a
双甲脒 Amitraz	3.83 ± 0.58b	2.85 ± 0.49ab	47.53 ± 4.59ab	59.33 ± 5.04ab
茚虫威 Indoxacarb	5.25 ± 0.58ab	2.35 ± 0.30b	58.16 ± 3.77a	58.75 ± 5.59ab

2.4 已交配雌虫所产子代的成虫性比和卵性比

经 3 种杀虫剂 LC₂₀ 处理 48 h 后, 已交配的普通大蓟马雌虫所产子代成虫性比发生不同程度的改变 (图 1), 但同对照的子代成虫性比 (0.52) 之间的差异均未达到显著水平。溴氰虫酰胺、双甲脒和茚虫威处理的子代成虫性比分别

为 0.60、0.47 和 0.41, 其中, 溴氰虫酰胺和茚虫威之间存在显著差异 ($P<0.05$)。

溴氰虫酰胺、双甲脒、茚虫威处理和对照的子代卵性比分别为 0.51、0.38、0.40 和 0.43, 彼此之间无显著差异, 说明 3 种杀虫剂 LC₂₀ 处理对已交配的普通大蓟马雌虫的性别分配未表现出明显的干扰作用。

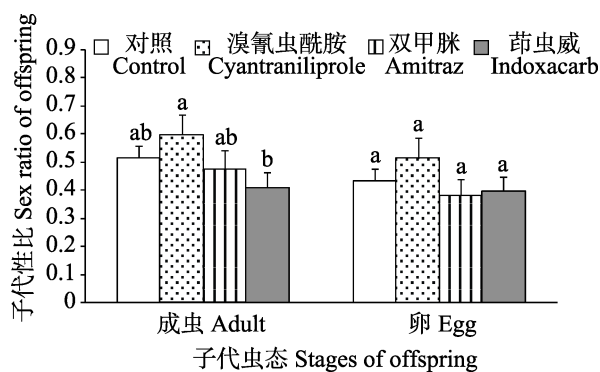


图1 杀虫剂亚致死浓度处理已交配的普通大蓟马雌虫后所产子代的成虫性比和卵性比

Fig. 1 Sex ratio of adult offspring and the estimated sex ratio of eggs laid after the mated females of *Megalurothrips usitatus* were sublethally exposed to insecticides for 48 h

图中数据为平均值 $\pm$ 标准误。柱上标有相同字母时,表示不同处理之间的差异在 $P>0.05$ 水平上未达到显著性(Duncan's多重比较法)。

Data are mean $\pm$ SE. Histograms with the same letters indicate no significant difference among different treatments at 0.05 level by Duncan's multiple range test.

3 结论与讨论

国内外有关蓟马性比的研究报道主要来自西花蓟马,其性比的变化受多种生物因子和非生物因子的影响(Reitz *et al.*, 2011; Bondy and Hunter, 2019; Wan *et al.*, 2019),其中包括化学杀虫剂和生物杀虫剂(Bielza and Guillen, 2015; Zhang *et al.*, 2015; Cao *et al.*, 2019; Liu *et al.*, 2019)。本研究选用了3种用来防治豇豆上的蓟马或其他有害生物的杀虫剂,评价其亚致死浓度对已交配的普通大蓟马雌虫的繁殖和子代性别分配的影响。试验结果表明,供试杀虫剂对已交配的普通大蓟马雌虫的毒杀效果依次为:茚虫威>双甲脒>溴氰虫酰胺。亲代雌虫经溴氰虫酰胺、双甲脒和茚虫威 LC_{20} 处理48 h后的存活时间均显著缩短。溴氰虫酰胺和双甲脒处理还能显著减少已交配雌虫的子代成虫数量。已交配雌虫所产子代的成虫性比和卵性比在药剂处理和对照之间均没有显著的差异。由此说明,氰虫酰胺、双甲脒和茚虫威 LC_{20} 对已交配的普通大蓟马雌虫的性别分配均未产生明显的干扰作用。

溴氰虫酰胺是一种鱼尼丁受体抑制剂,影响昆虫肌肉的运动,可防治蓟马、粉虱、蚜虫等吸汁害虫(Satelle *et al.*, 2008; Wang *et al.*, 2016; Gahukar and Reddy, 2018)。Bielza and Guillen (2015)用溴氰虫酰胺亚致死浓度(60-100 mg/L)浸泡20 s的菜豆豆荚饲喂西花蓟马已交配雌虫48 h,随后1 d内所产子代成虫性比显著偏雄,而对照的子代成虫性比高度偏雌。在本研究中,采用的是豇豆叶盘浸药10 s,所观察的是溴氰虫酰胺 LC_{20} 胁迫处理已交配的普通大蓟马雌虫48 h后在存活期间所产下的全部子代,其成虫性比(0.60)比对照(0.52)更加偏雄,但二者之间的差异未达显著水平。Wang等(2017)曾发现溴氰虫酰胺 LC_{10} 和 LC_{25} 处理烟粉虱*Bemisia tabaci* (Gennadius)成虫48 h后,雌虫的繁殖力下降,且产卵期缩短。由此可见,溴氰虫酰胺亚致死浓度胁迫处理成虫对蓟马类和粉虱类害虫的存活与繁殖力均产生不利的影响。

双甲脒是一种杀螨剂,也可防治鳞翅目和半翅目害虫,其作用机理跟章鱼胺受体有关(Wei *et al.*, 2011; Kita *et al.*, 2017; Rix and Cutler, 2017)。茚虫威为电压门控钠通道的阻滞剂,主要用于防治鳞翅目害虫(Lapied *et al.*, 2001; Han *et al.*, 2016; Thamilarasi *et al.*, 2017; Gahukar and Reddy, 2018)。双甲脒和茚虫威 LC_{20} 处理能改变普通大蓟马亲代雌虫的存活时间或子代成虫数量,但对已交配雌虫的性别分配没干扰作用。

本研究采用叶盘浸药法获得的溴氰虫酰胺在48 h内对普通大蓟马雌虫的 LC_{50} 远大于对西花蓟马雌虫的 LC_{50} (493 mg/L)(Bielza and Guillen, 2015)。这可能与生物测定条件的不同有很大关系。Bielza and Guillen (2015)用浸药30 s的菜豆豆荚(20 mm $\times$ 5 mm)胁迫处理10头西花蓟马雌虫4 d。同本研究相比,上述研究采用了更长的食物浸药时间和试虫胁迫时间,单头雌虫所拥有的食物表面积更大一些。此外,溴氰虫酰胺对蓟马(Jacobson and Kennedy, 2013; Bielza and Guillen, 2015)、粉虱(Civolani *et al.*, 2014; Rattan *et al.*, 2015)、木虱(Mustafa *et al.*, 2015)等吸汁害虫有明显的拒食作用,这一特性

使得试虫在浸药食物上的滞留时间大大缩短。

采用叶盘法研究蓟马性比可以估算出卵性比,但不足之处在于子代数偏少。在本研究中,普通大蓟马子代成虫数最多的是对照组的已交配雌虫,为 5.94 头/雌,但此值远小于以豇豆豆荚为食时的雌虫繁殖力 (Tang *et al.*, 2015)。食物的营养状况是影响蓟马繁殖和存活的重要因子 (Reitz *et al.*, 2011),所以,普通大蓟马性比调控的研究还应考虑食物因子的影响。

跟很多植食性蓟马一样,普通大蓟马的繁殖方式包括两性生殖和产雄孤雌生殖 (Arrhenotoky),其繁殖采用的是单倍-二倍体性别决定系统 (Haplodiploid sex-determination system) (Chang, 1988; Moritz, 1997)。对单倍-二倍体昆虫而言,根据受精囊内精子的数量与质量,雌性成虫可归为 2 类:获精受限雌虫 (Constrained females) 和获精非受限雌虫 (Unconstrained females) (Godfray, 1990; Henter, 2004),其性比调控可分为两个部分:交配期间的性比调控和交配后的性比调控。本文仅评价了杀虫剂亚致死浓度对交配后的普通大蓟马性比调控的影响。杀虫剂亚致死浓度对该虫在交配期间的性比调控是否起作用,值得进一步研究,因为杀虫剂亚致死浓度有可能对昆虫的交配行为与过程产生负面影响,导致交配成功率下降 (Bielza and Guillen, 2015) 或者雌虫所获精子的数量减少与质量变差 (de França *et al.*, 2017; Krueger *et al.*, 2017; 李正辉等, 2017; Wan *et al.*, 2019)。

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