

#### 4. Research report (Follow the guideline on the next page)

### **BACKGROUND**

Insect pests develop resistance to insecticides and represents a serious management program. The goals of Integrated Pest Management (IPM) are to devise methods that concurrently delay resistance and minimize the injury made by insects to field crops. Therefore, one of the centerpieces of IPM is the use of biological control agents. Researchers are now beginning to appreciate the usefulness of genomic tools in IPM towards management decisions. Genomics tools could characterize pest populations to gain a greater depth of knowledge for the development of certain pest control strategies. Recent advances of genome sequencing technologies allow the detection of large-scale gene polymorphisms, which are becoming economically feasible for insect pest. Genetic information such as temporal gene fluctuations, spatial distribution and ecological diversification within target, non-target, and natural enemy (e.g., parasitoids) populations can be utilized to predict insect pest outbreaks for deciding on its application control measures. With the insights gained by the researchers in terms of its morphology, ecology and behavior through the years, genomics tools can further deepen our understanding of the source population, route, and the mechanism of spread of the pest in different parts of the Philippines. There is a need to resolve the species complexes conundrum of *Comperiella calauanica* populations with differing ecological behavior as observed in the field.

### **METHODOLOGY**

*Comperiella sp.* samples were collected by BCRU in selected parts of Luzon and Mindanao as well as museum samples from the Ehime University Entomological Museum collection. All samples were DNA extracted using the QIAGEN Blood and Tissue DNEasy Kit (Qiagen, Hilden, Germany) following the modified protocol of Crane (2011). The amplification of the CO1 was carried out using the specified primers and thermal conditions from Serrana et al., (2020). PCR products were purified using the QIAquick 96 PCR Purification Kit (QIAGEN) following the manufacturer's instructions and sequenced in both directions by Eurofin Genomics, Inc, Tokyo, Japan. Sequences were assembled using CodonCode Aligner v. 5.1.5 (CodonCode Corporation, Dedham, MA, USA). Sequences were aligned via MAFFT v. 7.409 and the ambiguously aligned regions will be excluded using GBlocks 0.91b.

### **RESULTS**

Two *Comperiella* populations were collected in Laguna and Basilan and identified as *Comperiella calauanica*. However, preliminary CO1 analysis revealed otherwise with reference sequences from Vietnam (Figure 1). The preliminary CO1 analysis showed that the Laguna and Basilan populations diverged indicating there is a possible geographic separation of the two populations. Further analysis revealed that there is a genetic difference of 96% between the two populations suggesting that Basilan populations could be a new species. This prompted the researchers to

investigate the morphology and revealed that Basilan population is indeed a new species, *Comperiella basilan* (Barrion et al., 2022). Furthermore, it has been observed that the parasitoid rates of these species toward *Aspidiotus rigidus* are very low compared to *C. calauanica*.

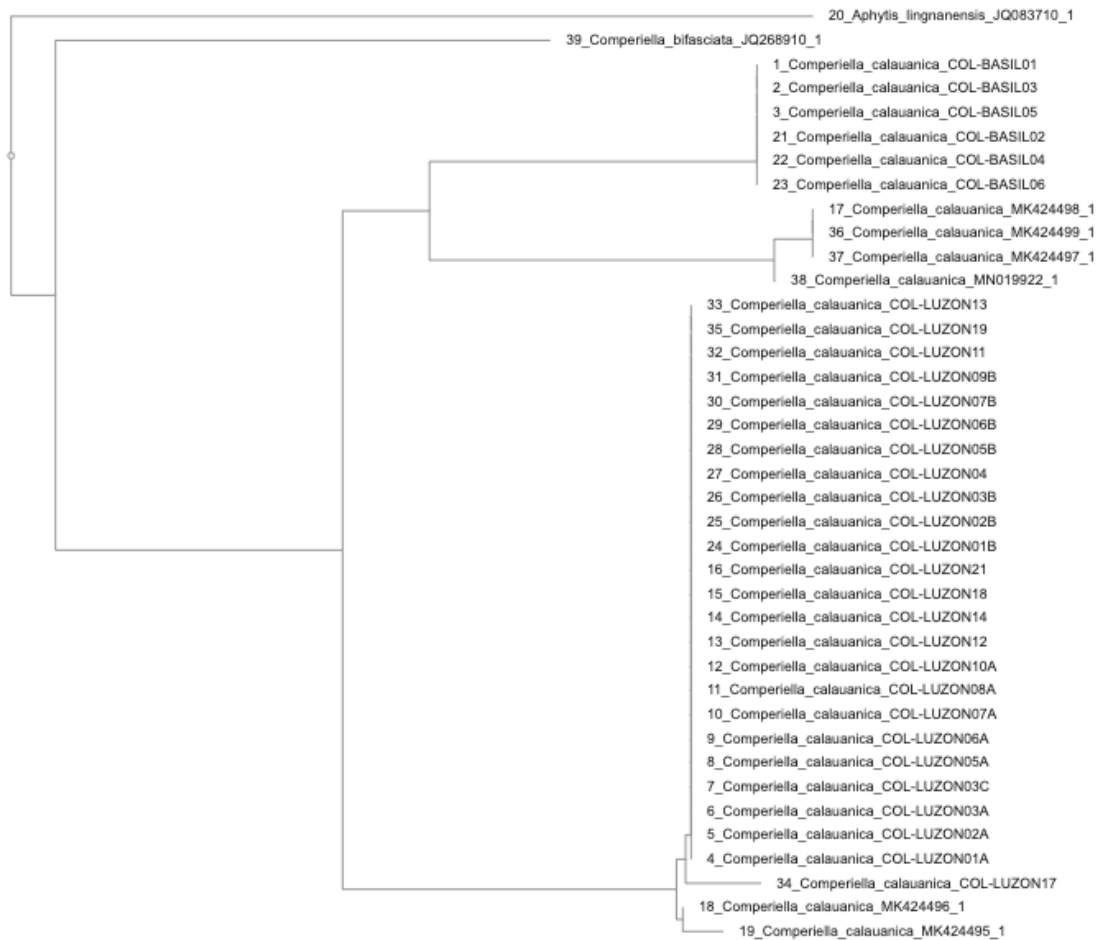


Figure 1. Preliminary analysis of *Comperiella calauanica* from Laguna and Basilan populations.

On the other hand, Figure 2 shows an updated phylogenetic relationship of *C. basilan* with other *Comperiella* species such as *C. bifasciata* and *C. indica*. It pointed out by an expert that the new species could be *C. indica*. Since the present researchers do not have any samples of *C. indica* to evaluate its morphology, the researchers obtained its reference CO1 sequence from NCBI. This revealed that *C. indica* is genetically different from *C. basilan*. Furthermore, the researchers included in this tree *C. calauanica* sequences from Vietnam (Dao et al., 2020). Some of the identified *C. calauanica* in Vietnam could be potentially misidentified as *C. basilan*. The red brackets indicate groupings for *C. calauanica*, *C. basilan*, and *C. indica*.

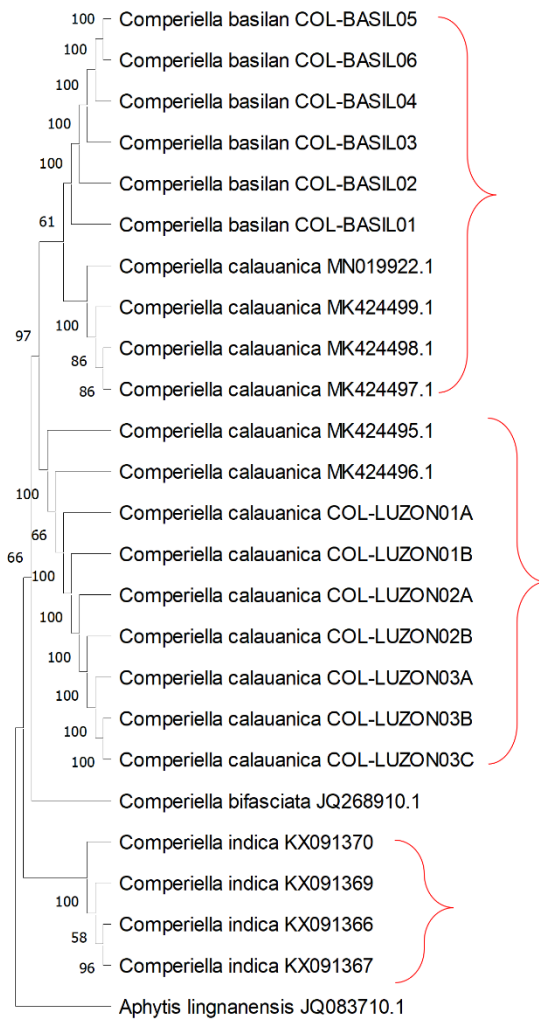


Figure 2. Phylogenetic analysis of *Comperiella calauanica*, *C. basilan* and *C. indica*.