

Figure S1. Pictures of the different study sites. **(A)** study site 1, *Lamium album* next to the wheat field. **(B)** study site 1, *Aegopodium podagraria* next to the dirt road. **(C)** study site 2, *B. pascuorum* on *Rubus fruticosus*. **(D)** study site 3, in the center of the photo: *B. pascuorum* on *Scabiosa coumbaria*.



Figure S2. Pictures of the different flowers present in the 3 study sites, presence in the different study sites is indicated by the number between brackets. (A) Symphytum officinale{1-2}; (B) Salva pratensis{3}; (C) Papaver rhoeas{3}; (D) Rubus fruticosus{2}; (E) Lamium album{1-2}; (F) Scabiosa columbaria{3}; (G) Malva neglecta{3}; (H) Geranium pusillum{1}; (I) Leucanthemum vulgare{3}; (J) Aegopodium podagraria{1-2}. All plants in study site 1 and 2 are wild plants and common native species in Belgium. S. officinale is a perennial plant with white or purple tube-shaped flowers, it has a flower period May to August. L. album is a perennial plant and flowers from April to October. It has white flowers produced in verticillasters. A. podagraria is a perennial plant which flowers from June to July. It has small white open flowers which are organized in umbels. G. pusillum is an annual plant with small purple flowers, and flowers from May to October. Rubus fruticosus is a bush from the Rosaceae family with white or pink flowers, flowering from June to August [1]. The flowers in study site 3 were planted, yet, all species are endogenous to Belgium. Salvia pratensis is a perennial plant with different flower colors ranging from violet to pink and white; all plants in study site 3 had violet flowers. The flower period ranges from May to July. Leucanthemum vulgare has composite white-yellow flowers, which are open from May to August. Papaver rhoeas is an annual plant with red flowers, flowering from May to July. Scabiosa columbaria has blue-purple flowers and flowers from July to September. Malva neglecta is a perennial plant with white or blue-purple flowers, all flowers on Site 3 were purple, and it flowers from June to September [2].

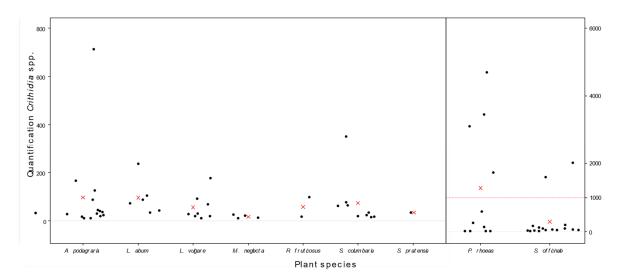


Figure S3. Dot plot of the quantification of *Crithidia* spp. on the flowers (only positive flowers are plotted) per plant species across all study sites. Red × denotes the mean quantity found on a plant species (only taking into account the positive flowers). Red dotted line represents the minimal infection dose if all infective particles are taken up, based upon values reported in literature, i.e. ca. 1000 infective cells [3]. Note the different scale for *P. rhoeas* and *S. officinale*. See also Table S4 for detailed quantification per plant species.

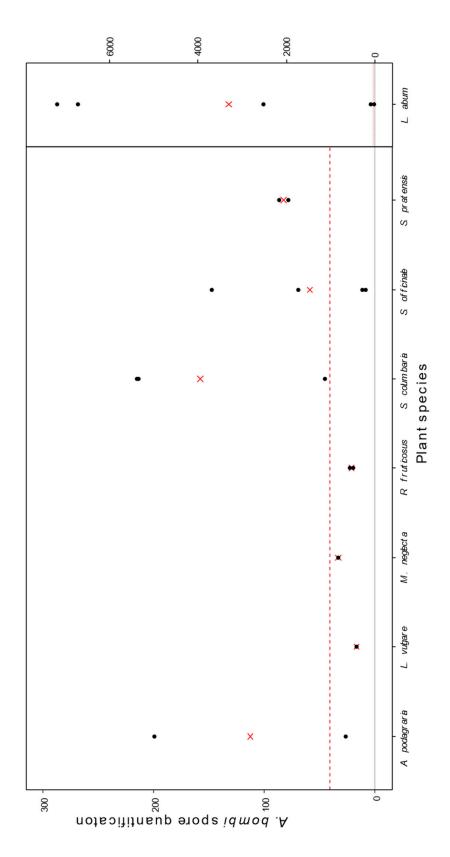


Figure S4. Dot plot of the quantification of *A. bombi* on the flowers (only positive flowers are plotted) per plant species across all study sites. Red × denotes the mean quantity found on a plant species (only taking into account the positive flowers). Red dotted line represents the minimal infection dose if all infective particles are taken up, based upon values reported in literature, i.e. ca. 40 oocysts [4]. Note the different scale for *L. album*. See also Table S 5 for detailed quantification per plant species.

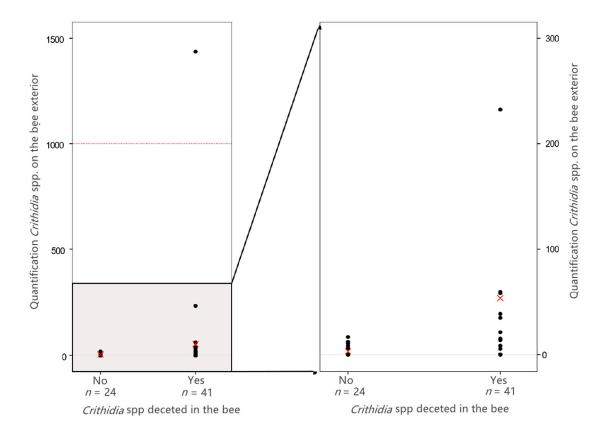


Figure S5. Dot plot of the quantification of *Crithidia* spp. found on the exterior of the bees across sites (i.e. all analyzed bees see Table S6 for more detail) (y-axis) the presence of *Crithidia* spp. within the bee is denoted on the x-axis. Right panel is an enlargement of the shaded rectangular on the left panel. Red × denotes the mean quantity found on the exterior of the bees. Red dotted line on the left panel represents the minimal infection dose if all infective particles are taken up, based upon values reported in literature, i.e. ca. 1000 infective cells [3].

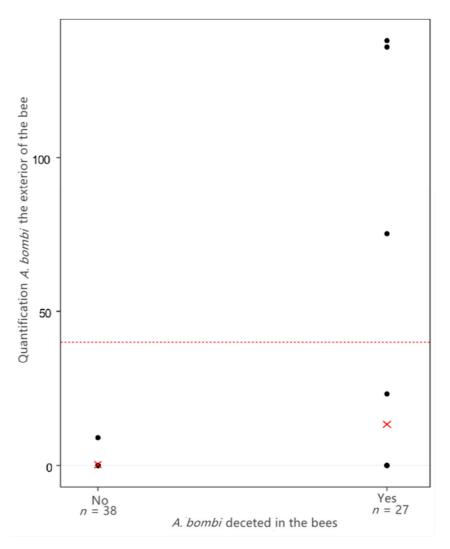


Figure S6. Dot plot of the quantification of *A. bombi* found on the exterior of the bees across sites (i.e. all analyzed bees see Table S6 for more detail) (y-axis) the presence of *A. bombi*. within the bee is denoted on the x-axis. Red × denotes the mean quantity found on the exterior of the bees. Red dotted line represents the minimal infection dose if all infective particles are taken up, based upon values reported in literature, i.e. ca. 40 oocysts [4].

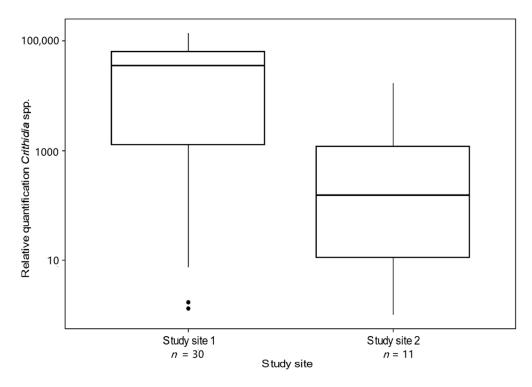


Figure S7. Boxplot of the Relative quantification of Crithidia spp. found in the bees per study site.

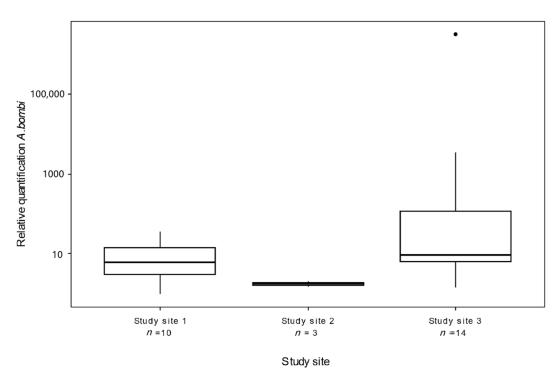


Figure S8. Boxplot of the Relative quantification of *A.bombi* found in the bees per study site.

Table S1. Flower species presence and abundance per species at the study sites. Species in bold were included in the network. The other species were not visited by any pollinator during 30 min monitoring and were therefore not included in the network.

| Study site 1 | | | |
|------------------------------|----------------|--|--|
| Species | Counts | | |
| Lamium album | 198 flowers | | |
| Aegopodium podagraria | 271 umbels | | |
| Symphytum officinale | 221 flowers | | |
| Geranium pusillum | 56 flowers | | |
| Taraxacum officinale | 4 flowers | | |
| Tripleurospermum inodorum | 57 flowers | | |
| Medicago lupulina | 9 flower heads | | |
| Argentina anserina 81 flower | | | |
| Ranunculus acris | 84 flowers | | |
| Achillea millefolium | 22 umbels | | |
| Study site 2 | | | |
| Species | Counts | | |
| Symphytum officinale | 250 flowers | | |
| Aegopodium podagraria | 41 umbels | | |
| Lamium album | 104 flowers | | |
| Rubus fruticosus | 36 flowers | | |
| Study site 3 | | | |
| Species | Counts | | |
| Papaver rhoeas | 18 flowers | | |
| Leucanthemum vulgare | 215 flowers | | |
| Scabiosa columbaria | 120 flowers | | |
| Malva neglecta | 204 flowers | | |
| Salvia pratensis | 83 flowers | | |

Table S2. Pollinator counts from the performed transects at each study site. Dipteran species were only determined up to genus level. Transects were done by walking twice alongside the whole length of the study site and recording the pollinators present on flowers by visual inspection. These transects were not included in the network construction and were only used to assess the overall presence and activity of pollinators at each site. This to set up a monitoring plan for the camera recordings (see section 2.2. Network construction in the main text).

| Study site 1 | | | | | |
|----------------------|-------|--|--|--|--|
| Species | Count | | | | |
| Bombus terrestris | 19 | | | | |
| Bombus pascuorum | 21 | | | | |
| Bombus lapidarius | 6 | | | | |
| Apis mellifera | 2 | | | | |
| Lucilia sp. | 28 | | | | |
| Eristalis sp. | 3 | | | | |
| Study site 2 | | | | | |
| Species | Count | | | | |
| Bombus pascuorum | 14 | | | | |
| Bombus pratorum | 1 | | | | |
| Bombus lapidarius | 1 | | | | |
| Study site3 | | | | | |
| Species | Count | | | | |
| Bombus pascuorum | 5 | | | | |
| Bombus terrestris | 2 | | | | |
| Bombus lapidarius | 1 | | | | |
| Heriades truncorum | 12 | | | | |
| Apis mellifera | 2 | | | | |
| <i>Eristalis</i> sp. | 3 | | | | |

Table S3. An overview of the number of sampled flowers for parasite analysis at each study site. Only the flower heads were sampled, and flowers were cut off in the field; for composite flowers such as *A. podagraria*, the whole umbel was sampled.

| Study site 1 | | | | | |
|-----------------------|---------|--|--|--|--|
| Species | Sampled | | | | |
| Lamium album | 24 | | | | |
| Aegopodium podagraria | 24 | | | | |
| Symphytum officinale | 24 | | | | |
| Geranium pusillum | 10 | | | | |
| Study site 2 | | | | | |
| Species | Sampled | | | | |
| Symphytum officinale | 25 | | | | |
| Aegopodium podagraria | 23 | | | | |
| Lamium album | 25 | | | | |
| Rubus fruticosus | 23 | | | | |
| Study site 3 | | | | | |
| Species | Sampled | | | | |
| Papaver rhoeas | 12 | | | | |
| Leucanthemum vulgare | 22 | | | | |
| Scabiosa columbaria | 20 | | | | |
| Malva neglecta | 21 | | | | |
| Salvia pratensis | 20 | | | | |

Table S4. An overview of the quantification of *Crithidia* spp. of the positive flowers per study site, qPCR quantification was based upon a standard curve (E = 87.2%; $R^2 = 0.991$ from a serial dilution of an in vitro culture of *C. mellificae* counted with a hemocytometer before DNA extraction).

| Study site | Plant species | Sample | Quantification Crithidia spp. | Study site | Plant species | Sample | Quantification Crithidia spp. |
|---------------|---------------|--------|-------------------------------|------------|---------------|--------|-------------------------------|
| 1 | L. album | D17 | 235 | 2 | A. podagraria | LaZ5 | 27 |
| 1 | S. officinale | Sp10 | 2020 | 2 | A. podagraria | LaZ6 | 36 |
| 1 | S. officinale | Sp9 | 1594 | 2 | A. podagraria | LaZ8 | 44 |
| 1 | A. podagraria | Z22 | 712 | 3 | S. columbaria | GeDu1 | 18 |
| 2 | R. fruticosus | LaB10 | 18 | 3 | S. columbaria | GeDu10 | 77 |
| 2 | R. fruticosus | LaB17 | 98 | 3 | S. columbaria | GeDu11 | 24 |
| 2 | L. album | LaD10 | 74 | 3 | S. columbaria | GeDu12 | 65 |
| 2 | L. album | LaD15 | 33 | 3 | S. columbaria | GeDu13 | 18 |
| 2 | L. album | LaD2 | 44 | 3 | S. columbaria | GeDu3 | 62 |
| 2 | L. album | LaD3 | 104 | 3 | S. columbaria | GeDu4 | 15 |
| 2 | L. album | LaD5 | 86 | 3 | S. columbaria | GeDu7 | 349 |
| 2 | S. officinale | LaS1 | 15 | 3 | S. columbaria | GeDu9 | 33 |
| 2 | S. officinale | LaS10 | 62 | 3 | M. neglecta | GeKa1 | 21 |
| 2 | S. officinale | LaS14 | 86 | 3 | M. neglecta | GeKa10 | 9 |
| 2 | S. officinale | LaS16 | 24 | 3 | M. neglecta | GeKa14 | 12 |
| 2 | S. officinale | LaS17 | 33 | 3 | M. neglecta | GeKa6 | 24 |
| 2 | S. officinale | LaS19 | 15 | 3 | P. rhoeas | GeKl1 | 4701 |
| 2 | S. officinale | LaS2 | 189 | 3 | P. rhoeas | GeKl10 | 12 |
| 2 | S. officinale | LaS20 | 59 | 3 | P. rhoeas | GeKl11 | 254 |
| 2 | S. officinale | LaS24 | 18 | 3 | P. rhoeas | GeKl2 | 3450 |
| 2 | S. officinale | LaS25 | 115 | 3 | P. rhoeas | GeKl3 | 580 |
| 2 | S. officinale | LaS3 | 160 | 3 | P. rhoeas | GeKl4 | 6 |
| 2 | S. officinale | LaS4 | 41 | 3 | P. rhoeas | GeKl5 | 3110 |
| 2 | S. officinale | LaS7 | 83 | 3 | P. rhoeas | GeKl6 | 8 |
| 2 | S. officinale | LaS8 | 47 | 3 | P. rhoeas | GeKl7 | 1730 |
| 2 | A. podagraria | LaZ1 | 166 | 3 | P. rhoeas | GeKl8 | 127 |
| 2 | A. podagraria | LaZ11 | 18 | 3 | P. rhoeas | GeKl9 | 11 |
| 2 | A. podagraria | LaZ13 | 86 | 3 | L. vulgare | GeMA1 | 92 |
| 2 | A. podagraria | LaZ14 | 18 | 3 | L. vulgare | GeMA10 | 12 |
| 2 | A. podagraria | LaZ17 | 12 | 3 | L. vulgare | GeMA18 | 68 |
| 2 | A. podagraria | LaZ2 | 24 | 3 | L. vulgare | GeMA19 | 18 |
| 2 | A. podagraria | LaZ20 | 127 | 3 | L. vulgare | GeMA2 | 27 |
| 2 | A. podagraria | LaZ21 | 30 | 3 | L. vulgare | GeMA20 | 30 |
| 2 | A. podagraria | LaZ3 | 41 | 3 | L. vulgare | GeMA22 | 21 |
| 2 | A. podagraria | LaZ4 | 12 | 3 | L. vulgare | GeMA8 | 175 |
| | | | | 3 | S. pratensis | GeVS6 | 33 |

Table S5. An overview of the quantification of *Apicystis bombi* of the positive flowers per study site, qPCR quantification was based upon a standard curve (E = 91.4%; $R^2 = 0.992$ from a serial dilution of oocysts isolated from the fat body of a *B. pascuorum* worker, which were counted with a hemocytometer before DNA extraction).

| Study site | Plant species | Sample | Quantification A. bombi |
|------------|---------------|--------|-------------------------|
| 1 | L. album | D10 | 7186 |
| 1 | L. album | D2 | 6713 |
| 1 | L. album | D13 | 2523 |
| 1 | S. officinale | Sp3 | 147 |
| 1 | L. album | D21 | 110 |
| 2 | A. podagraria | LaZ11 | 199 |
| 2 | S. officinale | LaS19 | 69 |
| 2 | A. podagraria | LaZ3 | 26 |
| 2 | L. album | LaD16 | 23 |
| 2 | R. fruticosus | LaB23 | 22 |
| 2 | R. fruticosus | LaB17 | 19 |
| 2 | S. officinale | LaS20 | 11 |
| 2 | S. officinale | LaS18 | 8 |
| 3 | S. columbaria | GeDU16 | 215 |
| 3 | S. columbaria | GeDU8 | 213 |
| 3 | S. pratensis | GeVS6 | 86 |
| 3 | S. pratensis | GeVS19 | 78 |
| 3 | S. columbaria | GeDU15 | 45 |
| 3 | M. neglecta | GeKa18 | 33 |
| 3 | L. vulgare | GeMa16 | 16 |

Table S6. An overview of the number of sampled bee species at each study site and the number of *C. bombi* and *A. bombi* positive individuals (i.e. parasites were detected internally). Only the most prevalent bee species was sampled for parasite analysis. As in study site 1 the three Bombus species had a similar abundance, all three species were sampled.

| Study site 1 | | | | | |
|--------------------|-----------------|-------------------|-------------------|--|--|
| Species | Sampled | C. bombi positive | A. bombi positive | | |
| Bombus terrestris | errestris 12 11 | | 3 | | |
| Bombus pascuorum | 12 | 10 | 5 | | |
| Bombus lapidarius | 10 | 9 | 2 | | |
| Study site 2 | | | | | |
| Species | Sampled | C. bombi positive | A. bombi positive | | |
| Bombus pascuorum | 16 | 11 | 3 | | |
| Study site 3 | | | | | |
| Species | Sampled | C. bombi positive | A. bombi positive | | |
| Heriades truncorum | 15 | 0 | 14 | | |

Table S7. Results of the GLMM (using Model 2, see main text) for the separate parasites. The presence/absence data of *A. bombi* or *Crithidia spp*. (both confirmed by Sanger sequencing) served as a binomial response variable for which the link function log of the odds ratio (logit) was used. The natural logarithm of the normalized weighted closeness was used as a fixed variable. Site and flower species served as random variable, where flower species was nested within site.

| Crithidia spp. | | | | | |
|----------------------|--------|----|-----------|---------|--|
| Fixed effect | β | df | □2 | p-value | |
| Normalised closeness | 4.458 | 4 | 7.852 | 0.005 | |
| Apicystis bombi | | | | | |
| Fixed effect | β | df | □2 | p-value | |
| Normalised closeness | 0.8816 | 4 | 0.835 | 0.361 | |

References

- 1 H. Eggeltje and D. Tande Lid, Veldgids Nederlandse flora, 9e ed. Zeist: KNNV uitgeverij, 2015.
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- 3 M. C. Otterstatter and J. D. Thomson, "Does pathogen spillover from commercially reared bumble bees threaten wild pollinators?," *PLoS One*, vol. 3, no. 7, p. e2771, Jan. 2008, doi: 10.1371/journal.pone.0002771.
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