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**Appendix I: Symbiont list identifying potentially deleterious symbionts of concern for clean stock and commercial bumble bee rearing**

The list has been sorted into two groupings based on the concern of the symbiont’s presence in rearing operations: Priority/Of Concern and High Uncertainty but Potential, with select evidence presented in the main text summarized here. It is important to note that this is likely not a fully inclusive list, given current unknowns, and new symbionts of concern are likely to arise. Any clean stock program should visually monitor for problematic symptoms in individuals and colonies and verify causative agents.

**Priority/Of Concern**

Symbiont	Evidence
<b>Protozoa</b>	
<i>Apicystis bombi</i> Liu, MacFarlane, and Pengelly; Ophrocystidae	High virulence (Jones & Brown 2014; Rutrecht & Brown 2008); Potential for spillover (Graystock et al. 2013b); Possibly introduced with non-native <i>Bombus</i> (Arbetman et al. 2013)
<i>Crithidia bombi</i> Lipa and Triggiani; Trypanosomatidae	Virulence context dependent, but may be high (Sadd & Barribeau 2013); Easily transmitted and has been common in commercial colonies (Graystock et al. 2013b; Murray et al. 2013); High spillover potential (Colla et al. 2006)
<i>Crithidia expoeki</i> Schmid-Hempel and Tognazzo; Trypanosomatidae	Limited study, but assumed like <i>C. bombi</i>
<b>Fungi</b>	
<i>Vairimorpha (Nosema) bombi</i> Fantham and Porter; Microsporidia: Nosematidae	High virulence (Otti & Schmid-Hempel 2007; Otti & Schmid-Hempel 2008); Found in commercial distributed colonies (Graystock et al. 2013b; Murray et al. 2013)
<i>Vairimorpha (Nosema) ceranae</i> Fries et al.; Microsporidia: Nosematidae	Bumble bees susceptible, increases mortality (Graystock et al. 2013a)
<b>Viruses (Li et al. 2011)</b>	
<i>Deformed wing virus (DWW)</i> ; Iflaviridae	Replication in bumble bees (Levitt et al. 2013; Li et al. 2011); Pathology in commercially reared bumble bees (Genersch et al. 2006)
<i>AKI-complex: Kashmir bee virus (KBV); Acute Bee Paralysis Virus (ABPV), Kashmir Bee Virus (KBV), Israeli Acute Paralysis Virus (IAPV)</i> Dicistroviridae	Infective to bumble bees, long-lasting infectivity (Bailey & Gibbs 1964); Can be common in bumble bees (McMahon et al. 2015); Virulence may be dose- and transmission route-dependent, but can be high (Meeus et al. 2014; Niu et al. 2016; Wang et al. 2018)

### High Uncertainty but Potential

Symbiont	Evidence
<b>Bacteria</b>	
<i>Spiroplasma apis</i> Mouches et al.; Mollicutes: Spiroplasmataceae	Disease association in honey bees and presence of bacteria in bumble bees indicates pathogenic potential, but not verified (Clark et al. 1985; Meeus et al. 2012)
<i>Spiroplasma melliferum</i> Clark et al.; Mollicutes: Spiroplasmataceae	Associated with disease in honey bees and presence of bacteria in bumble bees indicates pathogenic potential, but not verified (Clark et al. 1985; Meeus et al. 2012)
<b>Viruses</b>	
<i>Black queen cell virus</i> (BQCV); <i>Chronic Bee Paralysis Virus</i> (CBPV); <i>Cloudy Wing Virus</i> (CWV); <i>Sacbrood virus</i> (SBV); <i>Slow Bee Paralysis Virus</i> (SBPV); Other “honey bee” RNA viruses	Found in bumble bees, including in commercial colonies (Choi et al. 2010; McMahon et al. 2015; Peng et al. 2011; Sachman-Ruiz et al. 2015; Singh et al. 2010); Need study to understand further occurrence and virulence in bumble bees (Tehel et al. 2016)
<b>Fungi</b>	
<i>Tubulosema pampeana</i> Plischuk et al.; Microsporidia; Tubulosematidae	Detected in bumble bees, affecting adipose tissue (Plischuk et al. 2017; Plischuk et al. 2015); Currently further pathology and effects unknown.

### Literature Cited

- Arbetman MP, Meeus I, Morales CL, Aizen MA, Smagghe G (2013) Alien parasite hitchhikes to Patagonia on invasive bumblebee. *Biological Invasions* 15: 489-494. <https://doi.org/10.1007/s10530-012-0311-0>
- Bailey L, Gibbs J (1964) Acute infection of bees with paralysis virus. *Journal of Insect Pathology* 6: 395-407.
- Choi Y, Lee M, Hong I, Kim N, Kim H, Byeon K, Yoon H (2010) Detection of honeybee virus from bumblebee (*Bombus terrestris* L. and *Bombus ignitus*) Korean Journal of Apiculture 25: 259-266.
- Clark TB, Whitcomb RF, Tully JG, Mouches C, Saillard C, Bové, J. M., Wróblewski H, Carle P, Rose DL, Henegar RB, Williamson DL (1985) *Spiroplasma melliferum*, a new species from the honeybee (*Apis mellifera*). *International Journal of Systematic and Evolutionary Microbiology* 35: 296-308. <https://doi.org/10.1099/00207713-35-3-296>.
- Colla SR, Otterstatter MC, Gegear RJ, Thomson JD (2006) Plight of the bumble bee: Pathogen spillover from commercial to wild populations. *Biological Conservation* 129: 461-467. <https://doi.org/10.1016/j.biocon.2005.11.013>
- Genersch E, Yue C, Fries I, de Miranda JR (2006) Detection of Deformed wing virus, a honey bee viral pathogen, in bumble bees (*Bombus terrestris* and *Bombus pascuorum*) with wing deformities. *J Invertebr Pathol* 91: 61-63. <https://doi.org/10.1016/j.jip.2005.10.002>

- Graystock P, Yates K, Darvill B, Goulson D, Hughes WOH (2013a) Emerging dangers: Deadly effects of an emergent parasite in a new pollinator host. *Journal of Invertebrate Pathology* 114: 114-119. doi:<https://doi.org/10.1016/j.jip.2013.06.005>.
- Graystock P, Yates K, Evison SEF, Darvill B, Goulson D, Hughes WOH (2013b) The Trojan hives: pollinator pathogens, imported and distributed in bumblebee colonies. *Journal of Applied Ecology* 50: 1207-1215. <https://doi.org/10.1111/1365-2664.12134>
- Jones CM, Brown MJF (2014) Parasites and genetic diversity in an invasive bumblebee. *Journal of Animal Ecology* 83: 1428-1440. doi:<https://doi.org/10.1111/1365-2656.12235>.
- Levitt AL, Singh R, Cox-Foster DL, Rajotte E, Hoover K, Ostiguy N, Holmes EC (2013) Cross-species transmission of honey bee viruses in associated arthropods. *Virus Res* 176: 232-240. <https://doi.org/10.1016/j.virusres.2013.06.013>
- Li J, Peng W, Wu J, Strange JP, Boncristiani H, Chen Y (2011) Cross-species infection of Deformed Wing Virus poses a new threat to pollinator conservation. *Journal of Economic Entomology* 104: 732-739. <https://doi.org/10.1603/EC10355>
- McMahon DP, Fürst MA, Caspar J, Theodorou P, Brown MJF, Paxton RJ (2015) A sting in the spit: widespread cross-infection of multiple RNA viruses across wild and managed bees. *Journal of Animal Ecology* 84: 615-624. <https://doi.org/10.1111/1365-2656.12345>
- Meeus I, de Miranda JR, de Graaf DC, Wäckers F, Smagghe G (2014) Effect of oral infection with Kashmir bee virus and Israeli acute paralysis virus on bumblebee (*Bombus terrestris*) reproductive success. *Journal of Invertebrate Pathology* 121: 64-69. <https://doi.org/10.1016/j.jip.2014.06.011>.
- Meeus I, Vercauteren V, Smagghe G (2012) Molecular detection of *Spiroplasma apis* and *Spiroplasma melliferum* in bees. *Journal of Invertebrate Pathology* 109: 172-174. <https://doi.org/10.1016/j.jip.2011.11.006>.
- Murray TE, Coffey MF, Kehoe E, Horgan FG (2013) Pathogen prevalence in commercially reared bumble bees and evidence of spillover in conspecific populations. *Biological Conservation* 159: 269-276. <http://dx.doi.org/10.1016/j.biocon.2012.10.021>.
- Niu J, Smagghe G, De Coninck DIM, Van Nieuwerburgh F, Deforce D, Meeus I (2016) In vivo study of Dicer-2-mediated immune response of the small interfering RNA pathway upon systemic infections of virulent and avirulent viruses in *Bombus terrestris*. *Insect Biochemistry and Molecular Biology* 70: 127-137. <https://doi.org/10.1016/j.ibmb.2015.12.006>.
- Otti O, Schmid-Hempel P (2007) *Nosema bombi*: A pollinator parasite with detrimental fitness effects. *Journal of Invertebrate Pathology* 96: 118-124. <https://doi.org/10.1016/j.jip.2007.03.016>.
- Otti O, Schmid-Hempel P (2008) A field experiment on the effect of *Nosema bombi* in colonies of the bumblebee *Bombus terrestris*. *Ecological Entomology* 33: 577-582. <https://doi.org/10.1111/j.1365-2311.2008.00998.x>
- Peng W, Li J, Boncristiani H, Strange JP, Hamilton M, Chen Y (2011) Host range expansion of honey bee Black Queen Cell Virus in the bumble bee, *Bombus huntii*. *Apidologie* 42: 650-658. <https://doi.org/10.1007/s13592-011-0061-5>
- Plischuk S, Salvarrey S, Arbulo N, Santos E, Skevington JH, Kelso S, Revainera PD, Maggi MD, Invernizzi C, Lange CE (2017) Pathogens, parasites, and parasitoids associated with bumble bees (*Bombus* spp.) from Uruguay. *Apidologie* 48: 298-310. <https://doi.org/10.1007/s13592-016-0474-2>
- Plischuk S, Sanscrainte ND, Becnel JJ, Estep AS, Lange CE (2015) *Tubulosema pampeana* sp. n. (Microsporidia, Tubulosematidae), a pathogen of the South American bumble bee *Bombus atratus*. *Journal of Invertebrate Pathology* 126: 31-42. doi:<https://doi.org/10.1016/j.jip.2015.01.006>.
- Rutrecht ST, Brown MJF (2008) The life-history impact and implications of multiple parasites for bumble bee queens. *International Journal for Parasitology* 38: 799-808. <https://doi.org/10.1016/j.ijpara.2007.11.004>.
- Sachman-Ruiz B, Narváez-Padilla V, Reynaud E (2015) Commercial *Bombus impatiens* as reservoirs of emerging infectious diseases in central México. *Biological Invasions* 17: 2043-2053. <https://doi.org/10.1007/s10530-015-0859-6>

- Sadd BM, Barribeau SM (2013) Heterogeneity in infection outcome: lessons from a bumblebee-trypanosome system. *Parasite Immunology* 35: 339-349. doi:<https://doi.org/10.1111/pim.12043>.
- Singh R, Levitt AL, Rajotte EG, Holmes EC, Ostiguy N, vanEngelsdorp D, Lipkin WI, dePamphilis CW, Toth AL, Cox-Foster DL (2010) RNA viruses in Hymenopteran pollinators: Evidence of inter-taxa virus transmission via pollen and potential impact on non-*Apis* Hymenopteran species. *PLOS ONE* 5: e14357. <https://doi.org/10.1371/journal.pone.0014357>
- Tehel A, Brown MJF, Paxton RJ (2016) Impact of managed honey bee viruses on wild bees. *Current Opinion in Virology* 19: 16-22. <https://doi.org/10.1016/j.coviro.2016.06.006>.
- Wang H, Meeus I, Piot N, Smaghe G (2018) Systemic Israeli acute paralysis virus (IAPV) infection in bumblebees (*Bombus terrestris*) through feeding and injection. *Journal of Invertebrate Pathology* 151: 158-164. <https://doi.org/10.1016/j.jip.2017.11.015>