

A scientific note on genetic profile of the mite *Varroa destructor* infesting apiaries in Rio Grande do Sul state, Brazil

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The *Varroa destructor* mite is a hematophagous ectoparasite widely known as the most important *Apis mellifera* parasite; it causes severe economic impact on beekeeping, since in most regions of the world colonies infested with *V. destructor* eventually collapse (Strapazzon et al. 2009; Rosenkranz et al. 2010). Different mite haplotypes present distinct reproductive capacity and virulence factors (Anderson and Trueman 2000; Garrido et al. 2003; Strapazzon et al. 2009), which contribute to infestation rates and consequent damage to honey bee colonies.

We examined the prevalence of Japan (*J*) and Korean (*K*) mitochondrial haplotypes of the mites infesting *A. mellifera* from southwestern Rio Grande do Sul state, an important Brazilian honey-producing region that borders Argentina (to the west) and Uruguay (to the south), both of which have severe problems with *V. destructor* infestations. Given the higher infestation levels in cold regions of Brazil (Moretto et al. 1991) and the tendency for beekeepers in this southernmost region of the country to copy management practices used in the neighboring countries, there is a considerable concern about *Varroa* infestations.

This study was carried out between May 2013 and November 2015 and sampled phoretic adult females of *V. destructor*, 47 in total, from adult workers from arbitrarily selected beehives. One known *V. destructor* haplotype *J* mite sample from Fernando de Noronha Island

(Pernambuco State) was collected and used for comparison. The molecular analysis of the CO-I mitochondrial gene using PCR-restriction enzyme, as described in Anderson and Fuchs (1998), and sequencing and alignment experiments indicated all 47 mite samples of the southwestern region of Rio Grande do Sul state belong to the Korean (*K*) haplotype pattern. Only the Fernando de Noronha Island sample showed the haplotype *J* pattern, although the Japan (*J*) haplotype was formerly reported in the region (Strapazzon et al. 2009). All PCR amplicons presented the expected 458 base pairs and had 100% identity to previously reported sequences.

The sequences obtained from our southwestern samples (GenBank accession numbers: KX458254.1 and KY380010-KY380055) were identical to one another and to the previously reported *V. destructor* Korean (*K*) haplotype, with the same genetic pattern that was found in Argentina (Guerra et al. 2010). As expected, only the *Xho*I restriction site was observed. The sequence obtained from the Fernando de Noronha Island sample (GenBank accession number: KX458253.1) were identical to the previously reported *V. destructor* Japan haplotype (Anderson and Trueman 2000) and showed the presence of both *Xho*I and *Sac*I restriction sites. None of the southwestern samples were cut by the *Sac*I endonuclease, as it is a characteristic of the *V. destructor* Korean haplotype, while the Fernando Noronha Island samples were cut by *Sac*I, as it is a characteristic of *V. destructor* Japan haplotype, as reported before (Anderson and Trueman 2000).

These data agree with previous studies that showed that the Japanese haplotype has more

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restricted distribution than the Korean haplotype (Anderson and Trueman 2000; Strapazzon et al. 2009; Rosenkranz et al. 2010) and was only found in Brazil in Fernando de Noronha Island (Guerra et al. 2010), Vale do Paraíba (São Paulo State), and Sul Fluminense (Rio de Janeiro State) (Pinto 2012).

We now have a new evidence to corroborate that the *V. destructor* Korean haplotype population is growing rapidly in southern Brazil, where the Japan haplotype formerly existed (Anderson 2000; Anderson and Trueman 2000; Carneiro et al. 2007) and support the hypothesis of genotype substitution occurring over time (Anderson and Trueman 2000; Guerra et al. 2010; Pinto 2012).

The intensity of damage caused by *V. destructor* mites has been shown to vary according to the infestation levels in a given region (Moretto and Mello 1999; Guerra et al. 2010; Pinto 2012). The honey bees present in the sampled region are characterized as Africanized bees (Wallberg et al. 2014), and it appears that the Africanized honey bees in Brazil are able to keep infestation rates at low levels, compared to what has been found in other countries (Moretto et al. 1991; Strapazzon et al. 2009); climate could be one of the factors involved (De Jong et al. 1984), despite the predominance of the *K* haplotype. Our results clarify the nature of the southwest Rio Grande do Sul *V. destructor* mite population and suggest that interactions between environment and reproduction ability of genetically distinct populations of *V. destructor* infesting Africanized honey bees throughout Brazil may play a significant role in prevalence and levels of infections. To our knowledge, this is the first comprehensive *V. destructor* genetic profile survey in this region bordering between subtropical and temperate regions of Brazil.

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Contributions CEOS and CEPL performed the experiments, participated in the interpretation of data, and wrote the paper. DDJ, PMP, ADC, and JTB conceived this research, designed the experiments, and reviewed the manuscript.

Note scientifique sur le profil génétique de l'acarien *Varroa destructor* infestant les ruchers dans l'État du Rio Grande do Sul, au Brésil.

Eine wissenschaftliche Notiz über genetisches profil der milbe *Varroa Destructor* befallen Bienenstöcke in Rio Grande do Sul Staat, Brasilien.

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