Towards a successful resistance and virulence management strategy for CpGV

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Abstract

Cydia pomonella *granulovirus is a cornerstone in the control of codling moth (*Cydia pomonella*) in organic pome fruit production. Since emergence of first cases of resistance to commercial CpGV preparations, new resistance-breaking CpGV products have been developed. This contribution provides some considerations on resistance and virulence management for CpGV products.*

Keywords: Codling moth, *Cydia pomonella* granulovirus, CpGV resistance, resistance-breaking viruses, virulence management

Introduction

Cydia pomonella (codling moth, CM) is a severe pest of organic and inegrated apple and pear plantation. Cydia pomonella granulovirus (CpGV) is virus pathogen of CM, which has been successfully used for CM control for more than 25 years. CpGV products are important to control CM in organic pome fruit production (Lacey *et al*., 2008). However, resistance to commercial CpGV products has been observed since 2004 in about 40, mainly organic orchards in different European countries, including Germany (22 orchards), France (3), Switzerland (2), Italy (6), Austria (2), the Netherlands (2) and the Czech Republique (1) (Fritsch *et al.,* 2005; Sauphanor *et al*., 2006, Schmitt *et al.,* 2013).

Known CpGV isolates can be grouped according to genome sequence analysis into five genome groups A-E. Molecular analyses revealed that in most cases CpGV resistance is targeted against CpGVs from genome group A, which was present in all previous commercial CpGV products in Europe (Fig.1) (Gebhardt *et al.,* 2014).

CpGV genome groups B-E are able to break this resistance. Recently, evidence for a second type of CpGV resistance became available from a very few cases in Germany. This resistance is not only targeted against genome group A CpGVs but also against genome group B-D (Jehle *et al*., submitted). For a successful resistance and virulence management it will be important to use the full natural biodiversity of CpGV. Therefore, new products, for example MadexPlus and MadexMAX (Andermatt Biocontrol) or Carpovirusine Evo2 (NPP-Arysta), have been developed and registered. To secure a continuous success in CM control in organic production it will be essential to put the control on a broad basis and to diversify the control methods. Here we present some conclusions from ongoing research which should help to sustain the efficacy of CpGV products in long-term.

Using the natural biodiversity of CpGV isolates

For historic reasons, all commercial products in Europe contained a single CpGV isolate, i.e. CpGV-M (genome group A). Resistance in most orchards is targeted against this isolate, whereas other genome groups B-E can break CpGV resistance. We performed next generation sequencing of the commercial resistance breaking products MadexMAX

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and Carpovirusine Evo2. It was found that both MadexMAX and Carpovirusine Evo2 contain a similar composition of genotypes with a genome group E as main component. Whether there are minor differences in single nucleotide positions could not be detected. In terms of resistance management we consider both products as equivalent. This finding indicates that concerning resistance management neither mixing nor rotation of both products will have a promising effect on the potential to delay or avoid resistance selection. If CM populations are able to develop resistance against one of both products it will be very likely that both products will fail. Therefore, for a long-term resistance management of CpGV products the mode of infection, especially on detailed molecular level for different isolates is important. Based on these most likely highly fine-tuned differences, the registration and use of further CpGV isolates from different genome groups and further investigations on the existing biodiversity of CpGV is essential. Biocontrol companies are encouraged to develop further products based on other CpGV genome groups than A and E.

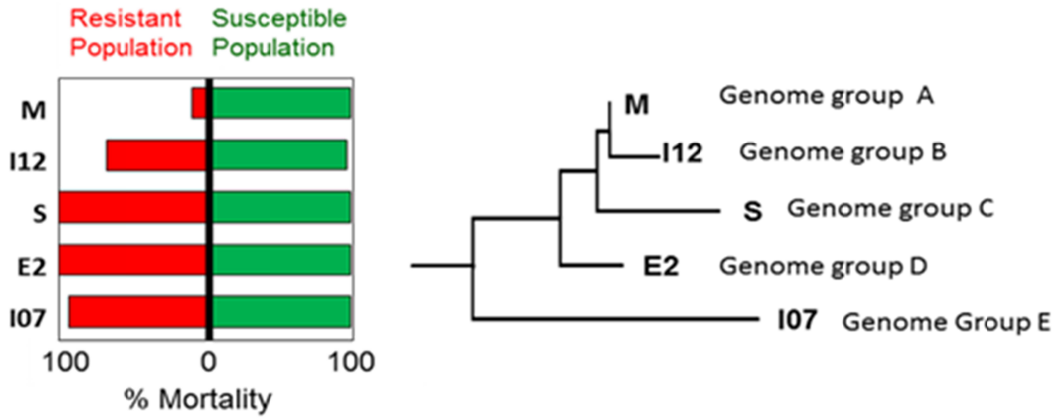

Figure 1: Activity and phylogeny of CpGV isolates. Known CpGV isolates can be grouped into five different genome groups A-E. For details see Gebhardt et al. (2014).

Resistance Management

In general, CpGV products derive from a natural virus isolate which is genetically more or less homogeneous (genome group). The genetically most homogenous preparation of a baculovirus is an *in-vivo* or *in-vitro* cloned genotype, which origins from a single viral genome and differs only in spontaneous mutations. It has been demonstrated for several baculoviruses that genotype mixtures can be more effective than pure genotypes (Lopez-Ferber *et al.,* 2003). CpGV isolates used in biological control are not fully homogenous but a mixture of more or less similar genotypes. Mixing different isolates would increase in the genetic diversity within the product and may lead to further synergistic effects. Therefore, mixing of different genome groups may be a promising approach to raise the efficacy of CpGV products. On the other hand, if a product contains significant genotype mixtures, each genotype will be present in a lower concentration than in a homogenous product and target populations are continuously and simultaneously exposed to the different genotypes at lowered concentration. Such a continuous exposition may increase the likelihood of resistance development in a target population. Therefore, a limited mix of few genome groups may be useful in terms of efficacy and resistance management, but we do not recommend having only a single mixture containing all genome groups in the same preparation as they may provoke the selection of a CM population with very broad resistance. Rotation of products containing different genome groups will be more sustainable in terms of resistance management.

Virulence Management

Viruses are biological entities which are subjected to evolutionary processes such as mutation or selection. Depending on the host, they may change more or less quickly in their genetic character. In nature, there is an arms-race between virus and host insect, development of resistance is followed by breaking of resistance. This biological alterability makes biological agents different from chemicals. It has been shown, that the activity of a CpGV isolate can be improved by a selection process on a resistant CM strain (Berling *et al.*, 2009; Graillot *et al.*, 2015). After a couple of passages through resistant CM larvae the final CpGV isolate was more virulent than the starting material. Therefore, the biological system of CpGV and CM allows managing the virulence of CpGV by performing this natural evolutionary process in the laboratory. This strategy may help in the future to develop new resistance-breaking CpGV products.

Outlook

Though, the knowledge about the molecular diversity of CpGV and the mechanisms of resistance breaking is far better understood than a few years ago, intensive research is necessary to learn about the mechanism(s) how CM can develop resistance to granulovirus infection. Only with this knowledge a full resistance and virulence management strategy can be implemented.

Acknowledgements

We acknowledge financial support of BMEL (2809OE097) and DFG (Je245/14-1).

References

Berling, M., Blachère-Lopez, C., Soubabère, O., Lery, X., Bonhomme, A., Sauphanor, B. & López-Ferber, M. (2009). *Cydia pomonella* granulovirus genotypes overcome virus resistance in the codling moth and improve virus efficiency by selection against resistant hosts. *Applied and Environmental Microbiology* **75**: 925-930.

Fritsch, E., Undorf-Spahn, K., Kienzle, J., Zebitz, C.P.W. & Huber, J. (2005). Apfelwickler-Granulovirus: Erste Hinweise auf Unterschiede in der Empfindlichkeit lokaler Apfelwickler-Populationen. *Nachrichtenblatt des Deutschen Pflanzenschutzdienstes* **57**: 29-34.

Gebhardt, M., Eberle, K.E., Radtke, P. & Jehle, J. A. (2014). Baculovirus resistance in codling moth is virus-isolate dependent and the consequence of a mutation in viral gene pe38. *Proceedings of the National Academy of Sciences of the United States of America (PNAS)* **111**: 15711-15716.

Graillot, B., Berling, M., Blachere-López, C., Siegwart, M., Besse, S. & López-Ferber, M. (2015). Progressive adaptation of a CpGV Isolate to codling moth populations resistant to CpGV-M. *Viruses* **6**: 5135-5144.

Lacey, L.A., Thomson, D., Vincent, C. & Arthurs, S.P. (2008). Codling moth granulovirus: a comprehensive review. *Biocontrol Science and Technology* **18**: 639-663.

López-Ferber, M., Simón, O., Williams, T. & Caballero, P. (2003). Defective or effective? Mutualistic interactions between virus genotypes. *Proceedings of the Royal Society B: Biological Sciences* **270**: 2249-2255.

Sauphanor, B., Berling, M., Toubon, J.-F., Reyes, M., Delnatte, J. & Allemoz, P. (2006). Carpocapse des pommes. Cas de résistance au virus de la granulose en vergers biologique. *Phytoma-La Défense des Végétaux* **590**: 24-27.

Citation of the full publication

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