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Transgenerational effects on pest evolution

1 Transgenerational effects of insecticides - implications for rapid pest evolution in  
2 agroecosystems

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13 **Highlights**

- 14 • Insecticide-induced effects can be transgenerationally inherited.
- 15 • Epigenetic modifications are heritable.
- 16 • Epigenetic modifications are responsive to insecticide-induced stress.
- 17 • Pesticide use may directly and indirectly drive the evolution of insect pests in
- 18 agroecosystems via epigenetic processes.

19 **Abstract**

20 Although pesticides are a major selective force in driving the evolution of insect pests,

21 the evolutionary processes that give rise to insecticide resistance remain poorly understood.

22 Insecticide resistance has been widely observed to increase with frequent and intense insecticide

23 exposure, but can be lost following the relaxation of insecticide use. One possible but rarely

24 explored explanation is that insecticide resistance may be associated with epigenetic

25 modifications, which influence the patterning of gene expression without changing underlying

26 DNA sequence. Epigenetic modifications such as DNA methylation, histone modifications, and

27 small RNAs have been observed to be heritable in arthropods, but their role in the context of

28 rapid evolution of insecticide resistance remain poorly understood. Here, we discuss evidence

29 supporting how: 1) insecticide-induced effects can be transgenerationally inherited, 2) epigenetic

30 modifications are heritable, and 3) epigenetic modifications are responsive to pesticide and

31 xenobiotic stress. Therefore, pesticides may drive the evolution of resistance via epigenetic

32 processes. Moreover, insect pests primed by pesticides may be more tolerant of other stress,

33 further enhancing their success in adapting to agroecosystems. Resolving the role of epigenetic

34 modifications in the rapid evolution of insect pests has the potential to lead to new approaches

35 for integrated pest management as well as improve our understanding of how anthropogenic

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36 stress may drive the evolution of insect pests.

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## 38 **Introduction**

39         The pesticide treadmill describes how agricultural insect pests evolve resistance in  
40 response to frequently used pesticides, rendering them ineffective. Pesticides are pervasive in  
41 agriculture, and are a major selective force driving the evolution of insect pests in  
42 agroecosystems [1]. Although insecticide resistance has been documented in a wide range of  
43 insect pests [2] and the genetic basis of major gene resistance has been mapped in key pests for  
44 select insecticides [3], the broader evolutionary processes that give rise to insecticide resistance  
45 remain poorly understood [4,5]. Farmers and entomologists have observed that insecticide  
46 resistance increases with the frequency of exposure to particular insecticides [6–8], but can be  
47 lost following the relaxation of insecticide use [9–11]. The rapid gain and loss of resistance  
48 appears to occur far more rapidly than expected based upon mutation rates [12,13], suggesting  
49 that insecticides themselves may increase the rate of mutation or cause physiological changes in  
50 pest organisms [5]. One possible explanation that has been relatively unexplored is that the  
51 evolution of insecticide resistance results from epigenetic modifications, which are heritable and  
52 influence gene expression without changing the underlying DNA sequence.

53         The evolution of insecticide resistance has been considered an evolutionary paradox [5],  
54 in that pest species which have experienced repeated genetic bottlenecks due to invasion and  
55 selection remain able to adapt very rapidly, despite limited genetic diversity. The same insect  
56 pests have evolved resistance to insecticides in all of the major classes [14], and are expected to  
57 evolve resistance to future chemistries [15]. Extreme genetic bottlenecks also do not appear to  
58 limit the likelihood that insecticide resistance evolves. For example, all Colorado potato beetle  
59 (*Leptinotarsa decemlineata* Say) populations in Europe are descended from the introduction of a

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60 single female, or single mtDNA haplotype [16]. Despite this strong historic bottleneck, *L.*  
61 *decemlineata* populations in Eurasia have evolved resistance to a wide range of insecticides in  
62 Europe, the Middle East, and East Asia [6,17,18]. There is a seeming inevitability of insecticide  
63 resistance developing in pests, where new phenotypes arise following environmental stress at  
64 rates that may not be explained by natural selection. Indeed, Skinner et al. [19] argued how  
65 epigenetic processes fit within a neo-Lamarckian framework, because environmental epigenetic  
66 patterning can influence transgenerational transmission of phenotypic variation. By influencing  
67 epigenetic modifications, xenobiotic and environmental stressors can directly influence the  
68 phenotypic responses of organisms to their environment.

69 Epigenetics is the field of study that examines how environmental factors influence  
70 heritable changes in gene expression. There are several epigenetic mechanisms that are heritable  
71 and could underlie transgenerational effects of insecticides: DNA methylation [20], histone  
72 modifications [21], and heritable noncoding RNA [22]. Here, we discuss evidence supporting  
73 how 1) insecticide-induced effects can be transgenerationally inherited, 2) epigenetic  
74 modifications are heritable, and 3) epigenetic modifications are responsive to insecticide-induced  
75 stress. We draw on other model systems from a diverse body of literature, including genetics,  
76 epigenetics, and toxicology to identify gaps in our understanding around the evolution of  
77 insecticide resistance in insect pests. We close with a discussion of the implications of epigenetic  
78 processes for insect fitness in intensively-managed agroecosystems.

79

### 80 **Insecticide-induced hormetic effects can be heritable**

81 Insecticides not only select for insecticide resistance and point mutations at target sites,  
82 but they can also affect physiological and life-history traits [23]. In particular, exposure to

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83 sublethal dosages of insecticides can incur stress and lead to increased phenotypic variation [24].  
84 Stress responses can lead to hormesis, a well-known phenomenon from toxicological literature,  
85 where small dosages can stimulate biological functions whereas large dosages are detrimental or  
86 lethal [24]. Hormetic responses include activation of stress response pathways in a variety of  
87 taxa from microbes, plants, and animals. They are not related to any special class of compounds,  
88 as hormetic effects have been reported for over 240 different chemical classes [23]. Sublethal  
89 exposure to insecticides can induce hormetic effects and lead to variety of positive life history  
90 effects, such as mating success [25], fecundity [26], and body size [27]. By positively  
91 influencing traits associated with fitness, hormetic effects may play an important role in pest  
92 evolution.

93         There is evidence that individuals exposed to stressful conditions, either abiotic or biotic,  
94 can prime gene expression in their offspring to be able to better tolerate stress [28,29].  
95 Insecticides have been shown to induce transgenerational insecticide induced hormetic effects,  
96 but thus far the results have been difficult to interpret. For example, *Myzus persicae* aphids  
97 treated with sublethal levels of imidacloprid produce offspring that survive longer when exposed  
98 to food/water stress, but tolerance to insecticide stress is unchanged [30]. Similarly, although  
99 sublethal levels of precocene (an antagonist to Juvenile hormone) stimulate reproduction in *M.*  
100 *persicae*, the results are not passed on to subsequent generations [31\*]. Although chemical-  
101 induced hormesis has been reported from many groups and these changes have also been  
102 reported to be inherited [23] the genetic, epigenetic, and toxicological basis of hormesis is still  
103 poorly understood [5,32].

104

105 **Epigenetic modification and transgenerational inheritance**

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106           Epigenetic modifications have been shown to be heritable [20]. DNA methylation, the  
107 addition of a methyl group to the 5 carbon position of cytosine a nucleotide (usually the cytosine  
108 in CpG dinucleotides), is a well-documented mechanism of epigenetic inheritance that can  
109 influence phenotypic variation (Table 1), and is found in most, if not all, orders of insects [32].  
110 Methylation in insects is largely found within coding regions, and is closely linked with gene  
111 expression and alternative splicing - where a single gene can generate a diversity of gene  
112 transcripts of differing length, based on which exons are translated [33]. Methylation can occur  
113 at any location in the genome, but the effects of DNA methylation vary based on its location in  
114 the genome (Figure 1): A) changes in DNA methylation at the promoter region can influence  
115 gene expression in downstream genomic regions [34], B) methylation suppresses gene  
116 expression of transposable elements (TEs, which are mobile genetic elements responsible for the  
117 majority of mutations in many genomes) and prevent TE mobilization [35], and C) Gene body  
118 methylation can increase gene expression [32], as well as an increase in the number of alternative  
119 splice variants [36]. Changes in methylation patterns in arthropods can be associated with  
120 changes in levels of resistance to insecticides. *Myzus persicae*, can gain insecticide resistance  
121 through the duplication of esterase genes and subsequent overexpression of esterases [37]. After  
122 suspending insecticide exposure, extra copies of esterase genes can be methylated, leading to a  
123 loss of resistance. It is possible that these aphid populations could quickly become resistant again  
124 following demethylation of these amplified genes.

125           Histone modifications include additions of acetyl or methyl groups on the histone  
126 proteins around which nuclear DNA is wrapped, which can influence gene regulation and  
127 expression [38]. The full effects of these modifications are not well known, especially in  
128 arthropods. However, it does appear that some histone modifications are able to be transmitted

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129 transgenerationally [39]. Different noncoding RNA (ncRNA) [22] can be inherited through either  
130 the male or female gametes, though most current research does not incorporate analysis of  
131 heritable RNA. Certain types of small RNA can direct and maintain DNA methylation and  
132 histone modification, and therefore affect chromatin structure [40]. DNA methylation, histone  
133 modifications, and ncRNAs form a constellation of interacting effects that result in a phenotypic  
134 response [41]. To fully understand how epigenetic modifications influence transgenerational  
135 phenotypic inheritance, it would be optimal to assess all three mechanisms simultaneously  
136 through concurrent small RNA-seq, bisulfate-treated DNA-seq, and histone modification assays,  
137 in as many tissues and individuals as possible. Ideally, multiple generations would be sequenced,  
138 to determine if changes in epigenetics and gene expression differ consistently between  
139 treatments. Because the cost of sequencing is the major limiting factor for these studies,  
140 projected lower sequencing costs in the future should enable these types of studies.

141

**142 Epigenetic modifications are responsive to xenobiotic stress**

143 Exposure to insecticides and other xenobiotic compounds can alter DNA methylation  
144 status in arthropods, and these epigenetic changes can persist for at least several generations  
145 [20,42,43]. Table 1 lists a number of examples of stress leading to epigenetic changes in  
146 arthropods. Studies focusing on insects are few in number, so our scope is broadened to include  
147 examples from aquatic ecotoxicology literature, which includes a number of non-insect  
148 arthropods. Oppold (2015) found that exposure of mosquitoes to a fungicide leads to heritable  
149 changes in methylation and decreases in sensitivity to imidacloprid, an insecticide. Methylated  
150 cytosines also spontaneously deaminate, becoming thymines, at a higher rate than non-  
151 methylated cytosines, which can lead to higher mutation rates in methylated regions [44]. If

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152 genes that are associated with resistance are methylated, which leads to increased expression and  
153 increased mutation rate, then genes that are most upregulated in response to insecticide resistance  
154 may also be the most likely to experience spontaneous deamination.

155 Both the role of histone modifications and small RNA in modifying epigenetic responses  
156 to toxins are less understood than DNA methylation in arthropods, though it has been shown that  
157 methylation and histone modifications tend to be co-located in the genome [38]. Kishimoto et al.  
158 [45\*] showed that parental hormetic responses to oxidative stress can be epigenetically  
159 transmitted to descendants via histone modifications. A wide range of environmental chemicals,  
160 such as heavy metals, air pollutants, dioxins, and endocrine disruptors, can alter histone  
161 modifications [46], but it is unknown whether these changes are heritable. We have not found  
162 any studies on arthropods examining if insecticides can induce transgenerational small RNAs  
163 responses. Small RNAs have been found to interact with histone modifications [47], so changes  
164 in small RNAs may be implicated in the transgenerational inheritance of stress phenotypes as  
165 well.

166

**167 Implications for transgenerational effects on insect fitness in agroecosystems**

168 We hypothesize that pesticide use can directly and indirectly drive the evolution of insect  
169 pests in agroecosystems via epigenetic processes (Figure 2). Pesticides may directly stimulate the  
170 expression of advantageous phenotypes, which may be underwritten by epigenetic modifications.  
171 Continued insecticide use on populations developing resistance would thus operate as ‘natural  
172 selection’ and selectively increase the frequency of insect phenotypes that are adaptive to  
173 pesticides. Indirectly, pesticide use may maintain stressful environments that hormetically prime  
174 insect pests to become more tolerant of stressful conditions. For instance, sublethal exposure to

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175 insecticides can influence adult body size of the *L. decemlineata* [23], which may allow insect  
176 pests to be better able to tolerate overwintering conditions [48]. Insecticides can also increase  
177 female fecundity [49] or propensity to mate [25], which can increase population size.

178 The phenotypic traits of insect pests that allow them to thrive under insecticide exposure  
179 may also facilitate global invasions. For example, *L. decemlineata* is a globally-invasive pest that  
180 is expanding its range northwards into the Arctic Circle [48]. Insecticide exposure appears to  
181 stimulate the beetle to invest more in fat bodies and have a higher metabolic rate than control  
182 beetles [50]. While the higher metabolic rate and larger fat bodies may enable beetles to better  
183 detoxify chemicals, higher fat body reserves enable small individuals to overwinter successfully  
184 [51,52]. For example, sublethal applications of the pyrethroid deltamethrin on resistant *L.*  
185 *decemlineata* populations can have stimulatory effects rendering exposed individuals larger  
186 which is also inherited to the next generation (Lindström, unpublished data).

187 To date, most of the research examining the role of pesticides or xenobiotics in epigenetic  
188 change come from the field of aquatic toxicology [53\*\*], where environmental exposure to  
189 toxins can be highly variable and difficult to predict. In contrast, pesticide use in agroecosystems  
190 is intentionally part of an active pest management system, where insect responses to stresses can  
191 cause positive feedbacks on subsequent management decisions. Agroecosystems are also highly  
192 controlled systems, which allows for greater experimental control for field and landscape level  
193 studies. Along these lines, it would be important to know how epigenetic responses to the same  
194 insecticides may vary among individuals, populations, and species. Such information would help  
195 provide insight on whether epigenetic responses can be broadly predictable across individuals  
196 and species, and possibly, how pesticide resistance may be better managed. A combination of  
197 new genomic tools, epigenetic assays, and computationally-intensive approaches may allow us to

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198 better understand to what extent epigenetic responses within insects help drive the pesticide  
199 treadmill.

200

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207

## 208 **References**

- 209 1. Food and Agriculture Organization of the United Nations: *FAOSTAT Statistics Database*.  
210 2014.
- 211 2. Insecticide Resistance Action Committee: **Arthropod Pesticide Resistance Database**  
212 <http://www.irac-online.org>. Accessed September 15, 2017.
- 213 3. Ffrench-Constant RH: **Insecticide resistance comes of age**. *Genome Biol.* 2014, **15**:106.
- 214 4. Ffrench-Constant RH: **The molecular genetics of insecticide resistance**. *Genetics* 2013,  
215 **194**:807–15.
- 216 5. Gressel J: **Low pesticide rates may hasten the evolution of resistance by increasing**  
217 **mutation frequencies**. *Pest Manag. Sci.* 2011, **67**:253–257.
- 218 6. Tang L De, Qiu BL, Cuthbertson AGS, Ren SX: **Status of insecticide resistance and**  
219 **selection for imidacloprid resistance in the ladybird beetle *Propylaea japonica***  
220 **(Thunberg)**. *Pestic. Biochem. Physiol.* 2015, **123**:87–92.

## Transgenerational effects on pest evolution

- 221 7. Malekmohammadi M: **Resistance of Colorado potato beetle (*Leptinotarsa decemlineata***  
222 **Say) to commonly used insecticides in Iran.** *J. Asia. Pac. Entomol.* 2014, **17**:213–220.
- 223 8. Zhou C, Liu Y, Yu W, Deng Z, Gao M, Liu F, Mu W: **Monitoring resistance of field**  
224 **populations of diamondback moth *Plutella xylostella* L. (Lepidoptera:**  
225 **Yponomeutidae) to five insecticides in South China: A ten-year case study.** *Crop*  
226 *Prot.* 2011, **30**:272–278.
- 227 9. Yang Y, Dong B, Xu H, Zheng X, Tian J, Heong K, Lu Z: **Decrease of insecticide**  
228 **resistance over generations without exposure to insecticides in *Nilaparvata lugens***  
229 **(Hemipteran: Delphacidae).** *J. Econ. Entomol.* 2014, **107**:1618–1625.
- 230 10. Foster SP, Denholm I, Devonshire AL: **The ups and downs of insecticide resistance in**  
231 **peach-potato aphids (*Myzus persicae*) in the UK.** *Crop Prot.* 2000, **19**:873–879.
- 232 11. Ffrench-Constant RH, Devonshire AL, White RP: **Spontaneous loss and reselection of**  
233 **resistance in extremely resistant *Myzus persicae* (Sulzer).** *Pestic. Biochem. Physiol.*  
234 1988, **30**:1–10.
- 235 12. Drake JW, Charlesworth B, Charlesworth D, Crow JF: **Rates of spontaneous mutation.**  
236 *Genetics* 1998, **148**:1667–1686.
- 237 13. Jablonka E, Raz G: **Transgenerational epigenetic inheritance: prevalence,**  
238 **mechanisms, and implications for the study of heredity and evolution.** *Q. Rev. Biol.*  
239 2009, **84**:131–176.
- 240 14. Sparks TC, Nauen R: **IRAC: Mode of action classification and insecticide resistance**  
241 **management.** *Pestic. Biochem. Physiol.* 2015, **121**:122–128.
- 242 15. fFrench-Constant RH: **Which came first: insecticides or resistance?** *Trends Genet.*  
243 2007, **23**:1–3.

## Transgenerational effects on pest evolution

- 244 16. Grapputo A, Boman S, Lindström L, Lyytinen A, Mappes J: **The voyage of an invasive**  
245 **species across continents: genetic diversity of North American and European**  
246 **Colorado potato beetle populations.** *Mol. Ecol.* 2005, **14**:4207–19.
- 247 17. Stankovic S, Zabel A, Kostic M, Manojlovic B, Rajkovic S: **Colorado potato beetle**  
248 **[*Leptinotarsa decemlineata* (Say)] resistance to organophosphates and carbamates in**  
249 **Serbia.** *J. Pest Sci. (2004).* 2004, **77**:11–15.
- 250 18. Jiang W, Wang Z, Xiong M, Lu W: **Insecticide resistance status of Colorado potato**  
251 **beetle (Coleoptera□: Chrysomelidae) adults in Northern Xinjiang Uygur**  
252 **Autonomous Region.** *J. Econ. Entomol.* 2017, **103**:1365–1371.
- 253 19. Skinner MK: **Environmental epigenetics and a unified theory of the molecular aspects**  
254 **of evolution: A neo-Lamarckian concept that facilitates neo-Darwinian evolution.**  
255 *Genome Biol. Evol.* 2015, **7**:1296–1302.
- 256 20. Vandegheuchte MB, Lemièrre F, Vanhaecke L, Vanden Berghe W, Janssen CR: **Direct**  
257 **and transgenerational impact on *Daphnia magna* of chemicals with a known effect on**  
258 **DNA methylation.** *Comp. Biochem. Physiol. C. Toxicol. Pharmacol.* 2010, **151**:278–85.
- 259 21. Niu Y, DesMarais TL, Tong Z, Yao Y, Costa M: **Oxidative stress alters global histone**  
260 **modification and DNA methylation.** *Free Radic. Biol. Med.* 2015, **82**:22–8.
- 261 22. Liebers R, Rassoulzadegan M, Lyko F: **Epigenetic regulation by heritable RNA.** *PLoS*  
262 *Genet.* 2014, **10**:e1004296.
- 263 23. Calabrese EJ, Blain RB: **The hormesis database: The occurrence of hormetic dose**  
264 **responses in the toxicological literature.** *Regul. Toxicol. Pharmacol.* 2011, **61**:73–81.
- 265 24. Calabrese EJ, Mattson MP: **Hormesis provides a generalized quantitative estimate of**  
266 **biological plasticity.** *J. Cell Commun. Signal.* 2011, **5**:25–38.

## Transgenerational effects on pest evolution

- 267 25. Haddi K, Mendes M V., Barcellos MS, Lino-Neto J, Freitas HL, Guedes RNC, Oliveira  
 268 EE, Narciso R, Guedes C, Oliveira EE: **Sexual success after stress? Imidacloprid-**  
 269 **induce hormesis in males of the neotropical stink bug *Euschistus heros*. *PLoS One***  
 270 **2016, 11:e0156616.**
- 271 26. Ayyanath MM, Scott-Dupree CD, Cutler GC: **Effect of low doses of precocene on**  
 272 **reproduction and gene expression in green peach aphid. *Chemosphere* 2015, 128:245–**  
 273 **251.**
- 274 27. Stanley JK, Perkins EJ, Habib T, Sims JG, Chappell P, Escalon BL, Wilbanks M, Garcia-  
 275 Reyero N: **The good, the bad, and the toxic: approaching hormesis in *Daphnia magna***  
 276 **exposed to an energetic compound. *Environ. Sci. Technol.* 2013, 47:9424–9433.**
- 277 28. Freitag D, Knorr E, Vogel H, Vilcinskas A: **Gender-and stressor-specific microRNA**  
 278 **expression in *Tribolium castaneum*. *Biol. Lett.* 2012, 8:860–863.**
- 279 29. Tidbury HJ, Pedersen AB, Boots M: **Within and transgenerational immune priming in**  
 280 **an insect to a DNA virus. *Proceedings. Biol. Sci.* 2011, 278:871–6.**
- 281 30. Rix RR, Ayyanath MM, Christopher Cutler G: **Sublethal concentrations of**  
 282 **imidacloprid increase reproduction, alter expression of detoxification genes, and**  
 283 **prime *Myzus persicae* for subsequent stress. *J. Pest Sci.* 2016, 89:1–9.**
- 284 \*31. Ayyanath M-M, Cutler GC, Scott-Dupree CD, Prithiviraj B, Kandasamy S, Prithiviraj K:  
 285 **Gene expression during imidacloprid-induced hormesis in green peach aphid. *Dose.***  
 286 ***Response.* 2014, 12:480–97.**
- 287 This study shows heritable changes in methylation following imidacloprid exposure in an  
 288 arthropod pest.
- 289
- 290 32. Glastad KM, Hunt BG, Yi S V, Goodisman MAD: **DNA methylation in insects: on the**

## Transgenerational effects on pest evolution

- 291           **brink of the epigenomic era. *Insect Mol. Biol.* 2011, **20**:553–65.**
- 292 33. Flores K, Wolschin F, Corneveaux JJ, Allen AN, Huentelman MJ, Amdam G V:  
293           **Genome-wide association between DNA methylation and alternative splicing in an**  
294           **invertebrate.** *BMC Genomics* 2012, **13**:480.
- 295 34. Hunt BG, Glastad KM, Yi S V, Goodisman MAD: **The function of intragenic DNA**  
296           **methylation: insights from insect epigenomes.** *Integr. Comp. Biol.* 2013, **53**:319–28.
- 297 35. Fablet M, Vieira C: **Evolvability, epigenetics and transposable elements.** *Biomol.*  
298           *Concepts* 2011, **2**:333–41.
- 299 36. Field LM, Devonshire AL, Ffrench-Constant RH, Forde BG: **Changes in DNA**  
300           **methylation are associated with loss of insecticide resistance in the peach-potato**  
301           **aphid *Myzus persicae* (Sulz.).** *FEBS Lett.* 1989, **243**:323–327.
- 302 37. Hunt BG, Glastad KM, Yi S V, Goodisman MAD: **Patterning and regulatory**  
303           **associations of DNA methylation are mirrored by histone modifications in insects.**  
304           *Genome Biol. Evol.* 2013, **5**:591–8.
- 305 38. Szyf M: **Nongenetic inheritance and transgenerational epigenetics.** *Trends Mol. Med.*  
306           2015, **21**:134–144.
- 307 39. Holoch D, Moazed D: **RNA-mediated epigenetic regulation of gene expression.** *Nat.*  
308           *Rev. Genet.* 2015, **16**:71–84.
- 309 40. Peschansky VJ, Wahlestedt C: **Non-coding RNAs as direct and indirect modulators of**  
310           **epigenetic regulation.** *Epigenetics* 2014, **9**:3–12.
- 311 41. Vandegehuchte MB, Kyndt T, Vanholme B, Haegeman A, Gheysen G, Janssen CR:  
312           **Occurrence of DNA methylation in *Daphnia magna* and influence of multigeneration**  
313           **Cd exposure.** *Environ. Int.* 2009, **35**:700–6.

## Transgenerational effects on pest evolution

- 314 42. Vandegehuchte MB, De Coninck D, Vandenbrouck T, De Coen WM, Janssen CR: **Gene**  
315 **transcription profiles, global DNA methylation and potential transgenerational**  
316 **epigenetic effects related to Zn exposure history in *Daphnia magna*. *Environ. Pollut.***  
317 2010, **158**:3323–9.
- 318 43. Poulos RC, Olivier J, Wong JWH: **The interaction between cytosine methylation and**  
319 **processes of DNA replication and repair shape the mutational landscape of cancer**  
320 **genomes. *Nucleic Acids Res.* 2017, **45**:7786–7795.**
- 321 44. Baccarelli A, Bollati V: **Epigenetics and environmental chemicals. *Curr. Opin. Pediatr.***  
322 2009, **21**:243–51.
- 323 \*45. Kishimoto S, Uno M, Okabe E, Nono M, Nishida E: **Environmental stresses induce**  
324 **transgenerationally inheritable survival advantages via germline-to-soma**  
325 **communication in *Caenorhabditis elegans*. *Nat. Commun.* 2017, **8**:14031.**
- 326 This study, in *C.elegans*, shows how stress resistance is transmitted to subsequent generations via  
327 epigenetic alterations.  
328
- 329 46. Rechavi O, Lev I: **Principles of transgenerational small RNA inheritance in**  
330 ***Caenorhabditis elegans*. *Curr. Biol.* 2017, **27**:R720–R730.**
- 331 47. Piironen S, Ketola T, Lyytinen A, Lindström L: **Energy use, diapause behaviour and**  
332 **northern range expansion potential in the invasive Colorado potato beetle. *Funct.***  
333 ***Ecol.* 2011, **25**:527–536.**
- 334 48. Alyokhin A, Guillemette R, Choban R: **Stimulatory and suppressive effects of**  
335 **novaluron on the Colorado potato beetle reproduction. *J. Econ. Entomol.* 2009,**  
336 **102**:2078–2083.
- 337 49. Piironen S, Boman S, Lyytinen A, Mappes J, Lindström L: **Sublethal effects of**

- 338 **deltamethrin exposure of parental generations on physiological traits and**  
339 **overwintering in *Leptinotarsa decemlineata*. *J. Appl. Entomol.* 2014, **138**:149–158.**
- 340 50. Piironen S, Lyytinen A, Lindström L: **Stress for invasion success? Temperature stress**  
341 **of preceding generations modifies the response to insecticide stress in an invasive**  
342 **pest insect. *Evol. Appl.* 2013, **6**:313–23.**
- 343 51. Lehmann P, Lyytinen A, Piironen S, Lindström L: **Northward range expansion**  
344 **requires synchronization of both overwintering behaviour and physiology with**  
345 **photoperiod in the invasive Colorado potato beetle (*Leptinotarsa decemlineata*).**  
346 *Oecologia* 2014, **176**.
- 347 52. Brander SM, Biales AD, Connon RE: **The role of epigenomics in aquatic toxicology.**  
348 *Environ. Toxicol. Chem.* 2017, **36**:2565–2573.
- 349 \*\*53. Oppold A, Kreß A, Bussche J Vanden, Diogo JB, Kuch U, Oehlmann J, Vandegehuchte  
350 MB, Müller R: **Epigenetic alterations and decreasing insecticide sensitivity of the**  
351 **Asian tiger mosquito *Aedes albopictus*. *Ecotoxicol. Environ. Saf.* 2015, **122**:45–53.**
- 352 Oppold *et al.* demonstrate that exposure to a toxic compound can affect epigenetic state for  
353 several generations, and that there is a phenotypic effect (decreased susceptibility to  
354 imidacloprid) associated with these methylation alterations.
- 355
- 356 54. Norouzitallab P, Baruah K, Vandegehuchte M, Van Stappen G, Catania F, Vanden  
357 Bussche J, Vanhaecke L, Sorgeloos P, Bossier P: **Environmental heat stress induces**  
358 **epigenetic transgenerational inheritance of robustness in parthenogenetic *Artemia***  
359 **model. *FASEB J.* 2014, **28**:3552–63.**
- 360 55. Vandegehuchte MB, Lemièrre F, Janssen CR: **Quantitative DNA-methylation in**  
361 ***Daphnia magna* and effects of multigeneration Zn exposure. *Comp. Biochem. Physiol.***  
362 ***C. Toxicol. Pharmacol.* 2009, **150**:343–8.**

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- 363 \*\*56. Asselman J, De Coninck DI, Beert E, Janssen CR, Orsini L, Pfrender ME, Decaestecker  
364 E, De Schamphelaere KA: **Bisulfite sequencing with *Daphnia* highlights a role for**  
365 **epigenetics in regulating stress response to *Microcystis* through preferential**  
366 **differential methylation of serine and threonine amino acids.** *Environ. Sci. Technol.*  
367 2017, **51**:924–931.

368 This study describes how stress-induced methylation patterns can be associated with certain  
369 codon and genes with certain functions relevant to a stress response.  
370

- 371 57. Kumar S, Kim Y: **An endoparasitoid wasp influences host DNA methylation.** *Sci. Rep.*  
372 2017, **7**:43287.
- 373 58. Seong K-H, Li D, Shimizu H, Nakamura R, Ishii S: **Inheritance of stress-induced, ATF-**  
374 **2-Dependent Epigenetic Change.** *Cell* 2011, **145**:1049–1061.

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## Transgenerational effects on pest evolution

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379 Table 1. Examples of epigenetic alterations following exposure to anthropogenic and “natural”,  
 380 (non-anthropogenic) stress.

|                         | Species  | Treatment  | Phenotypic Effects   | DNA Methylation  | Histone Modifications          | Transgenerational effect | Reference |
|-------------------------|--|--|--|--|--------------------------------|--------------------------|-----------|
| anthropogenic stressors | <i>Daphnia magna</i>                           | vinclozolin (fungicide)                            | body size  | reduced methylation  | n/a                            | N                        | [20]      |
|                         | <i>Daphnia magna</i>                           | 5-azacytidine (demethylating agents)               | reproduction, body size  | reduced methylation  | n/a                            | Y                        | [20]      |
|                         | <i>Myzus persicae</i> (Green Peach Aphid)      | imidacloprid (insecticide)                         | changes in gene expression, including heat shock protein                 | increased, decreased, or no change based on concentration  | n/a                            | Y                        | [31*]     |
|                         | <i>Aedes albopictus</i> (Asian Tiger Mosquito) | genistein (phytohormone)                           | decrease in sensitivity to imidacloprid                                  | cautious decrease  | n/a                            | Y                        | [54]      |
|                         | <i>Aedes albopictus</i> (Asian Tiger Mosquito) | vinclozolin (fungicide)                            | decrease in sensitivity to imidacloprid                                  | cautious increase  | n/a                            | Y                        | [54]      |
| "natural" stressors     | <i>Artemia</i> sp. (brine shrimp)              | heat stress  | increased Hsp70 production, heat tolerance, and resistance vs. pathogens | changes in methylation   | histones H3 and H4 acetylation | Y                        | [55]      |
|                         | <i>Daphnia magna</i>                           | zinc   | changes in gene expression   | reduced methylation  | n/a                            | Y                        | [56*]     |
|                         | <i>Daphnia magna</i>                           | toxic cyanobacterium <i>Microcystis aeruginosa</i> | n/a  | differential methylation primarily in exonic regions, enriched for serine/threonine amino acid codons and genes related to protein synthesis, transport and degradation, in genes susceptible to alternative splicing in response to <i>Microcystis</i> stress | n/a                            | N                        | [57]      |
|                         | <i>Plutella xylostella</i> (Diamondback Moth)  | endoparasitoid                                     | altered gene expression  | reduced methylation  | n/a                            | N                        | [58]      |

## Transgenerational effects on pest evolution

|  |   |                              |     |     |                            |   |      |
|--|---|------------------------------|-----|-----|----------------------------|---|------|
|  | <i>Drosophila melanogaster</i><br>(Fruit Fly) | Heat shock or osmotic stress | n/a | n/a | heterochromatin disruption | Y | [59] |
|--|---|------------------------------|-----|-----|----------------------------|---|------|

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382 Figure Legends.

383 Figure 1. Examples of how changes in methylation status of in different gene regions can effect  
 384 gene expression. Compared to the “normal” unmethylated region, A) has promoter methylation,  
 385 leading to decreased gene expression; B) exhibits methylation in transposable element regions,  
 386 leading to those elements not being expressed, and C) shows gene body methylation as found in  
 387 arthropods, leading to increased gene expression as well as an increased variety of splice variants  
 388 in those transcripts.

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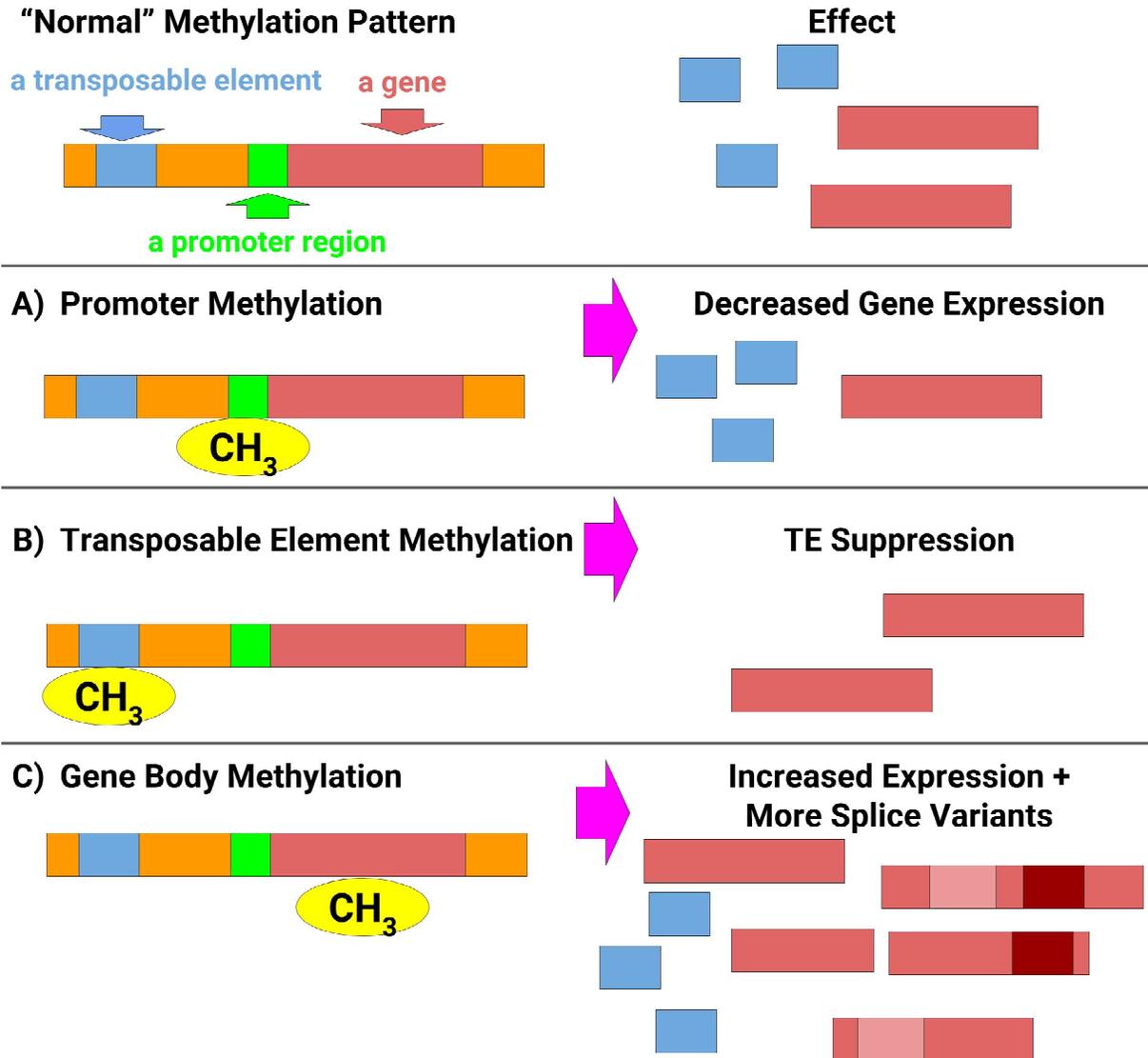
390 Figure 2. How exposure to a stressor may lead to heritable epigenetic changes that could lead to  
 391 stress-resistant phenotype in an invasive agricultural insect pests.

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## Mechanisms of Methylation and Gene Expression in Insects



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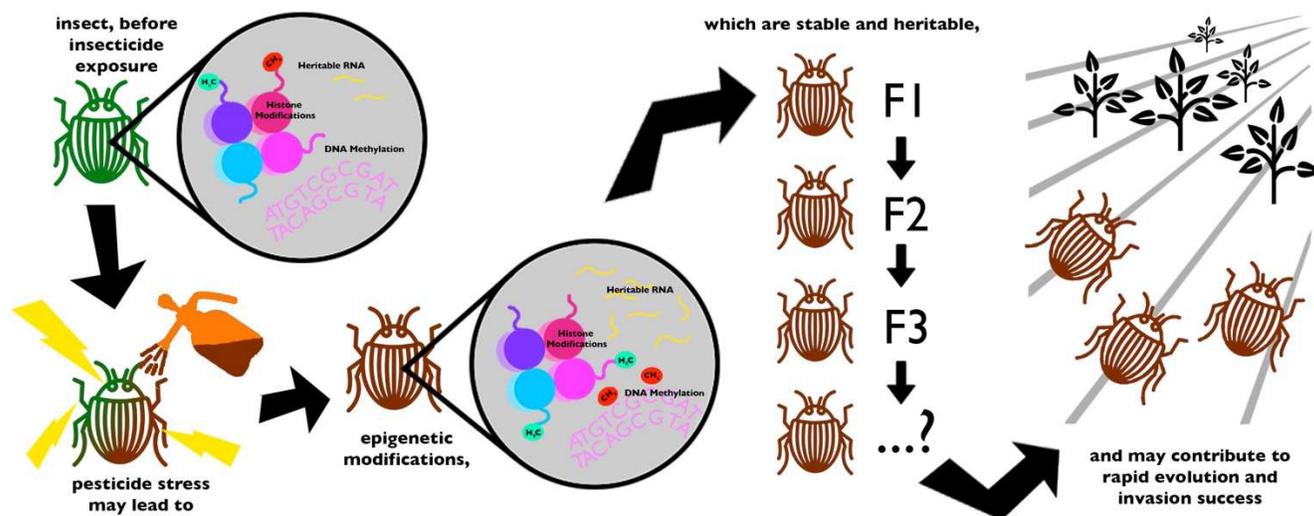
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399 Brevik et al. Figure 1.

## Transgenerational effects on pest evolution



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406 Brevik et al. Figure 2.

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