Ticks are among the most relevant arthropod vectors affecting human and domestic animals health, because they have capacity to transmit more disease agents to animals and humans than any other arthropod. Furthermore, ticks are able to cause toxicosis, irritation, allergy, and leave painful and lasting lesions in the skin which can lead to secondary infections.

Awareness on species of ticks biting humans may be useful for preventing the transmission of tick-borne diseases. In Argentina, there are several records of *Amblyomma* ticks parasitizing humans. However, in Misiones Province, northeast of the country, these records are scarce. In this area many people are exposed to tick bites due to the wide spread of rural activities and wildlife eco-tourism. The aim of this paper is to present information on ixodid species that can bite humans in northern Misiones Province, a region where knowledge about these parasites and their effect on human health is limited, with notes on their role in the potential transmission of rickettsial organisms.

**Materials and methods**

The sampled area is located at north of the province of Misiones and includes forest environments inside the Iguazú National Park (INP) and Puerto Península Provincial Park (PPPP), and farms located at northwest of the province: Werle (25° 34' 27"S, 54° 09' 22"W), Los Cedritos (25° 42' 49"S, 54° 02' 49"W) and Jacobo (25° 43' 58"S, 53° 57' 19"W). Within the INP, three sites were selected: 1) Macuco trail (25° 40' 38"S, 54° 26' 55"W), a tourist attraction of the Park, where people pass every day, 2) Former camping site Ñandú (25° 34' 27"S, 54° 09' 22"W), Los Cedritos (25° 42' 49"S, 54° 02' 49"W) and Jacobo (25° 43' 58"S, 53° 57' 19"W). Within the INP, three sites were selected: 1) Macuco trail (25° 40' 38"S, 54° 26' 55"W), a tourist attraction of the Park, where people pass every day, 2) Former camping site Ñandú (25° 42' 12"S, 54° 25' 32"W), where tourists are not allowed, but its re-opening is projected in the near future, 3) Apepú Ranger Station (25° 33' 54"S, 54° 17' 45"W), intangible area. In the PPPP only one site was selected (25° 43' 36"S, 54° 33' 03"W). Ticks were collected during successive sampling sessions at the forest and farms sites from April 2014 until July 2015. At each site of the INP and the PPPP, three animal trails were sampled by dragging white cloth flags of 1x1.5 m size for an hour, totaling three hours of sampling per session, in search...
of free-living ticks. In the farms, cattle and horses were revised looking for ticks. After each sampling session, the researchers returned to the laboratory where their bodies were examined and feeding ticks were recovered using dissection forceps.

All researchers are involved in the project “Eco-epidemiological study of ticks (Acari: Ixodidae) in northern Misiones, Argentina”, and were aware of the risk they were subjected due to walk through active animal trails. During the weeks after sampling, the wounds caused by ticks were controlled, and researchers made sure that no symptoms associated to tick-borne diseases appear.

Ticks were preserved in 1.5 ml tubes with 96° ethanol for subsequent determination by morphological characters following Guglielmone and Viñábal, Barros-Battesti et al. and Martins et al. and by comparison with specimens deposited in the tick collection of INTA Rafaela.

Results

A total of 282 ticks attached to the body of researchers were collected after 32 sampling sessions, the details on species and locations are shown in Table I. Of these ticks, 280 attached to the host while working in active wildlife trails in the INP and the PPPP. Also, one feeding tick was collected after visiting a farm at Gral. Manuel Belgrano Department, northeast Misiones province, and one at Puerto Iguazu city, northwest of the province.

From the *Amblyomma* ticks, 144 were larvae and 134 were nymphs. All larvae were determined to genus level. Regarding nymphs, 47 were *Amblyomma brasiliense* Araújo, 1908, 76 *Amblyomma coelesbs* Neumann, 1899, 10 *Amblyomma incisum* Neumann, 1906 and one *Amblyomma dubitatum* Neumann, 1899. One nympha of *Haemaphysalis juxtakochi* Cooley, 1946 and a male of *Amblyomma ovale* Koch, 1844 were also collected. After one of the visits to the farms, a female of *Rhipicephalus microplus* (Canestrini, 1888), was found parasitizing a researcher, and in the city of Puerto Iguazú, a male of *Rhipicephalus sanguineus* sensu lato, was found attached to a team member.

*Amblyomma* ticks were found mostly attached to the skin of limbs and trunk (n = 275), while *R. microplus* and *R. sanguineus* were found on the skin of the neck and head of the researchers. The specimens remained attached from minutes to days if they were unseen. The larvae and nymphs bites occurred mostly unnoticed, unlike the adult *A. ovale* bite, which caused a significant acute pain.

The main injuries caused by the ticks were inflammation with serous exudation and pustules accompanied by pruritus. They had a very variable duration, from one week to five months.

Discussion

The presence of *A. coelesbs* in Argentina was only confirmed for northern Misiones. The high number of *A. coelesbs* nymphs parasitizing humans during almost every month of the year (see Table 1) indicates that it is a very aggressive species to humans. *Amblyomma coelesbs* has been found infected with *Rickettsia amblyommii*, a rickettsia currently considered of unknown pathogenicity, but that could be responsible for causing disease in humans. Specimens of *A. brasiiliense* have been previously found infesting humans at northwest of the country. To date there are no records of infection with tick-borne human pathogens in this species. In the case of *A. incisum*, the finding of this work constitutes the first records of this tick biting humans in Argentina. *Amblyomma incisum* was found infected with *Rickettsia bellii* and *Rickettsia monteiroi* in southeastern Brazil, none involved as human pathogens. Further studies should be focused on the pathogenic agents potentially transmitted by these three tick species in northeastern Argentina.

*Amblyomma dubitatum* is reported for the first time in the INP, and this record is the first infesting humans in Misiones province. Lado et al. have detected the human pathogen *Rickettsia parkeri*, a spotted fever group (SFG) rickettsia, in *A. dubitatum* of Uruguay, and Monje et al. found in Argentina (Santa Fe Province) a specimen of *A. dubitatum* infected with *Rickettsia* sp. strain Atlantic rainforest, also a SFG rickettsia pathogenic to humans. These antecedents evidence the sanitary relevance of the findings of *A. dubitatum* parasitizing humans in Misiones Province.

The other *Amblyomma* species found on humans in this work was *A. ovale*. This species has been involved in the epidemiology of the human pathogen *Rickettsia* sp. strain Atlantic rainforest in an area of southeastern Brazil ecologically similar to the north of Misiones Province. This fact suggests that the epidemiological dynamics of *Rickettsia* sp. strain Atlantic rainforest in southeastern Brazil could be extrapolated to the sampled area during the current work.

The worldwide distributed taxon *Rhipicephalus sanguineus* (Latreille, 1806) is currently considered a species complex. In South America, the taxon *R. sanguineus* s.l is composed by at least two lineages: tropical and temperate. In Brazil, *R. sanguineus* s.l specimens belonging to the tropical lineage were found infected with *Rickettsia rickettsii*, the most pathogenic *Rickettsia* species, while in Argentina *R. sanguineus* s.l specimens belonging to the temperate lineage were found to be infected with other human pathogen, *Rickettsia massiliiae*. Because in this work *R. sanguineus* s.l was found biting humans, it is important to determine the taxonomic status and the role as vector of human pathogens of the *R. sanguineus* s.l populations inhabiting urban and rural areas of the north of Misiones Province.

*Haemaphysalis juxtakochi* and *R. microplus* have not been previously registered infesting humans in Misiones. Although *H. juxtakochi* has not been involved as vector of human pathogens to date, in Brazil this tick was found...
infected with an organism closely related to *Rickettsia rhipicephali*, a SFG rickettsia\textsuperscript{22}. Bites of *R. microplus* in humans are uncommon, even among rural workers, but there are previous records of *R. microplus* ticks attached to humans in other Argentine provinces as Corrientes, Entre Ríos and Formosa\textsuperscript{23}. This tick species was not certainly involved as vector of human pathogens so far.

Despite using protective clothing so ticks could not climb from the vegetation to the researchers, some body parts like hands or neck were exposed for methodological reasons, therefore ticks could access from clothing to other parts of the body. The immediate removal of the ticks decreased the risk of transmission of pathogens to the researchers, as shown in Sood et al.\textsuperscript{24} and Katavolos et al.\textsuperscript{25}. The cases in which they remained attached for more than an hour were fortuitous.

All findings reported in this article are new records for Misiones because this province has been understudied.

The study site is characterized by large areas of highly endangered Atlantic rainforest directly contacting rural environments and peri-urban settlements, which determines a high rate of contact between domestic and wild animals and man. Furthermore, the INP receives more than a million tourists each year (INP, 2007-2014, unpublished data) from all continents. These people enter the tourist area of the Park and expose directly to tick bites during trail hiking. These features define a high rate of exposure of rural workers, peri-urban dwellers and tourists from around the world to tick bites, which determines a high risk of transmission of tick-borne pathogens and highlights the need for further study of ticks eco-epidemiology and public health relevance in the province of Misiones. It is also necessary to raise public awareness about the risk of tick bites and infection with pathogens, and to inform about protection methods against ticks.

**Acknowledgements:** We thank Nilso Molina and the research team of the INMeT for field assistance and samples donation, and Dr. Daniel Salomon for his comments on an early version of this article. We acknowledge the cooperation of the Administration of National Parks (APN), MEyRNR Misiones and the farm owners Jacobo Bernardi, Otto Waidelich and Alfonso Werle. Financial support was given by Instituto
Nacional de Medicina Tropical (INMeT) to DL, and by INTA, Asociación Cooperadora INTA Rafaela and Agencia Nacional de Promoción Científica y Tecnológica (PICT 526) to SN.

Conflict of interests: None to declare

References


All travel has its advantages. If the passenger visits better countries, he may learn to improve his own, and if fortune carries him to worst, he may learn to enjoy it.

Todos los viajes tienen ventajas. Si el viajero visita los mejores países, puede aprender a mejorar el propio, y si la fortuna lo lleva a los peores, puede aprender a disfrutar el suyo.

Samuel Johnson (1709-1784)

A journey to the western islands of Scotland (1775). London: Cape, 1951, p 217