Journal of Invertebrate Pathology 206 (2024) 108168

Contents lists available at ScienceDirect

Journal of Invertebrate Pathology

journal homepage: www.elsevier.com/locate/jip

Occurrence of *Nosema ceranae, Ascosphaera apis* and trypanosomatids in *Vespa orientalis* linneus 1771

Karen Power [a,*,1], Giovanni Cilia [b,1], Ernesto Ragusa [c], Roberto Rizzo [d], Laura Bortolotti [b], Paola Maiolino [e]

[a] Department of Biology, University of Naples Federico II, 80126 Naples, Italy
[b] Council for Agricultural Research and Agricultural Economics Analysis, Centre for Agriculture and Environment Research (CREA-AA), 40128 Bologna, Italy
[c] Department of Agricultural, Food and Forest Sciences, University of Palermo, 90128 Palermo, Italy
[d] Council for Agricultural Research and Agricultural Economics Analysis, Research Centre for Plant Protection and Certification, 90145 Palermo, Italy
[e] Department of Veterinary Medicine and Animal Productions, University of Naples Federico II, 80137 Naples, Italy

ARTICLE INFO

Keywords:
Grocot
Histopathology
Microsporidia
Oriental hornet
Trypanosomatida

ABSTRACT

Vespa orientalis is spreading across the Italian and European territories leading to new interactions among species, which could lead to the transmission of pathogens between species. Detection of honey bee viruses in *V. orientalis* has already been revealed in both adults and larvae, while no information is available regarding parasitic occurrence. Sixty adult hornets collected across apiaries in the South of Italy were subjected to cytological, histopathological and biomolecular examination to evaluate the occurrence of *Nosema ceranae*, *Ascosphaera apis*, *Lotmaria passim, Crithidia mellificae,* and *Crithidia bombi.*

Cytological examination revealed the presence of *Nosema* spores in 38.33% of individuals while histopathological analysis showed the presence of L. passim-like elements in the rectum of two examined specimens and the presence of fungal hyphae in the small intestine of another hornet. Biomolecular investigation revealed that *N. ceranae* was the most prevalent pathogen (50.0%), followed by *A. apis (*6.66%), *L. passim* (6.66%) and *C. bombi* (6.0%).

1. Introduction

Involuntary anthropic introduction, climate change (Ward and Masters, 2007; Renault et al., 2018) and habitat loss have led to the introduction and expansion of new and old species in new territories, leading to creation of new interactions among species which could change the epidemiology of pathogens (Crowl et al., 2008). This is particularly true for terrestrial arthropods, as it was established a close association between the increase of temperatures and the establishment of species outside their natural range (Hulme, 2017).

In the beekeeping field the introduction of alien species such as *Varroa destructor* (Warner et al., 2024), *Aethina tumida* (Schäfer et al., 2010) or *Vespa velutina* (Laurino et al., 2020; Diéguez-Antón et al., 2022) is a phenomenon which has proved to cause great economical damage to the beekeeping sector due to their impact on honey bee colonies.

The Oriental hornet (*Vespa orientalis* L.) is native to the southeastern Mediterranean, north-eastern and eastern Africa, the Middle East, Central Asia (Archer, 1998; Cetkovicí, 2003), Malta and southern Italy. However, it has expanded its areal to central and northern Italy (Bressi et al., 2019; Graziani and Cianferoni, 2021), as well as other European countries (Hernández et al., 2013; Sánchez et al., 2019; Castro and del Pico, 2021; Ceccolini, 2021; Gereys et al., 2021; Zachi and Ruicănescu, 2021) and South America (Ríos et al, 2020; du Buysson, 1904; Dvořák, 2006; Otis et al., 2023).

Social hornets of the Vespidae family appear to be particularly dangerous for the wellbeing of the honey bee (*Apis mellifera* L.) colonies due to their predatory and plundering behavior towards honey bees and their colonies (Matsuura, 1991; Matsuura and Yamane, 1990). Indeed, hornet foragers perform continuous patrolling flights in front of the hive to catch honey bees (Zucca et al., 2024; Werenkraut et al., 2022) or enter the hive to rob honey, pollen and honey bee larvae (Morse, 1978; Abrol, 1994) which are used as sources of food. Dietary habits of this species vary according to age. In fact, while adults feed mainly on carbohydrates (Ibrahim and Mazeed, 1967), such as rotten fruit and honey, brood is fed

* Corresponding author.
E-mail address: karen.power@unina.it (K. Power).
[1] These authors equally contributed.

https://doi.org/10.1016/j.jip.2024.108168
Received 4 May 2024; Received in revised form 29 June 2024; Accepted 11 July 2024
Available online 14 July 2024

mainly proteins, particularly insects (Archer, 1998; Hernández et al., 2013; Cini et al., 2018).

The interaction between hornets and honey bees could lead to the passage of pathogens between the two species, and hornets could act as vectors for honey bee pathogens (Mazzei et al., 2014; Marzoli et al., 2021).

Previous studies have reported the presence of various honey bee pathogens in the species of *Vespa* (Nowar, 2016; Gabín-García et al., 2021), with a specific focus on viruses (Forzan et al., 2017; Mazzei et al., 2018, 2019; Dalmon et al., 2019; Highfield et al., 2020; Rodríguez-Flores et al., 2022; Power et al., 2022, 2023), confirming their replicability in Vespids and underlining the possible role of these insects in spreading pathogens potentially lethal to honey bees (Yañez et al., 2012; Mazzei et al., 2018, 2019; Yang et al., 2019). Honey bee viruses are responsible for high mortality of honey bees and impairment of productions, which are also associated to fungi and protozoa (Lin et al., 2023; Neov et al., 2019).

Nosema sp. is a globally diffused microsporidia of honey bees (Grupe and Quandt, 2020). Microsporidia are obligate intracellular eukaryotic parasites, nowadays classified as fungi (Adl et al., 2005), able to cause substantial damage to honey bee colonies. Two different species have been characterized in Italy: *Nosema apis* and *Nosema ceranae*. *N. apis* is responsible for Nosemosis type A, which is associated to diarrhea, crawling bees, and winter losses (Fries, 1991), while *N. ceranae* is responsible for Nosemosis type C, which progresses with no evident clinical signs leading in the end to high colony mortality (Higes, et al., 2008a; Paxton, 2010; Botías et al., 2013). Nowadays, *N. ceranae* is the main microsporidia infecting bee colonies, having completely replaced *N. apis* in the last years, not only in Italy but also in other European and Mediterranean countries (Martín-Hernández et al., 2012). *Ascosphaera apis* (Spiltoir, 1955; Spiltoir and Olive, 1955) is also an important mycotic agent which affects the developing brood causing Chalkbrood disease (Flores et al., 2005a). The disease is characterized by death of honey bee larvae and their transformation into the so-called "chalkbrood mummies": dehydrated white, gray or black larvae of hard consistency (Aronstein and Murray, 2010). Transmission of *N. ceranae* and *A. apis* can occur via ingestion of spores which accumulate in the hive, beehive products, beekeeping material as well as on food sources like pollen and nectar (Flores et al., 2005b; Aronstein and Murray, 2010; Martín-Hernández et al., 2018; Pereira et al., 2019).

Honey bee trypanosomatids are the least-studied group of bee parasites, however the scientific interest towards these species has grown in the past years helping increasing data about their prevalence in honey bees (Runckel et al., 2011; Schwarz et al., 2015; Castelli et al., 2019; Williams et al., 2019; Hall et al., 2021; Michalczyk et al., 2022; Yamamoto et al., 2023; Bordin et al., 2022; Buendía-Abad et al., 2023; Iller et al., 2024). *Crithidia mellificae* and *Lotmaria passim,* which until 2014 was identified as *C. mellificae* (Cepero et al., 2014), have been associated to impairment of colony wellbeing as they colonize the gut lumen (Gómez-Moracho et al., 2020) and induce microbiome and immune modifications (Schwarz et al., 2016; Liu et al., 2020), also in association to *N. ceranae* (Schwarz and Evans, 2013). On the contrary, *C. bombi,* which affects bumblebees, seems to have no direct effect on honey bee colonies, although it appears that honey bees may play a role in the epidemiology of its transmission.

Given the lack of epidemiological data on non-viral pathogens, especially in Italy, this study aimed to verify the possible presence of the most common honey bee fungi (*N. ceranae* and A. *apis*) and trypanosomatids (*C. mellificae, C. bombi* and *L. passim*) in adult individuals of *V. orientalis*.

2. Materials and methods

Sixty adults *V. orientalis* were collected from seven different apiaries (A-G) located in the Sicily and Campania regions (South of Italy) (Table 1).

Table 1
Sampling site list.

Sampling site ID	Province	Coordinates	Samples ID
Pa1	Palermo	38.11142821090812, 13.351422249826987	A1-A6
Pa2	Palermo	38.098709696638224, 13.320493674095824	B1-B7
Av	Avellino	40.832859461289594, 14.845954603567128	C1-C7
Sa	Salerno	40.26745003430254, 14.939167255349021	D1-D7
Na	Naples	40.835056151950106, 14.109914052525768	E1-E9
Ca	Caserta	41.24987356538574, 14.066506340434836	F1-F9
Pa	Palermo	38.07854376431349, 13.334164047497351	G1-G15

Individuals were sampled during their predatory activity in the proximity of the hives using a sweep net and transported in 50 mL tubes to the laboratory of Veterinary General Pathology and Anatomical Pathology of the Department of Veterinary Medicine and Animal Productions, University of Naples "Federico II". Adults of *V. orientalis* were then processed as previously described (Power et al., 2022) and analyzed through cytological, histopathological and biomolecular techniques to verify the presence of honey bee pathogens, namely *N. ceranae, A. apis*, *L. passim, C. mellificae*, and *C. bombi*. Samples were individually homogenized in 1,5 mL of water and 50 µl of suspension was smeared on a slide and stained with MGG Quick Stain (Bio-Optica, Milan, Italy). Cytological slides were then observed by light microscopy. DNA was extracted from the remaining suspension using the DNA Blood and Tissue kit (Qiagen, Hilden, Germany) following the manufacturer instructions. The extracted DNAs were kept at −80 °C until the qPCR analysis.

A quantitative Real-Time PCR (qPCR) analysis was performed to determine the abundance of each pathogen in the samples using the extracted DNA. The primers used for the qPCRs are reported in Table 2.

A total reaction volume of 10 µL was produced for each target gene using SYBR™ green assays with forward and reverse primers and nucleic acid extract adding 2 µl of extracted DNA, using the SYBR PowerUp™ SYBR™ Green Master Mix (ThermoFisher, Waltham, MA, USA), as reported in previous studies (Cilia et al., 2021, 2022; Tiritelli et al., 2024). The qPCRs were carried out using a QuantStudio™ 3 Real-Time PCR System (ThermoFisher Scientific), according to the protocols for each gene sequence (James and Skinner, 2005; Huang et al., 2015; Xu et al., 2018; Cilia et al., 2018a; Bartolomé et al., 2018). DNA previously extracted from positive honey bees were employed as positive controls for each pathogen investigated, while sterile water was used as a negative control. All the analyses were carried out in duplicate.

A standard curve was created for each of the target genes by amplifying serially diluted recombinant plasmids containing the pathogen-specific DNA fragment from 1×10^1 to 1×10^9 copies in a qPCR assay on QuantStudio™ 3 Real-Time PCR System (ThermoFisher Scientific), as reported in previous studies using the amplification and quantification protocols (James and Skinner 2005; Huang et al., 2015; Xu et al., 2018; Cilia et al., 2018a; Bartolomé et al., 2018).

Moreover, 10 samples collected from the B site were formalin fixed and processed for histopathological analysis as previously described (Power et al., 2020). Slides were then stained with Hematoxylin and Eosin (HE) and Grocott staining to evidence possible fungi.

3. Results

Overall, 65.0 % of investigated *V. orientalis* were positive to at least one of the tested pathogens. All investigated individuals were negative

Table 2
List of primers used for pathogen detection in *Vespa orientalis*.

Target	Primer name	Sequence (5′-3′)	Reference
Ascosphaera apis	A_apis_3-F1	TGTCTGTGCGGCTAGGTG	(James and Skinner 2005)
	A_apis_3-R1	CCACTAGAAGTAAATGATGGTTAGA	
Nosema ceranae	Hsp70_F	GGGATTACAAGTGCTTAGAGTGATT	(Cilia et al., 2018a)
	Hsp70_R	TGTCAAGCCCATAAGCAAGTG	
Lotmaria passim	LpRPB_F	CCCATACCAGCGATCCTCA ATGAACTTCGCCACCTCATCA	(Bartolomé et al., 2018)
	LpRPB_R		
Crithidia mellificae	Cmel_Cyt_b_F	TAAATTCACTACCTCAAATTCAATAACATAATCAT	(Xu et al., 2018)
	Cmel_Cyt_b_R	ATTTATTGTTGTAATCGGTTTTATTGGATATGT	
Crithidia bombi	C.bombi_119Fw	CCAACGGTGAGCCGCATTCAGT	(Huang et al., 2015)
	C.bombi_119Rv	CGCGTGTCGCCCAGAACATTGA	

for *C. mellificae. N. ceranae* was the most prevalent pathogen (50.0 %), followed by *A. apis* and *L. passim* (6.66 %) and *C. bombi* (6.0 %). Besides, co-infections were identified in 3 specimens (5.0 %). The association between *N. ceranae* and *L. passim* was detected in 2 individuals (3.33 %), while the association between *N. ceranae* and *C. bombi* was revealed in only one hornet (1.66 %).

Concerning pathogen abundance, the higher load was detected for *N. ceranae* ($1.07 \times 10^6 \pm 5.31 \times 10^6$), followed by *L. passim* ($1.26 \times 10^4 \pm 9.18 \times 10^4$), *A. apis* ($6.54 \times 10^3 \pm 4.19 \times 10^4$) and *C. bombi* ($1.34 \times 10^2 \pm 8.12 \times 10^2$). For a comprehensive view of individual infection, data are available in Table S1.

The cytological examination revealed the presence of *Nosema* spores in 23 individuals (38.33 %) (Fig. 1A-B), while no sample was found positive for *A. apis, L. passim, C. bombi* and *C. mellificae*.

Histopathological analysis showed the presence of *L. passim*-like elements in the rectum of two examined hornets (20.0 %) (Fig. 2A-2B) associated to haemocyte recruitment (Fig. 2C), and Grocott staining emphasized the presence of fungal hyphae in the small intestine (Fig. 3A) of another hornet (Fig. 3B-C). No sample showed the presence of *Nosema* spp. and *Crithidia* spp.

4. Discussion and conclusions

Microsporidia and trypanosomatids are the most prevalent eukaryotic gut parasites of wild bees and honey bees and their role in honey bee colony impairment is well known (Higes et al., 2008a; Runckel et al., 2011; Cornman et al., 2012; Schwarz et al., 2015). Natural and laboratory infection with *N. ceranae, A. apis* and *L. passim* had already been reported in different Arthropoda such as *Andrena*, *Bombus*, *Melipona*, *Tetragonula*, *Osmia*, *Halictus* and *Lasioglossum* species (reviewed in Nanetti et al., 2021a). Moreover, *Aethina tumida* (the small hive beetle) was found to carry *N. ceranae, A. apis, L. passim, C. bombi* and *C. mellificae* in three different previous studies (Cilia et al., 2018b; de Landa et al., 2021; Nanetti et al., 2021b).

Considering the genus *Vespa*, identification of *Nosema* spp., *L. passim, C. mellificae* and *C. bombi*, was already reported in Spain in *V. crabro* and *V. velutina* (Gabín-García et al., 2021) but never in *V. orientalis*. Moreover, this is the first report of the presence of *A. apis* in *Vespa* genus.

A previous study carried out in northern Italy by Zucca et al. (2024)) analyzed V. *orientalis* samples for several bee pathogens using microscopic and molecular approaches. Hornets were positive for DWV, BQCV and SBV, while, conversely to the results of this investigation, they were negative for the *Nosema* spp, *L. passim* and *C. mellificae*.

The samples analyzed in our study presented higher prevalence of *N. ceranae* compared to the other gut parasites. These finding are dissimilar to those reported in *V. crabro* and *V. velutina*, in which *N. ceranae* was detected less frequently (3.2 % in *V. crabro* and 2.5 % in *V. velutina*) than both trypanosomatids (*L. passim* with a prevalence of 16.1 % in *V. crabro* and 3.8 % in *V. velutina*, while *C. bombi* 12.9 % in *V. crabro* and 17.7 % in *V. velutina*) (Gabín-García et al., 2021). Moreover, the same study described the presence of *C. mellificae* in both species of *Vespa* (3.2 % *V. crabro* and 8.9 % *V. velutina*) while it was not detected in any of the samples analyzed in our study.

Suspension smears revealed only the presence of *N. ceranae* and not of *A. apis*, *L. passim*, *C. mellificae*, and *C. bombi*. Indeed, cytological examination on its own is known to be less specific compared to molecular approaches (da Silva and Langoni, 2001; Meymandi et al., 2010; Otranto et al., 2011) and discrepancy in results has already been reported (Gabín-García et al., 2021).

In general, the average pathogen abundance detected in the investigated *V. orientalis* individuals was lower than the threshold ($>1 \times 10^6$ copies) generally considered for symptomatic infection load of pathogens infecting honey bee workers (Mazzei et al., 2014; Martín-Hernández et al., 2018; Cilia et al., 2020; Chen et al., 2021). Only the abundance of *N. ceranae* exceeded this limit in few individuals. Although abundance does not provide definitive epidemiological information especially related to non-*Apis* species, these findings indicated a widespread occurrence of *N. ceranae* among *V. orientalis* probably due to honey bee predation or to the great presence of *Nosema* spores in the environment (McMahon et al., 2015; Porrini et al., 2016; Radzevičiūtė et al., 2017; Alger et al., 2019; Tiritelli et al., 2024).

Transmission of honey bee gastrointestinal pathogens relies on oral-fecal route to infect new hosts, therefore it is likely to occur via ingestion of contaminated hive products or feed. Both microsporidia, fungi and

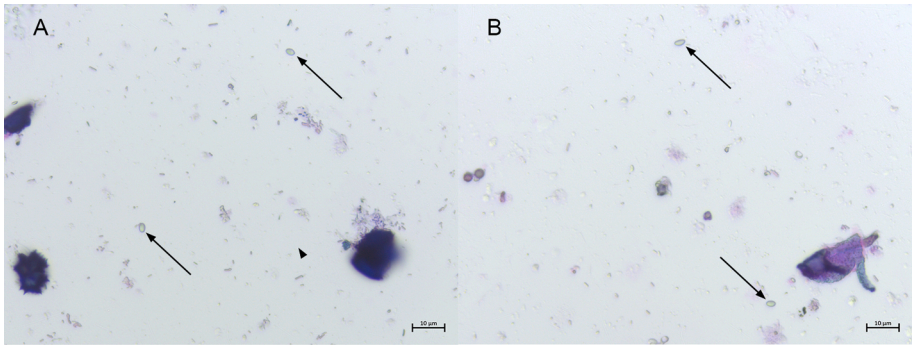

Fig. 1. Cytological examination of homogenized samples of *V. orientalis*. Presence of *Nosema* spp. spores (arrow). 40X. MGG Quik stain.

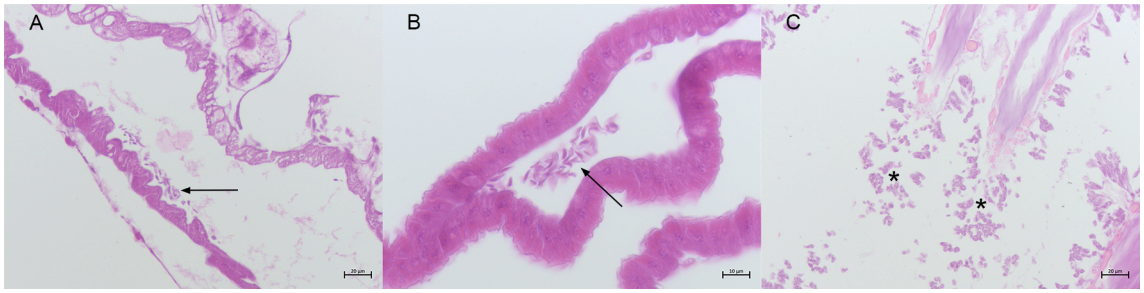

Fig. 2. Rectum of *V. orientalis*. A) Presence of *Lotmaria*-like forms adjacent to the epithelium. 20X. H&E. B) Presence of Lotmaria-like forms adjacent to the epithelium 40X. H&E. C) Presence of *Lotmaria*-like forms and haemocytes adjacent to the epithelium and in the lumen of the rectum. 20X. H&E.

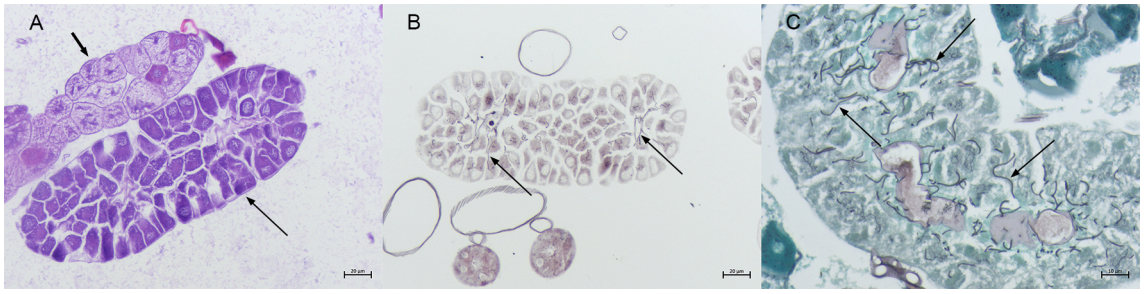

Fig. 3. Small intestine of *V. orientalis*. A) Healthy small intestine. 20X. H&E. B) Presence of fungal hyphae (arrow) 20X. Grocott stain. C) Presence of fungal hyphae (arrow) 40X. Grocott stain.

trypanosomatids, are highly resistant pathogens, and, parasite intake could have occurred during foraging activities, trophallaxis or vertically (Higes et al., 2008b; Graystock et al., 2015). However, we cannot discard the hypothesis of parasite uptake through ingestion of infected honey bees as it occurs for viruses (Yañez et al., 2020; Power et al., 2023), and more studies are needed to clarify this point.

In conclusion, *V. orientalis* in addition to honey bee viruses, can also harbor different species of enteropathogenic parasites and perhaps contribute to their spread.

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

CRediT authorship contribution statement

Karen Power: Writing – original draft, Methodology, Conceptualization. **Giovanni Cilia:** Writing – original draft, Methodology, Conceptualization. **Ernesto Ragusa:** Investigation. **Roberto Rizzo:** Validation. **Laura Bortolotti:** Writing – review & editing, Supervision. **Paola Maiolino:** Writing – review & editing, Supervision.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

The authors thank Dr. Claire Power for helping editing the figures in the article, and Dr. Carmine Vitagliano for assisting in laboratory practices.

Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jip.2024.108168.

References

Abrol, D.P., 1994. Ecology, behaviour and management of social wasp, *Vespa velutina* smith (Hym., vespidae), attacking honeybee colonies. Korean J. Apic. 9, 5–10. https://doi.org/10.1163/187631293X00226.

Adl, S.M., Simpson, A.G., Farmer, M.A., Andersen, R.A., Anderson, O.R., Barta, J.R., Bowser, S.S., Brugerolle, G., Fensome, R.A., Fredericq, S., James, T.Y., Karpov, S., Kugrens, P., Krug, J., Lane, C.E., Lewis, L.A., Lodge, J., Lynn, D.H., Mann, D.G., McCourt, R.M., Mendoza, L., Moestrup, O., Mozley-Standridge, S.E., Nerad, T.A., Shearer, C.A., Smirnov, A.V., Spiegel, F.W., Taylor, M.F., 2005. The new higher level classification of eukaryotes with emphasis on the taxonomy of protists. J. Eukaryot. Microbiol. 52 (5), 399–451. https://doi.org/10.1111/j.1550-7408.2005.00053.x.

Alger, S.A., Burnham, P.A., Boncristiani, H.F., Brody, A.K., 2019. RNA virus spillover from managed honeybees (Apis mellifera) to wild bumblebees (Bombus spp.). PLoS One 14 (6), e0217822.

Archer, M.E., 1998. Taxonomy, distribution and nesting biology of *Vespa Orientalis* L. Entomol. Mon. Mag. 134, 45–51.

Aronstein, K.A., Murray, K.D., 2010. Chalkbrood disease in honey bees. J. Invertebr. Pathol. 103 (Suppl 1), S20–S2099. https://doi.org/10.1016/j.jip.2009.06.018.

Bartolomé, C., Buendía, M., Benito, M., De la Rúa, P., Ornosa, C., Martín-Hernández, R., Higes, M., Maside, X., 2018. A new multiplex PCR protocol to detect mixed trypanosomatid infections in species of *Apis* and *Bombus*. J. Invertebr. Pathol. 154, 37–41. https://doi.org/10.1016/j.jip.2018.03.015.

Bordin, F., Zulian, L., Granato, A., Caldon, M., Colamonico, R., Toson, M., Trevisan, L., Biasion, L., Mutinelli, F., 2022. Presence of Known and Emerging Honey Bee Pathogens in Apiaries of Veneto Region (Northeast of Italy) during Spring 2020 and 2021. Appl. Sci. 12 https://doi.org/10.3390/app12042134.

Botías, C., Martín-Hernández, R., Barrios, L., Meana, A., Higes, M., 2013. *Nosema* spp. infection and its negative effects on honey bees (*Apis mellifera iberiensis*) at the colony level. Vet. Res. 44 https://doi.org/10.1186/1297-9716-44-25.

Bressi. N., Colla. A., Tomasin, G., 2019. Orientali Verso Nord: Insediamento Di Una Popolazione Urbana Di Calabrone Orientale (*Vespa Orientalis* Linnaeus 1771) a Trieste, NE Italy (Hymenoptera, Vespidae). Atti del Museo Civico di Storia Naturale di Trieste 60 (2019) 273–275.

Buendía-Abad, M., Martín-Hernández, R., Higes, M., 2023. Trypanosomatids in honey bee colonies in Spain: a new specific qPCR method for specific quantification of Lotmaria passim, Crithidia mellificae and Crithidia bombi. J. Invertebr. Pathol. 201, 108004 https://doi.org/10.1016/j.jip.2023.108004.

Castelli, L., Branchiccela, B., Invernizzi, C., Tomasco, I., Basualdo, M., Rodriguez, M., Zunino, P., Antúnez, K., 2019. Detection of Lotmaria passim in Africanized and European honey bees from Uruguay, Argentina and Chile. J. Invertebr. Pathol. 160, 95–97. https://doi.org/10.1016/j.jip.2018.11.004.

Castro, L., del Pico, C., 2021. Sobre el problema de *Vespa orientalis* Linnaeus1771 (Hymenoptera: vespidae) en el sur de españa. Rev. Gaditana Entomologia. 12 (1), 183–206.

Ceccolini, F., 2021. More records of *Vespa orientalis* linnaeus 1771 in the south of the Iberian peninsula (Hymenoptera: vespidae: vespinae). Arquivos Entomolóxicos. 24, 335–338.

Cepero, A., Ravoet, J., Gómez-Moracho, T., Bernal, J.L., Del Nozal, M.J., Bartolomé, C., Maside, X., Meana, A., González-Porto, A.V., de Graaf, D.C., Martín-Hernández, R., Higes, M., 2014. Holistic screening of collapsing honey bee colonies in Spain: a case study. BMC. Res. Notes 7, 649. https://doi.org/10.1186/1756-0500-7-649.

Cetkovicí A. A review of the European distribution of the oriental hornet (Hymenoptera, Vespidae: *Vespa orientalis* l.). Ekologija. 2003. 37 (1–2), 1–22.

Chen, G., Wu, Y., Deng, J., Wen, Z., Wang, S., Chen, Y., Hu, F., Zheng, H., 2021. Seasonal variation of viral infections between the eastern honey bee (Apis cerana) and the western honey bee (Apis mellifera). Microbiologyopen. 10 (1), e1162.

Cilia, G., Garrido, C., Bonetto, M., Tesoriero, D., Nanetti, A., 2020. Effect of Api-Bioxal® and ApiHerb® Treatments against Nosema ceranae Infection in Apis mellifera Investigated by Two qPCR Methods. Vet Sci. 7 (3), 125. https://doi.org/10.3390/vetsci7030125.

Cilia, G., Zavatta, L., Ranalli, R., Nanetti, A., Bortolotti, L., 2021. Replicative deformed wing virus found in the head of adults from symptomatic commercial bumblebee (*Bombus terrestris*) colonies. Vet Sci. 8 (7), 117. https://doi.org/10.3390/vetsci8070117.

Cilia, G., Cabbri, R., Maiorana, G., Cardaio, I., Dall'Olio, R., Nanetti, A., 2018a. A novel TaqMan® assay for *Nosema ceranae* quantification in honey bee, based on the protein coding gene Hsp70. Eur. J. Protistol. 63, 44–50. https://doi.org/10.1016/j.ejop.2018.01.007.

Cilia, G., Cardaio, I., dos Santos, P.E.J., Ellis, J.D., Nanetti, A., 2018b. The first detection of *Nosema ceranae* (Microsporidia) in the small hive beetle, *Aethina tumida* Murray (Coleoptera: Nitidulidae). Apidologie 49, 619–624. https://doi.org/10.1007/s13592-018-0589-8.

Cilia, G., Flaminio, S., Zavatta, L., Ranalli, R., Quaranta, M., Bortolotti, L., Nanetti, A., 2022. Occurrence of honey bee (*Apis mellifera* L.) pathogens in wild pollinators in Northern Italy. Front. Cell. Infect. Microbiol. 12, 907489 https://doi.org/10.3389/fcimb.2022.907489.

Cini, A., Cappa, F., Petrocelli, I., Pepiciello, I., Bortolotti, L., Cervo, R., 2018. Competition between the native and the introduced hornets vespa crabro and vespa velutina: a comparison of potentially relevant life-history traits. Ecol. Entomol. 43, 351–362. https://doi.org/10.1111/een.12507.

Cornman, R.S., Tarpy, D.R., Chen, Y., Jeffreys, L., Lopez, D., van Pettis, J.S., Engelsdorp, D., Evans, J.D., 2012. Pathogen webs in collapsing honey bee colonies. PLoS One 7, e43562.

Crowl, T.A., Crist, T.O., Parmenter, R.R., Belovsky, G., Lugo, A.E., 2008. The spread of invasive species and infectious disease as drivers of ecosystem change. Front. Ecol. Environ. 6, 238–246.

da Silva, A.V., Langoni, H., 2001. The detection of *Toxoplasma gondii* by comparing cytology, histopathology, bioassay in mice, and the polymerase chain reaction (PCR). Vet. Parasitol. 97 (3), 191–198. https://doi.org/10.1016/s0304-4017(01)00404-6.

Dalmon, A., Gayral, P., Decante, D., Klopp, C., Bigot, D., Thomasson, M., Herniou, E.A., Alaux, C., Le Conte, Y., 2019. Viruses in the invasive hornet *Vespa velutina*. Viruses 11 (11), 1041. https://doi.org/10.3390/v11111041.

de Landa, G.F., Porrini, M.P., Revainera, P., Porrini, D.P., Farina, J., Correa-Benítez, A., Maggi, M.D., Eguaras, M.J., Quintana, S., 2021. Pathogens detection in the small hive beetle (*Aethina tumida* (Coleoptera: Nitidulidae)). Neotrop. Entomol. 50 (2), 312–316. https://doi.org/10.1007/s13744-020-00812-8.

Diéguez-Antón, A., Rodríguez-Flores, M.S., Escuredo, O., Seijo, M.C., 2022. Monitoring Study in Honeybee Colonies Stressed by the Invasive Hornet *Vespa velutina*. Vet Sci. 9 (4), 183. https://doi.org/10.3390/vetsci9040183.

du Buysson, R., 1904. Monographie Des Guèpes Ou Vespa. Ann. Soc Entomol. Fr. 73, 485–556.

Dvořák, L., 2006. Oriental Hornet *Vespa Orientalis* Linnaeus 1771 Found in Mexico (Hymenoptera, Vespidae, Vespinae). Entomol. Probl. 36, 80.

Flores, J.M., Spivak, M., Gutiérrez, I., 2005a. Spores of *Ascosphaera apis* contained in wax foundation can infect honeybee brood. Vet. Microbiol. 108 (1–2), 141–144. https://doi.org/10.1016/j.vetmic.2005.03.005.

Flores, J.M., Gutierrez, I., Espejo, R., 2005b. The role of pollen in chalk brood disease in *Apis mellifera*: transmission and predisposing conditions. Mycologia 97, 1171–1176.

Forzan, M., Felicioli, A., Sagona, S., Bandecchi, P., Mazzei, M., 2017. Complete genome sequence of deformed wing virus isolated from *Vespa crabro* in Italy. Genome Announc. 5 (40), e00961–e01017. https://doi.org/10.1128/genomeA.00961-17.

Fries, I., 1991. *Nosema apis* – a parasite in the honey bee colony. Bee World 74, 5–19.

Gabín-García, L.B., Bartolomé, C., Guerra-Tort, C., Rojas-Nossa, S.V., Llovo, J., Maside, X., 2021. Identification of pathogens in the invasive hornet *Vespa velutina* and in native Hymenoptera (Apidae, Vespidae) from SW-Europe. Sci. Rep. 11 (1), 11233.

Gereys, B., Coache, A., Filippi, G., 2021. Preísence en France meítropolitaine d'un frelon allochtone: vespa orientalis linnaeus 1771 (Le frelon oriental) (Hymenoptera, vespidae, vespinae). Faunitaxys 9 (32), 1–5.

Gómez-Moracho, T., Buendía-Abad, M., Benito, M., García-Palencia, P., Barrios, L., Bartolomé, C., Maside, X., Meana, A., Jiménez-Antón, M.D., Olías-Molero, A.I., Alunda, J.M., Martín-Hernández, R., Higes, M., 2020. Experimental evidence of harmful effects of *Crithidia mellificae* and *Lotmaria passim* on honey bees. Int. J. Parasitol. 50 (13), 1117–1124. https://doi.org/10.1016/j.ijpara.2020.06.009.

Graystock, P., Goulson, D., Hughes, W.O., 1813. Parasites in bloom: flowers aid dispersal and transmission of pollinator parasites within and between bee species. Proc. Biol. Sci. 2015 (282), 20151371. https://doi.org/10.1098/rspb.2015.1371.

Graziani, F., Cianferoni, F., 2021. The Northernmost Record of *Vespa orientalis* Linnaeu (Hymenoptera: Vespidae) in Peninsular Italy. Revta. Gad. Entom. 12, 173–178.

Grupe, A.C., Quandt, C.A., 2020. A growing pandemic: a review of *Nosema Parasites* in globally distributed domesticated and native bees. PLoS Pathog. 16 (6), e1008580.

Hall, R.J., Pragert, H., Phiri, B.J., Fan, Q.H., Li, X., Parnell, A., Stanislawek, W.L., McDonald, C.M., Ha, H.J., McDonald, W., Taylor, M., 2021. Apicultural practice and disease prevalence in Apis mellifera, New Zealand: a longitudinal study. J Apic Res. 2021. 60(5):644–658. doi; 10.1080/00218839.2021.1936422.

Higes, M., Martín-Hernández, R., Botías, C., Bailón, E.G., González-Porto, A.V., Barrios, L., Del Nozal, M.J., Bernal, J.L., Jiménez, J.J., Palencia, P.G., Meana, A., 2008a. How natural infection by *Nosema ceranae* causes honeybee colony collapse. Environ. Microbiol. 10 (10), 2659–2669. https://doi.org/10.1111/j.1462-2920.2008.01687.x.

Higes, M., Martín-Hernández, R., Garrido-Bailón, E., García-Palencia, P., Meana, A., 2008b. Detection of infective *Nosema ceranae* (Microsporidia) spores in corbicular pollen of forager honeybees. J. Invertebr. Pathol. 97 (1), 76–78. https://doi.org/10.1016/j.jip.2007.06.002.

Highfield, A., Kevill, J., Mordecai, G., Hunt, J., Henderson, S., Sauvard, D., Feltwell, J., Martin, S.J., Sumner, S., Schroeder, D.C., 2020. Detection and replication of moku virus in honey bees and social wasps. Viruses 12 (6), 607. https://doi.org/10.3390/v12060607.

Hernández, R., García-Gans, F.J., Selfa, J., Rueda, J., 2013. Primera Cita De La Avispa Oriental Invasora *Vespa orientalis* Linnaeus 1771 (Hymenoptera: Vespidae) En La Península Ibérica. Boletín la Sociedad Entomológica Aragonesa. 2013. 52: 299–300.

Huang, W.F., Skyrm, K., Ruiter, R., Solter, L., 2015. Disease management in commercial bumble bee mass rearing, using production methods, multiplex PCR detection techniques, and regulatory assessment. J. Apic. Res. 2015. 54:516–524. doi; 10.1080/00218839.2016.1173352.

Hulme, P.E., 2017. Climate change and biological invasions: evidence, expectations, and response options. Biol. Rev. Camb. Philos. Soc. 92 (3), 1297–1313. https://doi.org/10.1111/brv.12282.

Ibrahim, M.M., Mazeed, M.M., 1967. Studies on the oriental hornet, *Vespa orientalis*. Agric. Res. Rev. 45, 163–180.

Iller, M., Lipczyńska-Ilczuk, K., Sokół, R., Borsuk, G., Bancerz-Kisiel, A., 2024. Phylogenetic analysis of the trypanosomatid parasite Lotmaria passim in honey bees (Apis mellifera) in Poland. J. Vet. Res. 68 (1), 123–127. https://doi.org/10.2478/jvetres-2024-0018.

James, R.R., Skinner, J.S., 2005. PCR diagnostic methods for *Ascosphaera* infections in bees. J. Invertebr. Pathol. 90 (2), 98–103. https://doi.org/10.1016/j.jip.2005.08.004.

Laurino, D., Lioy, S., Carisio, L., Manino, A., Porporato, M., 2020. *Vespa velutina*: an alien driver of honey bee colony losses. Diversity 12, 5. https://doi.org/10.3390/d12010005.

Lin, Z., Zheng, M., Li, Z., Ji, T., 2023. Editorial: biotic and abiotic stresses on honeybee physiology and colony health. Front. Physiol. 14, 1260547. https://doi.org/10.3389/fphys.2023.1260547.

Liu, Q., Lei, J., Darby, A.C., Kadowaki, T., 2020. Trypanosomatid parasite dynamically changes the transcriptome during infection and modifies honey bee physiology. Commun Biol. 3 (1), 51. https://doi.org/10.1038/s42003-020-0775-x.

Martín-Hernández, R., Bartolomé, C., Chejanovsky, N., Le Conte, Y., Dalmon, A., Dussaubat, C., García-Palencia, P., Meana, A., Pinto, M.A., Soroker, V., Higes, M., 2018. *Nosema ceranae* in *Apis mellifera*: a 12 years postdetection perspective. Environ. Microbiol. 20 (4), 1302–1329. https://doi.org/10.1111/1462-2920.14103.

Martín-Hernández, R., Botías, C., Bailón, E.G., Martínez-Salvador, A., Prieto, L., Meana, A., Higes, M., 2012. Microsporidia infecting Apis mellifera: coexistence or competition. Is *Nosema ceranae* replacing *Nosema apis*? Environ Microbiol. 2012. 14 (8):2127-38. doi: 10.1111/j.1462-2920.2011.02645.x.

Marzoli, F., Forzan, M., Bortolotti, L., Pacini, M.I., Rodríguez-Flores, M.S., Felicioli, A., Mazzei, M., 2021. Next generation sequencing study on RNA viruses of *Vespa velutina* and *Apis mellifera* sharing the same foraging area. Transbound. Emerg. Dis. 68, 2261–2273.

Matsuura, M., Yamane, S., 1990. Biology of Vespine Wasps. Berlin, Germany: Springer, 1990.

Matsuura M., 1991. "Vespa and Provespa"," in The Social Biology of Wasps. New York, NY, USA: Cornell University Press. 1991. 232–262.

Mazzei, M., Carrozza, M.L., Luisi, E., Forzan, M., Giusti, M., Sagona, S., Tolari, F., Felicioli, A., 2014. Infectivity of DWV associated to flower pollen: Experimental evidence of a horizontal transmission route. PLoS One 9, e113448.

Mazzei, M., Forzan, M., Cilia, G., Sagona, S., Bortolotti, L., Felicioli, A., 2018. First detection of replicative deformed wing virus (DWV)in *Vespa velutina nigrithorax*. Bull Insectol. 71, 211–216.

Mazzei, M., Cilia, G., Forzan, M., Lavazza, A., Mutinelli, F., Felicioli, A., 2019. Detection of replicative Kashmir Bee Virus and Black Queen Cell Virus in Asian hornet *Vespa velutina* (Lepelieter 1836) in Italy. Sci. Rep. 9 (1), 10091. https://doi.org/10.1038/s41598-019-46565-2.

McMahon, D.P., Fürst, M.A., Caspar, J., Theodorou, P., Brown, M.J.F., Paxton, R.J., 2015. A sting in the spit: widespread cross-infection of multiple RNA viruses across wild and managed bees. J. Anim. Ecol. 84 (3), 615–624. https://doi.org/10.1111/1365-2656.12345.

Meymandi, S.S., Bahmanyar, M., Dabiri, S., Aflatonian, M.R., Bahmanyar, S., Meymandi, M.S., 2010. Comparison of cytologic giemsa and real-time polymerase chain reaction technique for the diagnosis of cutaneous leishmaniasis on scraping smears. Acta Cytol. 54 (4), 539–545. https://doi.org/10.1159/000325174.

Michalczyk, M., Sokół, M., Bancerz-Kisiel, A., 2022. Presence of *Lotmaria passim*, *Crithidia mellificae* and *Nosema* spp. in differently aged *Apis mellifera* brood. J. Apic. Res. https://doi.org/10.1080/00218839.2022.2055265.

Morse, R.A., 1978. Honey bee pests, predators, and diseases. 1978. Ithaca, NY, USA: Cornell University Press.

Nanetti, A., Bortolotti, L., Cilia, G., 2021a. Pathogens Spillover from Honey Bees to Other Arthropods. Pathogens. 10 (8), 1044. https://doi.org/10.3390/pathogens10081044.

Nanetti, A., Ellis, J.D., Cardaio, I., Cilia, G., 2021b. Detection of *Lotmaria passim*, *Crithidia mellificae* and replicative forms of deformed wing virus and Kashmir Bee Virus in the Small Hive Beetle (*Aethina tumida*). Pathogens. 10 (3), 372. https://doi.org/10.3390/pathogens10030372.

Neov, B., Georgieva, A., Shumkova, R., Radoslavov, G., Hristov, P., 2019. Biotic and abiotic factors associated with colonies mortalities of managed honey bee (*Apis mellifera*). Diversity 11. https://doi.org/10.3390/d11120237.

Nowar, E.E., 2016. Oriental hornet (*Vespa orientalis*) as AFB disease vector to honeybee (*Apis mellifera* l.). Colonies. mid. East. J. App. Scien. 06 (04), 934–940.

Otis, G.W., Taylor, B.A., Mattila, H.R., 2023. Invasion potential of hornets (Hymenoptera: Vespidae: *Vespa* spp.). Front. Insect Sci. 3 (1145158) https://doi.org/10.3389/finsc.2023.1145158.

Otranto, D., Dantas-Torres, F., Weigl, S., Latrofa, M.S., Stanneck, D., Decaprariis, D., Capelli, G., Baneth, G., 2011. Diagnosis of *Hepatozoon canis* in young dogs by cytology and PCR. Parasit. Vectors 4, 55. https://doi.org/10.1186/1756-3305-4-55.

Paxton, R.J., 2010. Does infection by Nosema ceranae cause "Colony Collapse Disorder" in honey bees (*Apis mellifera*)? J. Apic. Res. 49, 80–84. https://doi.org/10.3896/IBRA.1.49.1.1.

Pereira, K.S., Meeus, I., Smagghe, G., 2019. Honey bee-collected pollen is a potential source of *Ascosphaera apis* infection in managed bumble bees. Sci. Rep. 9 (1), 4241. https://doi.org/10.1038/s41598-019-40804-2.

Porrini, C., Mutinelli, F., Bortolotti, L., Granato, A., Laurenson, L., Roberts, K., Gallina, A., Silvester, N., Medrzycki, P., Renzi, T., Sgolastra, F., Lodesani, M., 2016. The Status of Honey Bee Health in Italy: results from the Nationwide Bee Monitoring Network. PLoS One 11 (5), e0155411.

Power, K., Martano, M., Altamura, G., Maiolino, P., 2020. Histopathological findings in testes from apparently healthy drones of *Apis mellifera ligustica*. Vet Sci. 7 (3), 124. https://doi.org/10.3390/vetsci7030124.

Power, K., Altamura, G., Martano, M., Maiolino, P., 2022. Detection of honeybee viruses in *Vespa orientalis*. Front. Cell. Infect. Microbiol. 12, 896932 https://doi.org/10.3389/fcimb.2022.896932.

Power, K., Martano, M., Ragusa, E., Altamura, G., Maiolino, P., 2023. Detection of honey bee viruses in larvae of *Vespa orientalis*. Front. Cell. Infect. Microbiol. 13, 1207319. https://doi.org/10.3389/fcimb.2023.1207319.

Radzevičiūtė, R., Theodorou, P., Husemann, M., Japoshvili, G., Kirkitadze, G., Zhusupbaeva, A., Paxton, R.J., 2017. Replication of honey bee-associated RNA viruses across multiple bee species in apple orchards of Georgia. Germany and Kyrgyzstan. J Invertebr Pathol. 146, 14–23. https://doi.org/10.1016/j.jip.2017.04.002.

Renault, D., Laparie, M., McCauley, S.J., Bonte, D., 2018. Environmental adaptations, ecological filtering, and dispersal central to insect invasions. Annu. Rev. Entomol. 63, 345–368. https://doi.org/10.1146/annurev-ento-020117-043315.

Ríos, M., Barrera-Medina, R., Contreras, J.M., 2020. Primer Reporte Del Género *Vespa* Linnaeus (Hymenoptera: Vespidae: Vespinae) En Chile. Rev. Chil. Entomol. 2020. 46, 237–242. doi: 10.35249/rche.46.2.20.14.

Rodríguez-Flores, M.S., Mazzei, M., Felicioli, A., Diéguez-Antón, A., Seijo, M.C., 2022. Emerging risk of cross-species transmission of honey bee viruses in the presence of invasive vespid species. Insects. 14 (1), 6. https://doi.org/10.3390/insects14010006.

Runckel, C., Flenniken, M.L., Engel, J.C., Ruby, J.G., Ganem, D., Andino, R., DeRisi, J.L., 2011. Temporal analysis of the honey bee microbiome reveals four novel viruses and seasonal prevalence of known viruses, *Nosema*, and *Crithidia*. PLoS One 6, e20656.

Sánchez, I., Fajardo, M.C., Castro, M., 2019. Primeras citas del avispón oriental vespa orientalis Linnaeus 1771 (Hymenoptera: vespidae) para andalucía (España). Rev. la Sociedad Gaditana Hist. Natural 13, 11–14.

Schäfer, M.O., Ritter, W., Pettis, J.S., Neumann, P., 2010. Winter losses of honeybee colonies (Hymenoptera: Apidae): the role of infestations with *Aethina tumida* (Coleoptera: Nitidulidae) and *Varroa destructor* (Parasitiformes: Varroidae). J. Econ. Entomol. 103 (1), 10–16. https://doi.org/10.1603/ec09233.

Schwarz, R.S., Evans, J.D., 2013. Single and mixed-species trypanosome and microsporidia infections elicit distinct, ephemeral cellular and humoral immune responses in honey bees. Dev. Comp. Immunol. 40 (3–4), 300–310. https://doi.org/10.1016/j.dci.2013.03.010.

Schwarz, R.S., Bauchan, G.R., Murphy, C.A., Ravoet, J., de Graaf, D.C., Evans, J.D., 2015. Characterization of Two Species of Trypanosomatidae from the Honey Bee *Apis mellifera*: *Crithidia mellificae* Langridge and McGhee, and *Lotmaria passim* n. gen., n. sp. J. Eukaryot. Microbiol. 62 (5), 567–583. https://doi.org/10.1111/jeu.12209.

Schwarz, R.S., Moran, N.A., Evans, J.D., 2016. Early gut colonizers shape parasite susceptibility and microbiota composition in honey bee workers. PNAS 113 (33), 9345–9350. https://doi.org/10.1073/pnas.1606631113.

Spiltoir, C.F., 1955. Life cycle of *Ascosphaera apis*. Am. J. Bot. 42, 501–518.

Spiltoir, C.F., Olive, L.S., 1955. A reclassification of the genus *Pericystis* betts. Mycologia 47, 238–244.

Tiritelli, R., Flaminio, S., Zavatta, L., Ranalli, R., Giovanetti, M., Grasso, D.A., Leonardi, S., Bonforte, M., Boni, C.B., Cargnus, E., Catania, R., Coppola, F., Di Santo, M., Pusceddu, M., Quaranta, M., Bortolotti, L., Nanetti, A., Cilia, G., 2024. Ecological and social factors influence interspecific pathogens occurrence among bees. Sci. Rep. 14 (1), 5136. https://doi.org/10.1038/s41598-024-55718-x.

Ward, N.L., Masters, G.J., 2007. Linking climate change and species invasion: an illustration using insect herbivores. Glob. Change Biol. 2007. 13 (8), 1605–1615. doi: 10.1111/j.1365-2486.2007.01399.x.

Warner, S., Pokhrel, L.R., Akula, S.M., Ubah, C.S., Richards, S.L., Jensen, H., Kearney, G. D., 2024. A scoping review on the effects of Varroa mite (*Varroa destructor*) on global honey bee decline. Sci. Total Environ. 906, 167492 https://doi.org/10.1016/j.scitotenv.2023.167492.

Werenkraut, V., Arbetman, M.P., Fergnani, P.N., 2022. The Oriental Hornet (*Vespa orientalis* L.): a Threat to the Americas? Neotrop. Entomol. 51 (2), 330–338. https://doi.org/10.1007/s13744-021-00929-4.

Williams MKF, Tripodi AD, Szalanski AL. Molecular survey for the honey bee (Apis mellifera L.) trypanosome parasites Crithidia mellificae and Lotmaria passim. J Apic Res. 2019 58(4), 553–558. doi; 10.1080/00218839.2019.1568956.

Xu, G., Palmer-Young, E., Skyrm, K., Daly, T., Sylvia, M., Averill, A., Rich, S., 2018. Triplex real-time PCR for detection of *Crithidia mellificae* and *Lotmaria passim* in honey bees. Parasitol. Res. 117 (2), 623–628. https://doi.org/10.1007/s00436-017-5733-2.

Yamamoto, S., Nakamura, S., Nakayama, K., Kusakisako, K., Watanabe, K., Ikadai, H., Tanabe, T., 2023. Molecular detection of Lotmaria passim in honeybees in Japan. Parasitol. Int. 93, 102711 https://doi.org/10.1016/j.parint.2022.102711.

Yañez, O., Zheng, H.Q., Hu, F.L., Neumann, P., Dietemann, V., 2012. A scientific note on Israeli acute paralysis virus infection of Easternhoneybee *Apis cerana* and vespine predator *Vespa velutina*. Apidologie 43, 587–589.

Yañez, O., Piot, N., Dalmon, A., de Miranda, J.R., Chantawannakul, P., Panziera, D., Amiri, E., Smagghe, G., Schroeder, D., Chejanovsky, N., 2020. Bee Viruses: Routes of Infection in Hymenoptera. Front. Microbiol. 11, 943. https://doi.org/10.3389/fmicb.2020.00943.

Yang, D., Zhao, H., Shi, J., Xu, X., Wu, Y., Guo, R., Chen, D., Wang, X., Deng, S., Yang, S., Diao, Q., Hou, C., 2019. Discovery of aphid lethal paralysis virus in *Vespa velutina* and *Apis cerana* in China. Insects. 10 (6), 157. https://doi.org/10.3390/insects10060157.

Zachi, M., Ruicănescu, A., 2021. Vespa orientalis, a new alien species in romania. Travaux du Muséum National d'Histoire Naturelle "Grigore Antipa". 2021. 64 (1) 67– 72. doi: 10.3897/travaux.64.e61954.

Zucca, P., Granato, A., Mutinelli, F., Schiavon, E., Bordin, F., Dimech, M., Balbo, R.A., Mifsud, D., Dondi, M., Cipolat-Gotet, C., Rossmann, M.C., Ocepek, M.P., Scaravelli, D., Palei, M., Zinzula, L., Spanjol, K., 2024. The oriental hornet (*Vespa orientalis*) as a potential vector of honey bee's pathogens and a threat for public health in North-East Italy. Vet Med Sci. 10 (1), e1310.