PPP-final report

PPP projects which are under supervision of the "Topsectoren" must file a final report concerning the total project period. This form is used to report the content of the project. There is a separate form for the financial reporting.
The final report will be published on the TKI / topsector website. Therefore, please ensure that there is no confidential information in the final report.
The PPP-final report must be sent, at the latest, by the 1st of March 2020 to the "TKI's": info@tkitu.nl or info@tki-agrifood.nl. For Wageningen Research, the report has to be sent to the "Topsector secretary" of your respective institute.

General information	
PPP-number	KV 1409-045
Title	Mechanism of thrips resistance in Capsicum
Theme	T&U, Meer met Minder
Implementing institute	Plant Breeding, Wageningen University and Research
Project leader research (name + e-mail address)	Ben Vosman (ben.vosman@wur.nl)
Coordinator (on behalf of private partners)	Alejandro Lucatti (alejandro.lucatti@vegetableseeds.basf.com)
Project-website address	**-**
Start date	June 1, 2015
Final date	December 31, 2019

Approval by the coordinator of the consortium The final report must be discussed with the coordinator of the consortium. The "TKI's" appreciate additional comments concerning the final report.	
Assessment of the report by the coordinator on behalf of the consortium:	x Approved ☐ Not approved
Additional comments concerning the final report:	

Consortium	
Mention any changes in the composition of the project partners:	-

Summary of the project	
Problem definition	Thrips are among the major pests worldwide. They are difficult to control because of their cryptic habit, the larvae hide in closed buds and pupate in soil. This makes them difficult to reach by pesticide sprays, which limits their effectiveness. Recently we have discovered an effective source of thrips resistance in pepper and shown that the resistance was based on inhibition of larval development. The goal of the current project is to elucidate the mechanism and identify the gene(s) involved. This information is essential for the development of thrips resistant pepper varieties and may also results in leads that will help breeders to develop thrips resistant varieties in other crops as well.

Project goals	The goal of this project is to elucidate the mechanism of thrips resistance in pepper through identification of the gene(s) involved. To reach this goal we will use a molecular genetic approach including: (1) Identification of the gene(s) underlying the previously identified QTL for thrips resistance in pepper through fine mapping. (2) Elucidation of the resistance mechanism; which compound(s) inhibit the larval development? Which thrips species are affected by the QTL. (3) Study the evolution and distribution of the resistance gene(s) in the *Solanaceae*. (4) Study the effect of the genetic background on expression of the resistance.

Results	
Planned results in the original project plan	In the current project we want to obtain basic knowledge on the mechanism of thrips resistance in pepper through identification of the gene(s) involved. More specifically we want to get answers to the following fundamental questions: (1) Which genes are underlying the resistance encoded by the QTL on Chr. 6? (2) What is the resistance mechanism; which compound(s) inhibit the larval development? Are other thrips/insect species affected? (3) Are the gene(s) also present in related taxa and how have they evolved? (4) What is the effect of the genetic background on the resistance? Interaction between our chromosome 6 QTL and the QTL identified on chromosome 5 by Syngenta (PCT/EP2008/055374)?
Achieved results	The starting point of this project were two mapping studies in which accession CGN16975 was used as donor of thrips resistance. A study by WUR that identified a major resistance locus on chromosome 6, and a study by Syngenta that identified a QTL on chromosome 5. In the studies a different susceptible parent and a different evaluation protocol were used. Six metabolite QTLs, including two diterpene glycosides, have been identified that co-locate with the resistance QTL on chromosome 6, suggesting that they may play a role in the resistance encoded by the QTL (Maharijaya et al., 2018) In this project we first further characterized the effect of plant development on thrips resistance in *Capsicum*. The fraction of first instar larvae that did not develop into second instar was used as a resistance measure. Our results show that plants start to express thrips resistance when they are between four and eight weeks old. Furthermore, it was shown that youngest fully opened leaves of the resistant accession are significantly more resistant to thrips than older leaves, whereas young leaves of the susceptible accession are more susceptible than older leaves. Although the resistance is not equally expressed in all plant parts, it significantly reduced silvering damage in the whole plant damage assay. The preference of female adults to lay their eggs in younger compared to older leaves showed that the resistance mechanism is relevant and effective (Van Haperen et al., 2018). The resistance QTL on chromosome 6 has been fine mapped, using large set (3833) F3 plants, to a region of 0.4 Mbp harboring 15 genes. In addition to the fine mapping an RNAseq analysis has been carried out on F4-plants with and without the resistance QTL. Two of the 15 genes in the QTL region showed altered expression upon infestation with L1 larvae. Three genes, including the two that showed altered expression, contained a SNP that was predicted to lead to structural changes in the proteins, making these three genes the most likely candidates for thrips

	resistance (van Haperen, 2020; thesis chapter 3). The possible role of diterpene glycosides and other metabolites in the thrips resistance mechanism underlying the resistance QTL on chromosome 6 was further investigated using F4 plants that have the resistance allele and susceptibility allele in homozygous state for the QTL region. No significant differences in metabolite abundances were observed between the different groups of F4 plants for mock treated and thrips infested plants. From this analysis, it was concluded that diterpene glycosides do not play a role in the resistance mechanism encoded by the QTL on chromosome 6 (van Haperen, 2020; thesis chapter 4). Companies involved in the project transferred the resistance allele of the chromosome 6 QTL into four different susceptible backgrounds and the effect of the introgression on *F. occidentalis* and *Thrips tabaci* resistance was studied. Inhibition of larval development was used as resistance parameter in plants that have either the resistance or susceptibility allele in homozygous state for the QTL region, within the same genetic background. The resistance allele affected larval development of both thrips species in all tested backgrounds. Materials containing the resistance allele were shown to be more resistant than materials without the allele, thus validating the importance of the QTL. However, significant differences in thrips resistance level between different backgrounds were observed, indicating that genetic factors in the backgrounds play a role as well (van Haperen, 2020; thesis chapter 5). The possible interaction between the QTLs identified on chromosome 5 and 6 on both thrips species was studied using plant materials containing different QTL combinations in no-choice assays. The QTL on chromosome 5 did not affect thrips resistance in both larval development and silvering damage assays. In contrast, a significant effect of the QTL on chromosome 6 was found on both. It is likely that the NIL that was filed with the patent application has the resistance allele on chromosome 6 that originated from CGN16975. The findings from this study show that the QTL on chromosome 6 works independently from the QTL on chromosome 5 in all tested backgrounds (van Haperen, 2020; thesis chapter 6). Altogether, this work clearly shows that the resistance QTL identified on chromosome 6 has good potential in breeding thrips resistant varieties.
Explanation of changes relative to the project plan	Unfortunately we were not able to pinpoint a single gene or resistance component due to the fact that obtaining the recombinants needed for the fine mapping, took considerably more effort than anticipated. For that reason we could also not study the resistance gene in related species. Additional effort was put into a further characterization of the resistance in pepper.

What was the added value created by the project for:	
Participating "Knowledge Institutes" (scientific, new technologies, collaboration)	For WUR Plant Breeding the project was a successful resulting in a better characterization and understanding of thrips resistance. It resulted in a PhD thesis that will be defended publicly on March 24 (2020), as well as several oral presentations, posters, abstracts and one scientific paper published and more to follow soon.
Participating private partners (practical application of the results, within which period of time?)	The participating breeding companies obtained a lot of knowledge on thrips resistance in *Capsicum.* In addition they developed plant material with the resistance. These materials may result in new varieties. They have developed advanced plant material from the crosses.
Society (social, environment, economy)	When the thrips resistant varieties enter the market, they will need less or no chemical protection against thrips.

Possibly other stakeholders (spin-offs)	-

Follow-up	
Did the PPP result in one or more patents (first filings)?	no
Are there any follow-up projects planned? If yes, explain. (Contract research resulting from this project, additional funding, or new PPP projects)	The project contributed to acquiring a new TKI project on thrips resistance in *Chrysanthemum*.

Deliverables/products during the entire course of the PPP (provide the titles and/or a brief description of the products/deliverables or a link to a website.
Scientific articles: 1. Maharijaya, A., B. Vosman, K. Pelgrom, Y. Wahyuni, R.C.H. de Vos & R.E Voorrips (2018) Genetic variation in phytochemicals in leaves of pepper (*Capsicum*) in relation to thrips resistance. Arthropod-Plant Interactions https://doi.org/10.1007/s11829-018-9628-7 2. Van Haperen, P., R.E. Voorrips, J.J.A. van Loon & Ben Vosman (2018) The effect of plant development on thrips resistance in *Capsicum*. Arthropod-Plant Interactions https://doi.org/10.1007/s11829-018-9645-6 3. Van Haperen, P. (2020) Towards breeding thrips-resistant varieties in *Capsicum*. Ph.D. thesis, Wageningen University, Wageningen, the Netherlands. (public defense is on March 24, 2020)
External reports: -
Articles in professional journals/magazines: -
(Poster) presentations at workshops, seminars or symposia. **Oral presentations:** 1. Van Haperen, P. (2019) The effect of plant development on thrips resistance in Capsicum. EPS symposium Theme 2, February 1 2019, Wageningen, NL 2. Vosman, B. (2019) Insectenresistentie onderzoek. Vakopleiding voor plantenveredeling. 8 Maart 2019, Wageningen, Nl. 3. Van Haperen, P. (2019) Mechanism of thrips resistance in Capsicum. Transatlantic Summer School - Frontiers in Plant Sciences. May 27-31, 2019. Maria in der Aue, Germany. 4. Van Haperen, P. (2019) Thrips resistance in Capsicum - Exploring the role of QTLs in thrips resistance in 2 different Capsicum backgrounds. Plant Breeding and Biotechnology Symposium. June 11-13, 2019. Wageningen, NL. 5. Vosman, B. (2019) Breeding insect resistant crops. 10th Brazilian congress of plant breeding. 30 july 2019, Aguas de Lindoia/SP. 6. Vosman, B. (2019) Breeding insect resistant crops. Universidade federal de Lavras. 1 August 2019, Lavras/MG.

7. Voorrips, R.E. (2019) The effect of plant age and leaf age on thrips resistance in Capsicum. 17th Eucarpia meeting on genetics and breeding of Capsicum and Eggplant, 11-13 September 2019, Avignon, France.
8. Vosman, B. (2018) Enrichment of Crop Genepools with Wild Relative and Landrace Diversity. 4th International Conference “Plant Genetics & Breeding Technologies” July 12-13, 2018, Vienna.
9. Vosman, B. (2018) Breeding for thrips resistance in vegetables. TKI-TU netwerkevent 2018, April 3, Nieuwegein, NL.
10. Vosman, B. (2018) Different Mechanisms of Insect Resistance in tomato and pepper. 15th Solanaceae Conference, Sept 30th - Oct 4th 2018, Chiang Mai, Thailand.
11. Vosman, B. (2017) Host plant resistance against insect pests in pepper and tomato. Asian Solanaceous Round Table 2017(ASRT-2). February 23-25, 2017, Bangkok, Thailand
12. Vosman, B. (2017) Insectenresistentie onderzoek. Vakopleiding voor plantenveredeling, Wageningen 10 maart 2017.
13. Vosman, B. (2017) Breeding for Host Plant Resistance against insects in the post genomics era. International Symposium on Marine and Agricultural Genomics (ISMAG)”. April 19-21, 2017, organized by the Marine Genome 100+ Korea and National Agricultural Genome Program (NAGP) of South Korea. Seoul, South Korea.
14. Vosman, B. (2016) Host plant resistance towards insects. ICE 2016 XXV International congress of Entomology. September 25-30, 2016. Orlando Florida, USA.

Posters:

1. Van Haperen, P., Voorrips, R.E., Van Loon, J.J.A., Vosman, B. (2019) The effect of three QTLs and two backgrounds on thrips resistance in Capsicum. EPS Lunteren. April 9, 2019, Lunteren, NL.
2. Van Haperen, P., Voorrips, R.E., Van Loon, J.J.A., Vosman, B. (2018) The effect of plant development on thrips resistance in Capsicum. 2nd International CRC 973 Symposium, April 9 – April 11 2018, Berlin, Germany.
3. Van Haperen, P., Voorrips, R.E., Van Loon, J.J.A., Vosman, B. (2018) The effect of plant development on thrips resistance in Capsicum. TKI Horticulture and Propagation material Network Event 2018, April 3, Nieuwegein, NL.

Abstracts:

1. Van Haperen P., J.J.A. Van Loon, B. Vosman R.E. & Voorrips (2019) The effect of plant and leaf age on thrips resistance in *Capsicum* In: Innovations in Genetics and Breeding of Capsicum and Eggplant (eds V. Lefebvre & M-C Daunay) INRA, Avignon, Fr. PP 56.
2. Voorrips, R.E., G. Steenhuis-Broers, W. van ’t Westende & B. Vosman. (2016) Aphid resistance in a Capsicum collection. In: Proceedings of XVIth EUCARPIA Capsicum and Eggplant working group meeting (eds K. Ertsey-Peregi, Z. Füstos, G Palotás and G. Csilléry) Diamond congress Ltd, Budapest, Hungary. Page 66-68.

Symposium:

1. A symposium on host plant resistance against insects was organized as part of the International Conference on Entomology (ICE2016, Orlando, Fl.). https://esa.confex.com/esa/ice2016/meetingapp.cgi/Session/25614

TV/ radio / social media / newspaper:

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Remaining deliverables (techniques, devices, methods, etc.):

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