

Why don't queens get broody?

Parasites, mating status, and ovary development in *Bombus huntii* (Hymenoptera: Apidae).

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INTRODUCTION & BACKGROUND

- Bumble bees provide essential greenhouse pollination services¹.
- Concerns of introducing *Bombus* spp. and their parasites into novel environments has led to developing local species to provide pollination services^{2,3}.
- Bombus huntii*, native to intermountain regions of western North America, is an effective greenhouse tomato pollinator^{2,3}.
- 51% of wild-caught *B. huntii* queens did not initiate nests in captivity in 2015 and 2016.
- Parasite pressures on *Bombus* queen fecundity are poorly understood.

OBJECTIVES

- Diagnose parasites and pathogens, ovary development, and mating status of wild-caught *B. huntii* queens that failed to initiate nests.
- Assess if parasites alter ovary development.

MATERIALS AND METHODS

- Culled (-80° C) wild-caught (Fig. 1) *B. huntii* queens failing to initiate nests after 21 days in captivity (2015: n = 48; 2016: n = 32).
- Dissected and assessed macroparasites and ovary development (10x, Fig. 2).
- Slide mounted tissue samples and analyzed microparasites and sperm (400x).
- Extracted DNA with salting-out procedure⁴.
- Conducted PCR with microparasite-specific primers⁵ and positive and negative controls.
- Visualized PCR products for the presence of diagnostic bands on agarose gels (Fig. 4).
- Assessed consistency of mating status and relationship between parasite presence and ovary development with Fisher's Exact Tests implemented in R⁶.



Fig 1: Green Canyon, the collection site for many of the analyzed queens.



Fig 2: Queen dissection, showing the viscera and body cavity examined for parasites.

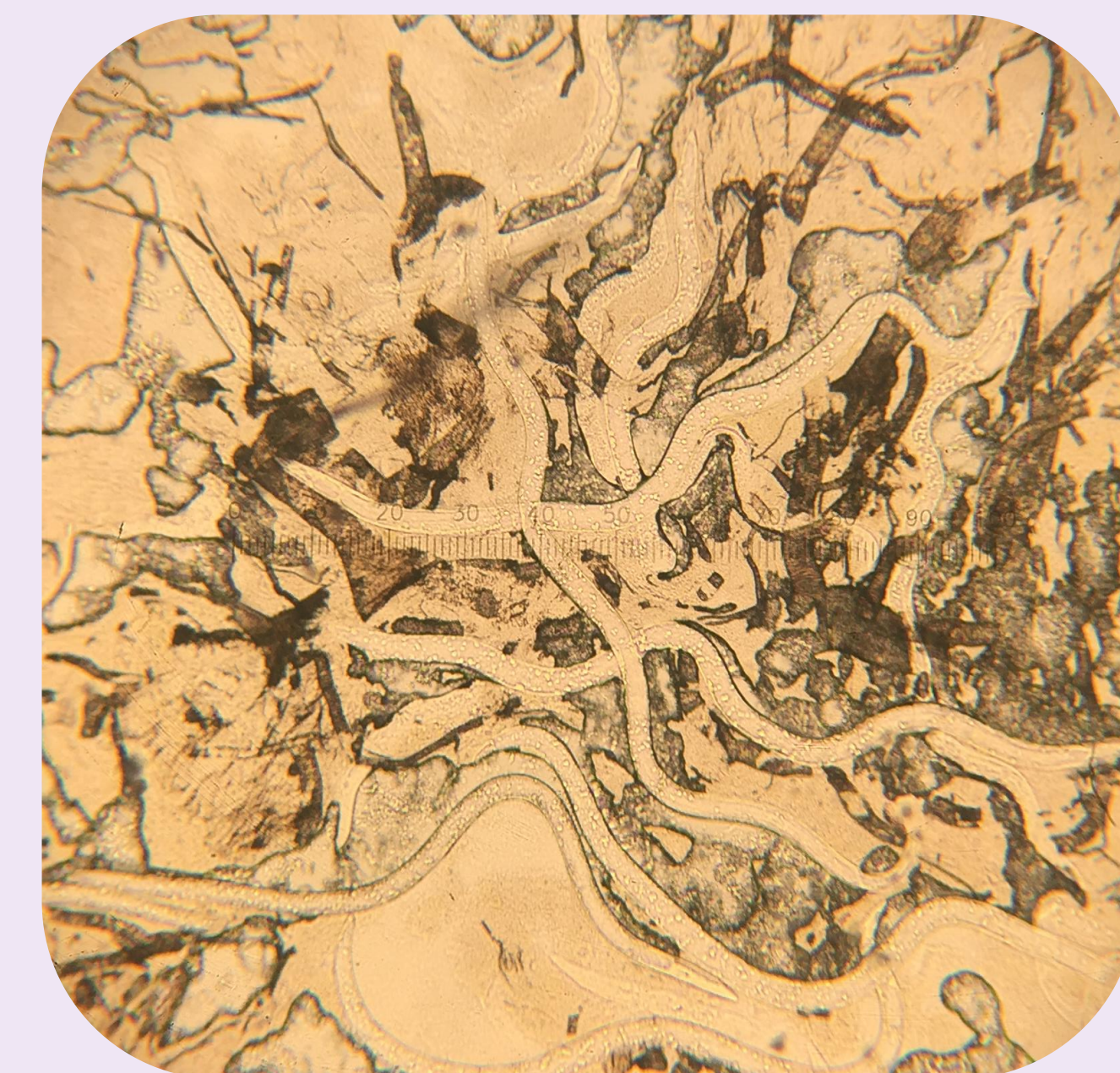


Fig 3: Slide mounted juvenile *Sphaerularia bombi* in tissue sample at 400x.

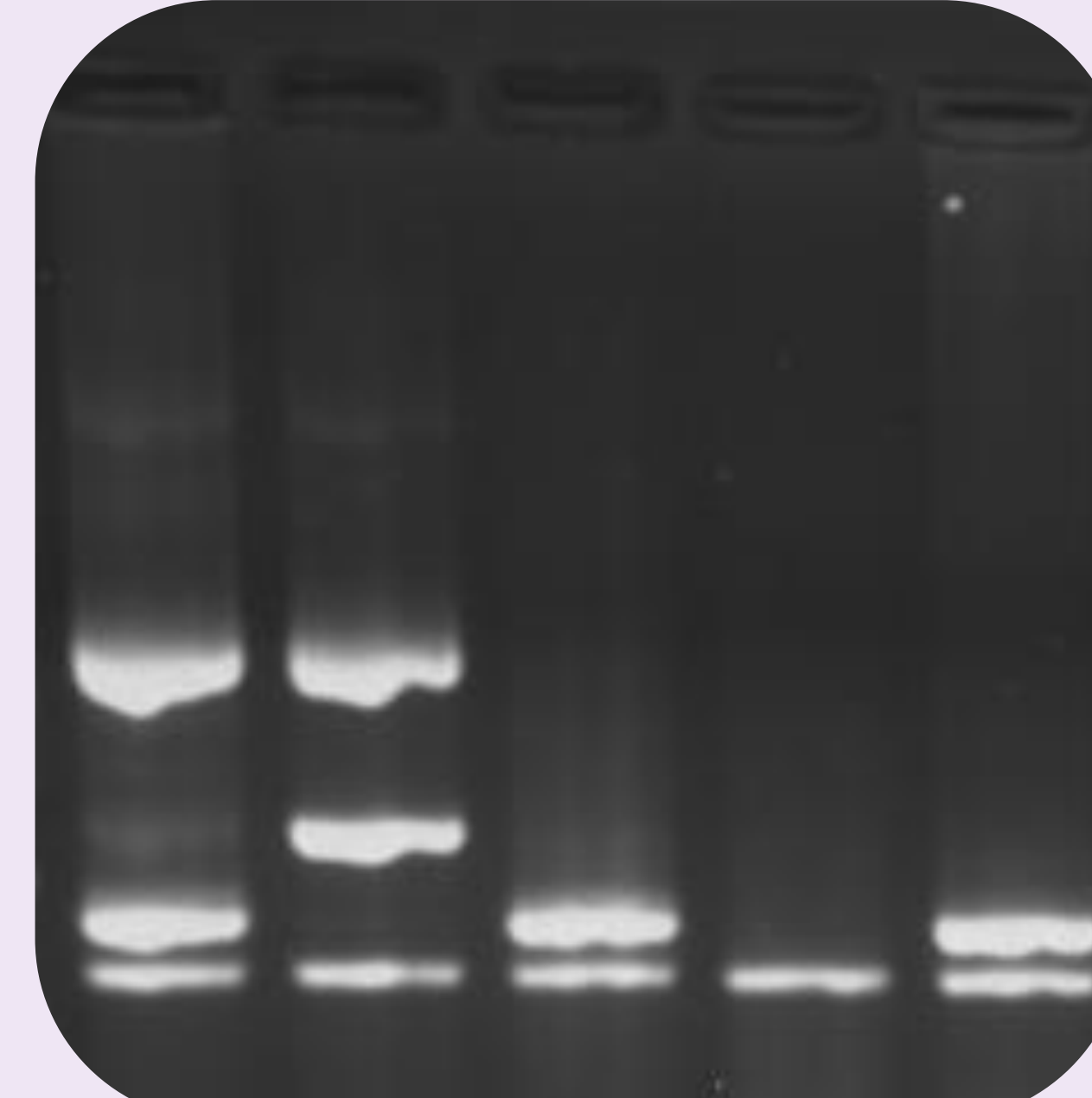
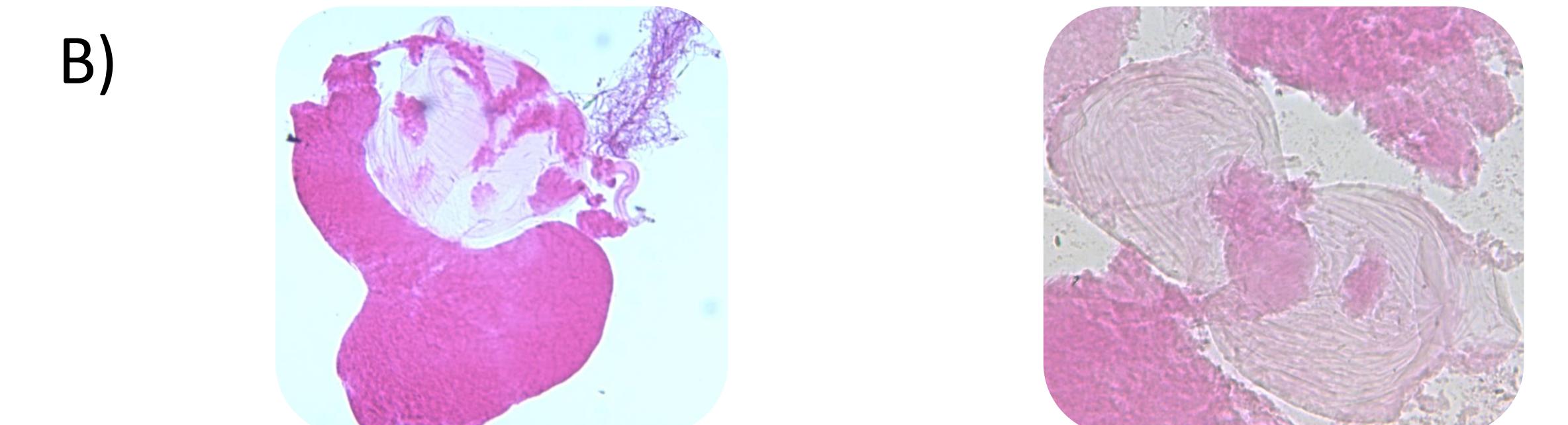
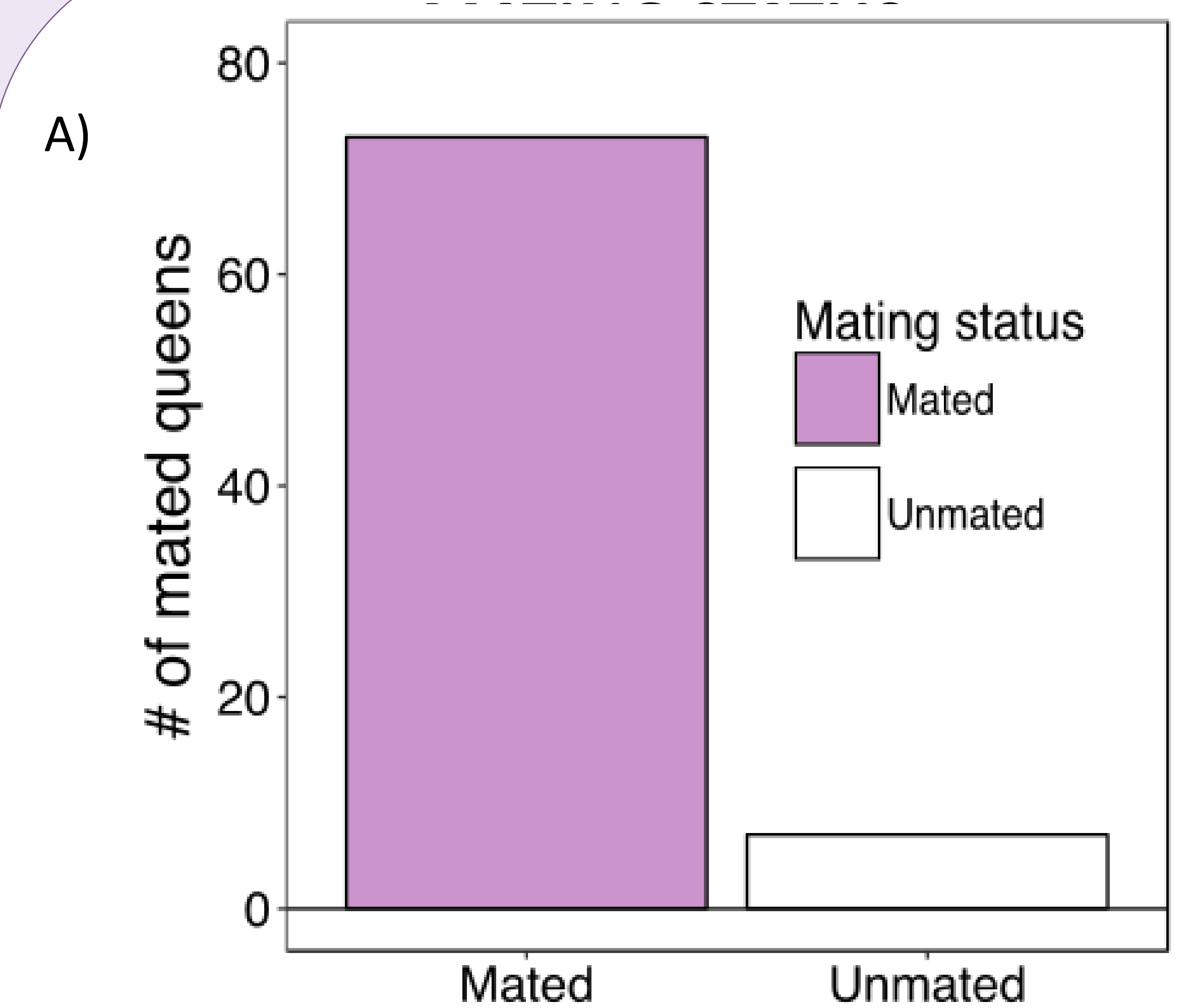


Fig 4: PCR products visualized in UV on an agarose gel to diagnose microparasites. Photo by Amber Tripodi.



Slide mounted spermatheca and sperm (dark pink) of mated queen (100x). Slide mounted spermatheca of unmated queen (100x).
 Fig 6: A) Mating status (presence or absence of sperm) B) spermathecae with (left) and without (right) sperm (100x).

PARASITISM

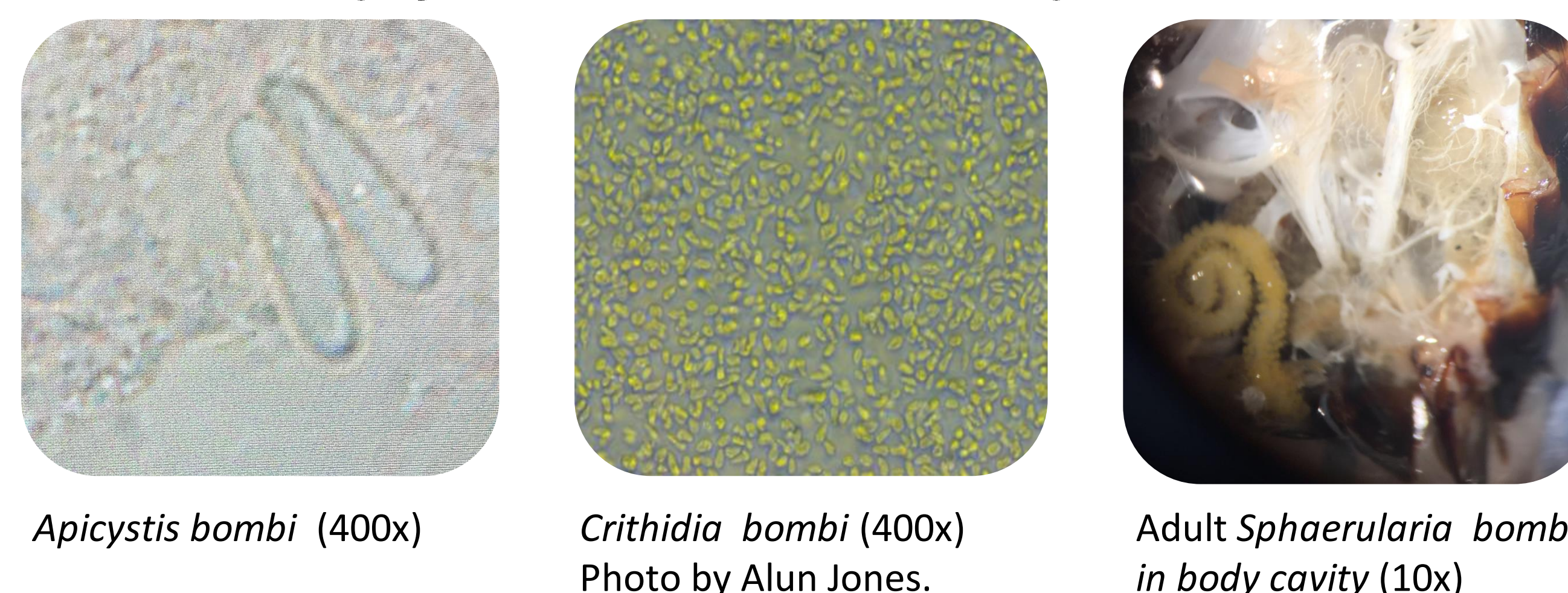
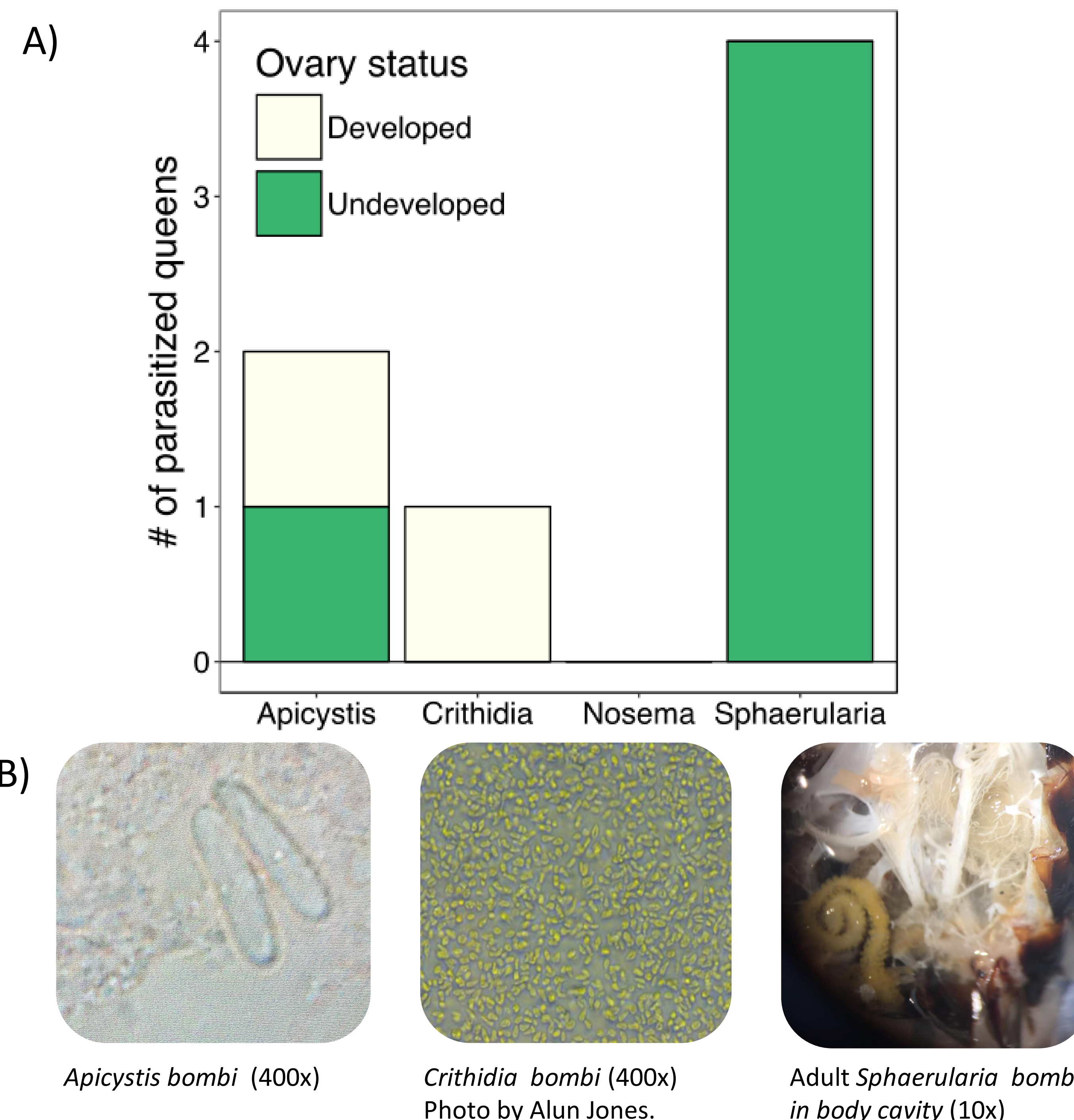


Fig 5: A) prevalence of each parasite by ovary development in broodless queens (2015 and 2016 combined). Fisher's Test p-value = 0.4286; B) parasites present in queens in this study.

RESULTS AND DISCUSSION

- Parasitism was rare, and not associated with ovary development (Fig. 5).
- No conopids, phorids, tracheal mites, *Nosema*, mermithids, or parasitic wasps were observed.
- Two queens had *Apicystis*, and one had *Crithidia* (Fig. 5).
- Sphaerularia bombi*, the queen-castrating nematode, was found in four queens (Fig. 5).
- All 2015 queens were mated and only seven were unmated in 2016, meaning mating status does not explain broodlessness (Fig. 6).
- Rearing room conditions and nutrition are potential factors leading to the broodless queen conundrum.

FUTURE DIRECTIONS

- Assess and compare queens that died before 21 days in captivity, successful nest founding queens, 2017 queens, and other candidate commercial pollination species (*B. vosnesenskii*).
- Evaluate effects of nutrition and rearing facility conditions on nest initiation.

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