ocular pain, low back pain, cutaneous rash, nausea, vomiting, diarrhea, and conjunctival injection. Serum samples were collected and diagnostic tests were performed including: reverse-transcriptase polymerase chain reaction (RT-PCR), NS1 antigen, IgM and IgG antibodies ELISA-based assays. Primary infection was diagnosed by the detection of NS1 antigen ELISA and/or RT-PCR and/or IgM ELISA in the absence of IgG ELISA. Secondary infection was determined by the presence of IgG ELISA and a positive NS1 antigen ELISA and/or RT-PCR and/or IgM ELISA.

Results: A total of 359 serum samples were collected in the study period. Dengue infection was diagnosed in 37.9% (136/359) of the patients, being 107 primary infections and 29 secondary infections. The most common serotype was DENV-3 in 77.5% of the cases. Differences were observed in the diagnostic performance of IgM ELISA (17.6% vs 55.2%, p≤0.01) and RT-PCR (74.8% and 31.0%, p≤0.01) in primary and secondary infections respectively. The combination of NS1 and IgM performed better than the other assays in detecting primary (91.9%) and secondary infections (96.6%). The most frequent symptoms associated with DENV infection were headaches, myalgias, and arthralgias.

Conclusion: We report an important outbreak of dengue infection caused by DENV-3 in Cajamarca, Peru. Our findings encourage the use of NS1 antigen and IgM co-detection. These findings demonstrate an increasing expansion of DENV-3 in Peru and highlight the importance of molecular diagnosis and serotype characterization among febrile patients to strengthen the Peruvian epidemiological surveillance.

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West Nile and Usutu Virus Circulation in Wild Birds and Equids in Piedmont, Liguria and Aosta Valley, Northwestern Italy, in 2018-2020

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Purpose: A mandatory plan on West Nile (WNV) and Usutu (USUV), aimed at an early detection of viral circulation, is implemented in Italy through an active and passive monitoring. Here we report data on WNV and USUV circulation from 2018 till 2020 in birds and equids in three Northwestern regions of Italy (Piedmont, Liguria and Aosta Valley).

Methods & Materials: Passive surveillance was carried out on wild birds found dead and as syndromic surveillance on neurological cases in equids. Active surveillance was performed on culled wild birds of target species (magpie, grey crow, jay). For each bird sampled, central nervous system and a pool of organs (heart, spleen and kidney) were analysed. For equids, analysis was carried out on blood samples and target organs (mainly spleen and central nervous system), from live and dead subjects, respectively. Viral RNA was extracted and purified using commercial kits and then assayed by real-time RT-PCRs detecting WNV Lineage 1 and 2, and USUV. All positive samples were sent to the OIE National reference Centre in Teramo (IZSTe) for confirmation.

Results: From January 2018 to December 2020 a total of 4602 animals were analysed by RT-PCR, of which 172 equids and 4430 wild birds. The formers ones comprised 124 samples in 2018, 29 in 2019 and 19 in 2020 while 1201 samples were collected from wild birds in 2018, 1492 in 2019 and 1737 in 2020, respectively. Among equids, 12 tested positive to WNV in 2018 while no positivities were found in 2019-2020. Among avifauna, 32 subjects tested positive to WNV in 2018, 37 in 2019 and 41 in 2020 while 4 subjects

resulted infected by USUV in 2018, none in 2019 and 2 ones in 2020. Positivities were found exclusively in samples from Piedmont and Liguria regions. WNV positive samples were all attributable to Lineage 2.

Conclusion: Surveillance on WNV and USUV allowed to detect viral circulation in birds and equids in Northwestern Italy (Piedmont and Liguria regions) in 2018-2020, aimed at reducing human risk of infection, mainly through a timely enforcement of blood donors screening.

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High Prevalence of Rickettsia Slovaca in Dermacentor Marginatus in Euganean Hills Regional Park

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Purpose: *Rickettsia slovaca* is a tick-borne zoonotic bacterium and is the etiological agent of SENLAT, Scalp Eschar and Neck Lymph-Adenopathy after Tick bite. Despite many human cases reported in Europe, its epidemiology is not completely understood.

To further understand this neglected tick-borne pathogen, we collected ticks and wild boar blood samples in 2017 to evaluate *R. slovaca* prevalence. Due to the high prevalence detected in *Dermacentor marginatus*, we investigated archive tick and blood samples collected previously in 2010 monitoring activities.

Methods & Materials: The research was conducted in Euganean Hills Regional Park, North-Eastern Italy. Sampling was performed in 2010 and 2017 and regarded both blood and ticks collected from wild boars at culling. In 2010, ticks were sampled from animals and from the vegetation using the dragging method.

Ticks were identified using identification keys and processed for DNA extraction. Biological samples were screened by real-time PCR targeting *Rickettsia gltA* gene and positive samples were tested by classical PCR targeting *gltA* and *ompB* genes for species identification.

Results: Overall, 89 wild boar blood samples were collected, 75 in 2010 and 14 in 2017. All samples tested negative. In addition, 254 tick samples were analysed, 233 sampled in 2010 and 21 in 2017.

Four different tick species were identified: *Dermacentor marginatus* (n=190), *Ixodes ricinus* (n=58), *Rhipicephalus sanguineus* (n=5), and *Hyalomma marginatum* (n=1).

A difference in *R. slovaca* prevalence was recorded among tick species, i.e, 23.68±6.05% in *D. marginatus* and 3.45±4.70% in *I. ricinus* while *R. sanguineus* and *H. marginatum* tested negative.

In addition, ticks collected by dragging were negative, while a prevalence of 18.68%±5.66 and 61.90±20.77 was detected in ticks from wild boars sampled in 2010 and 2017, respectively.

Conclusion: *D. marginatus* showed the highest prevalence of *R. slovaca*, revealing its important vectorial role. Comparing the tick sampling methods, dragging seems less informative than tick collection from wild boars. Since wild boars tested negative, their role in *R. slovaca* epidemiological cycle needs more investigations, while they act as a maintenance host of *D. marginatus* populations.

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