

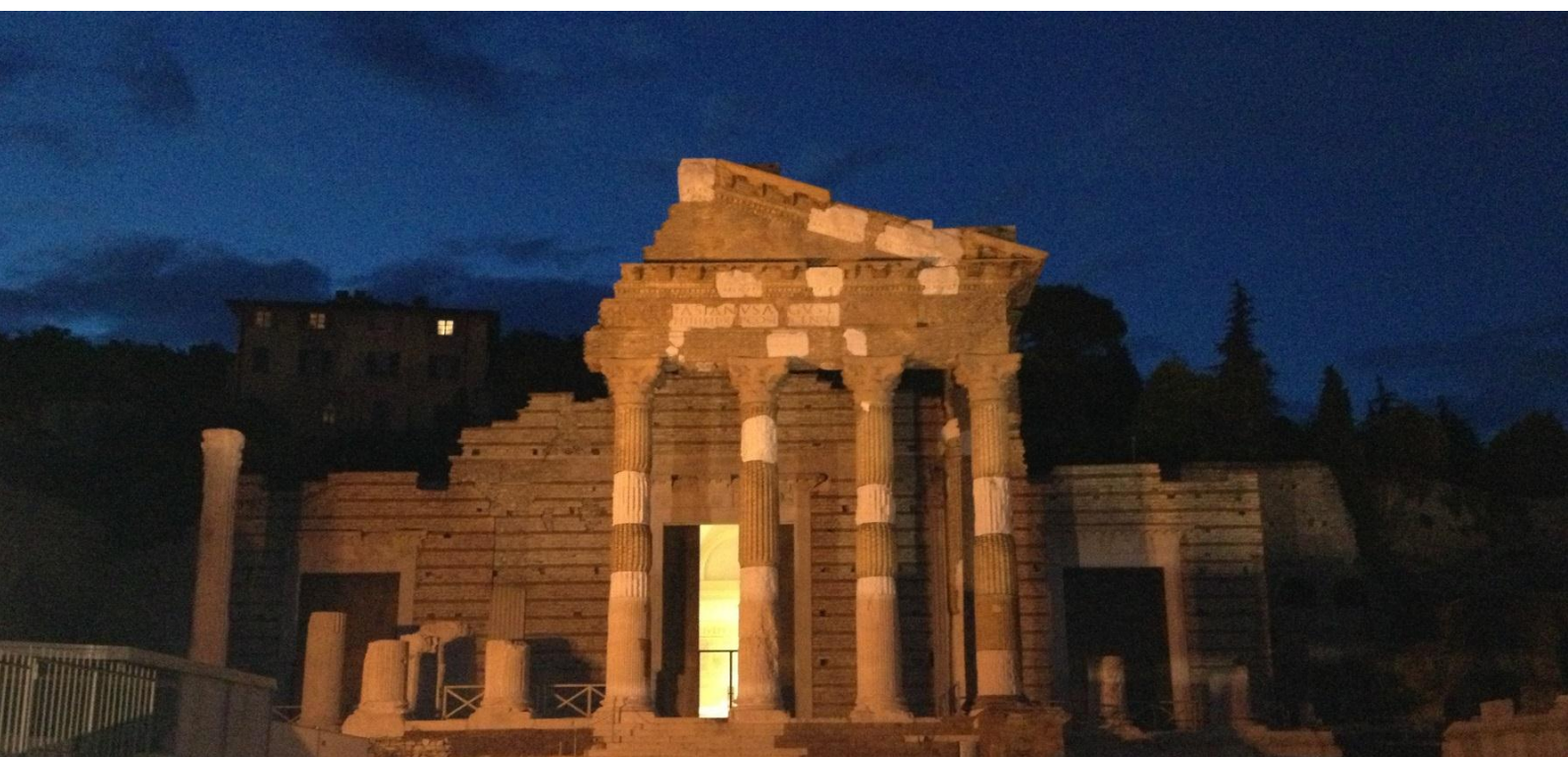
Brescia, Italy, 27-28 June 2013

"harmonized Approaches in monitoring wildlife
Population Health, And Ecology and Abundance"

APHAEA First Consultation Workshop

Abstract book

Two-days meeting organised by APHAEA in collaboration with IZSLER
(Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna)



Istituto Zooprofilattico Sperimentale della
Lombardia e dell'Emilia-Romagna
"B. Ubertini"

Outline Programme

Wednesday, 26th June, 2013 *Delegates arrival*

Thursday, 27th June, 2013 *Meeting place: IZSLER 9:00 – 18:10*

- 09:00 – 13:00 Morning for the core APHAEA partners only: where are we and how best to achieve our goals? (*Gortazar C. Ferroglio E. Staubach C., Ryser M.P. Herzig M., Ulrich R., Kuiken T. and others*)
- 13:00 – 13:30 Welcome and inscriptions of External Partners and EWDA colleagues and Posters setup
- 13:30 – 14:30 Lunch (free for all participants in the IZSLER canteen)
- 14:30 – 16:30 Invited Lectures (*Chair Staubach C.*).
A. "Harmonise monitoring of pathogens, surveillance plans" *Kuiken, T.*
B. "Population density/abundance" *Bonenfant, C.*
- 16:30 – 16:50 Coffee Break
- 16:50 – 18:10 APHAEA presentation session 1 (*Chair Kuiken T.*) 15'+5' discussion
1. "Monitoring wild bird populations for disease" *Esperón, F.*
2. "The inclusion of AIV of dabbling ducks in the surveillance of Poultry: a risk-based approach" *Galletti, G.*
3. "Bulgy-eyed (respiratory cryptosporidiosis) disease in red grouse from grouse (shooting) moors in Northern England" *Duff, P.*
4. "A decade of West Nile Virus surveillance in Hungary. Methods and lessons" *Erdélyi, K.*
- 19:30 Dinner

Friday, 28th June, 2013 Meeting place: IZSLER 9:00 – 17:00

- 06:30 – 08,30 Rodents snap-trap exercise on Monte Maddalena *Imholt, C. and Chiari M.*
- 09:00 – 09:10 APHAEA: introduction. *C. Gortazar*
- 09:10 – 09:35 APHAEA network development. *Ryser M.P.*
- 09:35 – 10:00 Upcoming APHAEA initiative: Pilot field studies *Staubach C.*
- 10:00 – 11:00 Invited Lectures (Chair *Ryser M.P.*)
- C. "Determinants of rodent population fluctuations and methodological approaches to estimate small herbivore density" *Imholt, C.*
- 11:00 – 11:30 Coffee Break
- 11:30 - 12:50 Presentation session 2 (Chair *Gortazar C.*)
5. "Bat White-Nose Syndrome and *Geomyces destructans* surveillance in Great Britain" *Barlow, A.*
6. "Serosurvey for Schmallenberg Virus in Alpine wild ungulates" *Chiari, M.*
7. "Wildlife health and surveillance in Norway" *das Neves, C.G.*
8. "Field monitoring on the fox population data in relation to sanitary surveillance in North-Eastern Italy" *Citterio, C.*
- 12:50 – 13,30 Poster discussion
- 13:30 – 14:30 Lunch (free for all participants in the IZSLER canteen)
- 14:30 – 15:30 Invited Lecture (Chair *Lavazza A.*)
- D. "Epidemiology, characterization and diagnosis of lagoviruses" *Capucci L.*
- 15:30 – 16:00 Conclusive remarks (Chair *Ferroglio E.*)
- E. "Integration of wildlife population monitoring with wildlife disease monitoring" *Gortazar C.*
- 16:00 – 17:00 Synthesis workshop Brescia
- *Ryser M.P.*
- *Gortazar C.*
- 17:00 Conclusions and farewell

Friday evening or at weekend delegates travel home

Invited Lectures

A. Bonenfant, C.

French National Center for Scientific Research, Université Claude Bernard, Villeurbanne, France

"Population density/abundance"

B. Kuiken, T.

Department of Viroscience, Erasmus University Medical Centre, Utrecht, The Netherlands

"Harmonise monitoring of pathogens, surveillance plans"

C. Imholt, C.

Friedrich-Loeffler-Institut, Federal Research Institute for Animal Health in Germany

"Rodents populations or rodent "borne" pathogen"

D. Capucci L.

Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna, Brescia, Italy

"Epidemiology, characterization and diagnosis of lagoviruses"

E. Gortazar C.

Instituto de Investigación en Recursos Cinegéticos, Ciudad Real, Spain

"Integration of wildlife population monitoring with wildlife disease monitoring"

Oral communications

1. Monitoring wild bird populations for disease

Esperón, F., Bosch, J.; de La Torre, A., Carballo, M., Muñoz, M.J.

Animal Health Research Center, National Institute for Agriculture and Food Research and Technology CISA-INIA, Valdeolmos, Madrid, Spain

Wild birds can play different roles in zoonotic or shared diseases, such as primary reservoirs, mechanical vectors, accidental hosts, “spill-over” hosts... As a consequence of that, before planning the monitoring study it's essential to take into account: 1) the nature and characteristics of the etiologic agent, 2) which species are related to the epidemiology of the disease and 3) the different aspects of the biology of these species. Also a major important issue is to adjust the human and financial resources to the purpose of the study.

Regarding the points 2 and 3, some questions which have to be solved along study design phase for each studied species are: What is its distribution? What is its phenology? What are their potential resources of their habitat? Concerning wild bird populations it could be interesting to know different parameters such as the geographical distribution, the population size, the population density and /or the abundance index. There are several bibliographic sources with reliable and standardized information about bird populations, ranging from regional to global scale. However, it is possible that a local scale we couldn't have enough basic information available about some population parameters. In these cases it may be necessary calculating the size of a population by means of an estimation of the real census. Popular methods are those based on bird surveys (mapping and transects), which they could be improved through habitat based stratification. Several starting considerations which should be taken into account in order to avoid compromising the success of the study are, firstly, the great biodiversity of a country that may cause the inclusion of multiple species of wild birds within a monitoring study (e.g. Spain hosts 337 species including native and introduced). Secondly, the population of study in a concrete area may be resident or migratory, breeding or wintering, colonial or individual, habitat generalist or specialist. Third, particularly, the size of the migratory bird populations varies considerably along the year and also can be affected with the weather conditions. Therefore, the estimation of the bird population must include different spatiotemporal scenarios, which are essential when we undertake a monitoring study.

Finally, when monitoring studies are going to be applied to national or international scale, “predictable models of habitat suitability” and “probabilistic models of species presence” are less time consuming tools. These studies are constructed from the available data both biological and the census. In this presentation, some different monitoring studies will be reviewed taking into account the spatiotemporal scale.

2. The inclusion of the AIVs in dabbling ducks risk, in the surveillance plan of AIV in Poultry

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² Regione Emilia Romagna, Servizi Veterinari, Bologna, Italy

³ Istituto Superiore Protezione e Ricerca Ambientale, Ozzano E. (BO), Italy

Avian Influenza Viruses (AIVs) have caused and are still causing a huge economic loss in the commercial poultry sector in Italy. Wild ducks, and dabbling ducks in particular, are recognized as the main epidemiological reservoir of AIVs, including both the H5 and H7 strains. The back yard poultry sector is supposed to behave as the eco-epidemiological bridge between wildlife and the commercial poultry sector either directly or indirectly through, for instance, contaminated vehicles and operators.

In Italy, there is currently a AIVs poultry surveillance plan in place; the plan is drawn considering specific risk factors (e.g. multispecies, low biosecurity, breeding system etc.). In deciding the sampling strategy, the risk posed by wildlife is poorly considered and not standardized. To improve the capacity of AIVs early detection in the different poultry sectors, and especially the detection of H5 and H7 LPAI, the wildlife risk should be taken into account. The *waterfowl risk* could be weighted in terms of distances between holdings and wetlands, number of birds in each one of the wetlands and AIV prevalence in waterfowl. Relevant data are already available: capture-recapture data of ringed birds caught during the same winter in Italy; shape file of wetlands of the Emilia-Romagna region and other neighboring regions; waterfowl winter census; average winter AIVs prevalence in waterfowls. The above data were used to obtain a *waterfowl risk* map of Emilia-Romagna region, using the weighted local density of infected waterfowl as a risk index. Capture-recapture data of ringed birds indicate the flight distances of birds in winter. Distances were organized according to a cumulative distribution and the percentiles of all the flight distances were calculated. Then a buffer around each wetland was calculated using the decreasing distance recorded in cumulative distribution percentiles. The number of waterfowl birds in each buffer was estimated as the bird population of the wetlands weighted for the percentiles associated with the distance that had generated the buffer. This value was multiplied by the average winter AIVs prevalence and divided by the buffer area in square km to obtain the density of infected waterfowl birds for each buffer.

The whole region was covered by a regular grid (cell size of 1km x 1km). The sum of the densities of infected waterfowl was calculated in each cell, giving an estimate of the *waterfowl risk*. This risk is represented by the density of infected birds that can arrive in each cell (with a probability determined by the distances) from the wetlands of Emilia-Romagna and the neighboring regions. The model runs for the winter period only, since data on AIV prevalence during summer are still lacking.

Future studies should be addressed to estimate the number of nesting pairs of dabbling ducks in the area, the reproductive rate, the summer AIV prevalence and, finally, the AIV prevalence in new born birds after fledging.

3. Bulgy-eyed (respiratory cryptosporidiosis) disease in red grouse from grouse (shooting) moors in Northern England

Duff¹ J.P., Errington¹ H., Wood² A.

¹ AHVLA (Animal Health and Veterinary Laboratory Agency) Diseases of Wildlife Scheme (DoWS) AHVLA Penrith England Cumbria CA11 9HS paul.duff@ahvla.gsi.gov.uk

² AHVLA DoWS, AHVLA Lasswade, International Research Centre, Pentlands Science Park, Bush Loan, Penicuik, Midlothian, EH26 0PZ.

The red grouse (*Lagopus lagopus ssp scoticus*) is a gamebird of upland heather moorland in Britain. The British red grouse has subspecies designation and feeds mainly on heather (*Erica* sp). Red grouse hunting we believe is a peculiarly British form of specialised hunting which begins each year on the 12th August (giving rise to the term established in the English language – ‘the glorious 12th’). Coveys of grouse are driven over treeless moors to shooting butts dug into the hills where the fast flying birds are shot from – there is no other game species hunted at these times – only grouse. The shot grouse may be eaten.

Grouse shooting is a multi-million pound industry and in places the only industry in these hills. The moorland is managed for the grouse, to improve heather growth which as a consequence have high population densities of grouse. This, as would be expected, encourages density dependent diseases, in particular trichostrongylosis which historically has been a significant limiting factor in maintaining grouse numbers. Trichostrongylosis in red grouse was a rich area for important publications on wildlife disease ecology, modelling and wildlife disease control.

In 2010 AHVLA Diseases of Wildlife Scheme (DoWS) and Diseases of Poultry Project identified the first cases of a new disease characterised by swollen eyes in affected birds. Histopathological evidence of respiratory cryptosporidiosis was seen and identified as *Cryptosporidium bayleii* which is considered to have low zoonotic potential. The disease will be described.

This new and emerging disease has not been recorded before in grouse was detected by opportunistic scanning surveillance. In the UK other forms of surveillance are utilised for wild bird surveillance. These include:

- scanning surveillance
- bird reserve, daily man patrol surveillance (for Avian Influenza Virus - AIV)
- trapped waterbird swab (cloaca and pharynx) surveillance (for AIV)
- swab surveillance of hunter-shot waterbirds intended for human consumption (for AIV)
- caged sentinel chicken surveillance (for West Nile Virus WNV) – used once in the UK
- targeted surveillance for WNV, prioritising coastal migration sites, raptors, corvids and migrant species of wild birds.
- tideline walking of beaches to detect seabird mass mortality events.

If time permits, these subjects will be discussed further.

4. A decade of West Nile Virus surveillance in Hungary. Methods and lessons

Erdélyi¹ K, Csörgő² T., Ferenczi³ E., Barna⁴ M., Szentpáli-Gavallér¹ K., Papp⁵ L., Soltész² Z., Fehérvár⁵ P., Bakonyi⁴ T.

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West-Nile virus (WNV) circulation in the Old World had primarily been documented and tracked through the diagnoses of human cases. The “silent” nature of the bird-mosquito cycle leading to the amplification of WNV and disease outbreaks in humans and horses had been taken for granted. However, the introduction, rapid spread and widely documented patho-ecological features of WNV infections in the Americas had partially flipped this perception to the opposite side, leading to the association of WNV outbreaks with high bird (primarily corvid) mortality regardless of geographic location. Some however retained the old wisdom of regarding avian host species as a homogenous entity amplifying and spreading the virus, but generally useless in epidemiological control, therefore retaining the focus on mosquito vectors as the “weakest” link in the epidemiological chain. The focus on mosquito vectors has practical merits, particularly when we consider the wide spectrum of other emerging pathogens that may also be detected and monitored through arthropod surveillance. Yet, one must also untangle the web of avian host related issues in order to unravel the true ecological factors driving the epidemiology of WNV in Europe (particularly factors related to introduction, endemicity, spread, etc.).

Our team has been documenting the introductions, virtually continuous presence, circulation and spread of WNV (both lineage 1 and 2 strains) in Hungary since 2003. The localised presence was followed by the expansion of lineage 2 WNV to Austria in 2008, Greece in 2010, and recently Italy in 2011. Although high rates of clinical disease and mortality of wild bird species were never associated with European WNV outbreaks, WNV related clinical disease and mortality has been regularly detected by a semi-targeted passive surveillance in both free ranging and captive wild birds during the past decade. Since 2003 we detected more than 50 cases of lethal WNV cases in goshawks (*A. gentilis*), which were the most reliable indicators of WNV circulation. Disease and mortality also occurred in other captive, non indigenous and indigenous bird species, as well as in free ranging red footed falcons (*F. vespertinus*) and sparrowhawks (*A. nisus*). Subclinical WNV infection was also detected in a wide range of bird species. Focal surveillance of mosquito vectors had also been initiated. The aim of our long term study is to identify the most potent amplifying, reservoir and sentinel host species, pinpoint the most likely migratory species implicated in the spread of WNV and provide baseline data for risk assessment, clarifying the potential of WNV for geographic spread and its potential impact on species conservation, wildlife, human and domestic animal health.

The presentation summarizes the evolution, technical features and results of this multidisciplinary research effort and provides an attempt to evaluate the strengths and weaknesses of this approach and applied methods.

5. Bat White-Nose Syndrome and *Geomyces destructans* surveillance in Great Britain

A. M. Barlow

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Geomyces destructans, a fungal infection of insectivorous hibernating bats is the cause of White-Nose Syndrome (WNS). It was first reported in hibernating bats in, New York State, in February 2006. WNS has caused mass die-offs of bats in Eastern USA and Canada. It is estimated that WNS has killed over 5.7 million bats in eastern North America. Nine bat species are affected and WNS or *G. destructans* is present in 22 U.S. states and five Canadian provinces. Mortality can be up to 100% in the worst affected hibernacula. This is the most catastrophic decline of Northern American wildlife caused by an infectious disease ever recorded. The depletion of this number of insectivorous bats may lead to a significant increase in insect population. These insects may be vectors for zoonotic diseases, diseases of farmed and wild animals and may also be crop pests. This level of mortality could lead to the possible extinction of rare bat species in North America. Severely affected bats can have visible fungal growths around the muzzle, ears and wing membrane. A specific fungus, *G. destructans* has consistently been isolated from affected bats. Cold, dark, humid hibernacula provide an ideal environment for *G. destructans*. The hibernating bats have lowered body temperatures and no immune response to infection. Evaporative water loss (EWL) during hibernation is 99% from the skin, mainly the wings. Fungal colonisation of the skin especially the wings causes increased water loss by the "wick" effect of fungal growth. Also there is loss of electrolytes and protective secretions, which moisturise and waterproof skin. Dehydration leads to increased arousal with shorter torpor bouts. Arousal and flight uses energy/fat reserves. Thus it is proposed that that hypotonic dehydration and emaciation are the causes of death of affected bats. *G. destructans* has now been isolated from bats in at least 14 continental European countries including France, Belgium and the Netherlands. It is also suspected to be present in at least four more including Denmark and Turkey. It has been detected in eight *Myotis* spp., six of which are present in GB, including Whiskered, Daubenton's, Natterer's, Bechstein's, Brandt's and Greater mouse-eared bat. However no mass mortality events have yet been reported. The Animal Health and Veterinary Laboratories Agency (AHVLA) wildlife group vets, working with bat researchers from the Bat Conservation Trust (BCT) and the Vincent Wildlife Trust, have been investigating cases of fungal infection in hibernating bats in Great Britain since January 2010. Surveillance has been extensive. There are currently almost 5000 members of BCT with 89 bat groups around GB. They have visited 543 hibernation sites over the last two winters. 48 incidents in Great Britain were investigated from January 2010 to March 2013. These were all been single or low numbers of dead bats with two samples received from suspect infection in a live bat. At least eight different bat species have been examined. A variety of fungi were isolated from skin fungal growths on the dead bats. All were considered to be opportunistic saprophytic fungi on dead tissue. No mass mortalities have reported and *G. destructans* has not been isolated from any of the samples from Great Britain. The present research suggests that *G. destructans* was probably introduced to North America from Europe into a naïve bat population. European bat species are likely to have co-evolved with *G. destructans* and thus have an innate resistance. However it is still unclear if indigenous British bats will be susceptible. Unpublished work by Puechmaille and others (2011) using ecological niche modeling have predicted that the potential distribution of *G. destructans* in Europe would include a large part of GB. So this Winter 2012/2013 the BCT/AHVLA/Northern Arizona University carried out a hibernacula survey using a specific *G. destructans* PCR to try and confirm its presence in GB. Six hibernacula were selected by; proximity to Continental Europe, if suspect cases had been found at these sites, number of bat species recorded and variety of hibernacula e.g. chalk, sandstone, brick, etc.. The results are awaited.

6. Serosurvey for Schmallenberg Virus in Alpine wild ungulates"

Chiari M., Sozzi E., Zanoni M., Alborali L.G., Lavazza A., Cordioli P.

IZSLER - Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna "Bruno Ubertini", Brescia, Italy

Schmallenberg virus (SBV), a novel member of the family *Bunyaviridae*, genus *Orthobunyavirus* and Simbu serogroup, was detected in cattle in North-Western Europe in 2011. SBV infections have been reported as the cause of congenital malformations and stillbirths in cattle, sheep, and goats. The first case in Italy was reported during mid-February 2012 in cattle in the Veneto region. Seropositivities in wild animals were assessed in various countries, but the occurrence of SBV infection and the presence of specific SBV antibodies have not yet been described in any wild ruminant species in the Alpine region. Therefore, we conducted a serosurvey to assess the presence of SBV-specific antibodies in free-ranging alpine ruminants in an area located between Stelvio and Adamello National Parks characterized by a high red deer density (16 animals/100 hectares). Serum samples from chamois (23) and red deer (352) hunted from 2007 to 2013 were tested by two different ELISA kit (ID Screen[®] Schmallenberg Virus Competition, IDvet, and IZSLER homemade kit) and confirmed by the virus-neutralization test (VNT), using Vero cells and SBV strain BH80/11-4 (RBV 1099-FLI), (Friedrich-Loeffler-Institut, Greifswald-Insel Riems, Germany).

All of the serum samples collected through September 2012 tested negative, whereas a single chamois serum and 21 red deer sera taken during the 2012-2013 hunting season tested positive.

Vector transmission of SBV has been demonstrated; in particular, SBV has been detected in *Culicoides* spp. in different European countries. Midges could play an active role in the spread of the virus, and their implication in the epidemiological cycle could explain the fast and wide spread of SBV. The diffusion of SBV in the study area could have occurred between the January and December 2012. Biting midges of the genus *Culicoides* are not usually active in the Alpine region from week 47–49 to week 12–14, depending on weather conditions. SBV infections could be occurring in the warmer months when *Culicoides* can reach high altitudes, including alpine summer pastures.

SBV is not (yet?) an OIE World Animal Health Information System notifiable disease, but the economic impact of the disease at farm level should be considered, particularly in the rural areas of the Alps. Conceivably, domestic ungulates grazing on common pastures with red deer and chamois during the summer could confer a role for wildlife in the epidemiology of SBV in the alpine environment.

To our knowledge, this is the first report of a SBV infection in Alpine wildlife in Italy. Thus, there should be the targeted surveillance of wild ruminants to monitor the spread of the virus and to assess the epidemiological role of wildlife at the interface with domestic animals.

7. Field monitoring on the fox population data in relation to sanitary surveillance in North-Eastern Italy

Obber¹ F., Capello¹ K., Gagliazzo¹ L., Mulatti¹ P., Mutinelli¹ F., De Nardi¹ M., Somnavilla² G., Berto³ M., Calabrese⁴ MS., Ferraro⁵ E., Carmignola⁶ G., Cadamuro⁷ A., Capelli¹ G., Citterio¹ CV.

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² Provincia di Belluno – Italy,

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⁷ Regione Friuli Venezia Giulia – Italy

In the last 7 years, North-Eastern Italy has been affected by severe epizootics in wild carnivore populations, spreading westwards from the eastern Italian border: a first epidemic of canine distemper started in the summer of 2006, lasting until 2010 and progressively spread to the Friuli Venezia Giulia, Trentino-Alto Adige and Veneto regions; then, from October 2008, a rabies epidemic occurred in the fox population, with the last case in February 2011 after repeated oral vaccination campaigns; finally, a second distemper epidemic, still ongoing, has been observed since 2012. Considering the unexpected dynamics of these diseases, and in particular for rabies the unexpected speed compared to the past (more than 100 km in a year), it could be hypothesized that the distribution and population dynamics of wild carnivores may have changed with respect to the past. In particular for the red fox, the most affected species and recognized reservoir for rabies, such changes could have occurred in parallel with modifications in the environmental conditions. Therefore, due to the role of the red fox in the cycle of different important zoonoses, from rabies to *Trichinella* and *Echinococcus multilocularis*, and considering also the bordering of our territory to other countries, it appears worthwhile to develop a system to monitor the fox status in the field, at least in terms of population distribution and trend, in order to anticipate possible epidemiological scenarios, to focus possible active surveillance programmes and to help risk assessment (e.g. for *Trichinella*).

In North-Eastern Italy, very few data are available concerning fox population, since foxes are hunted but there is a scarce hunting tradition and interest in this species; thus even hunting statistics are often not available. We therefore proposed a simple method, based on the following topics:

- Exploitation of daily recording from each campaign for monitoring of the effectiveness of rabies oral vaccination, by a specific fox hunting form. This activity, now finished after the successful eradication of rabies, was performed in different periods during the year (30 days after the conclusion of each vaccination campaign), and was aimed at evaluating the repeatability of the hunting effort, also obtaining indexes as the number of contacted foxes/time in case of fox hunting from fixed locations, or a Kilometric Abundance Index (IKA=fox number/km) in case of roaming.
- Improving and harmonizing the collection of regular hunting statistics, at least where some fox hunting tradition is present and is performed in team, by the use of a specific hunting card.
- Spring night counts, using a specific fox counting form, exploiting the spring censuses for red deer, in order to reach a more complete spatial distribution and a higher standardization in collecting data and IKA calculation.

An effort is in progress in order to: detect and remove/reduce the sources of bias in fox population counting, standardize counting methods and obtain robust and simple field population trend indicators.

8. Wildlife health and surveillance in Norway

Das Neves, C.G.

Norwegian Veterinary Institute, Department for Health Surveillance, Section of Veterinary Public Health, Wildlife group – Oslo, Norway

The Norwegian Veterinary Institute - NVI is a national biomedical research institute in the fields of animal health, fish health and food safety, whose primary functions include surveillance and diagnostic regarding different species and diseases as well as research and advisory support to the governing authorities.

The wildlife group, consisting of 4 veterinarians/researchers and 1 technician, is responsible for all wildlife species (both land and marine species) in cooperation many times with other departments at NVI and other institutions such as the Institute of Marine Research, the Norwegian Directorate for Nature Management or the Norwegian Institute for Nature Research.

NVI carries both passive and active surveillance programs based for example on routine necropsies of wild species sent to NVI, or on active collection of samples during hunting periods or specific field actions. The Health Surveillance Program for Wild Cervids is the largest active surveillance program in wildlife at NVI and is also the basis for different research projects carried by the group. Other programs have focused on diseases such as Chronic Wasting Disease, Tuberculosis, Rabies or Echinococcus and species such as wild boars. Furthermore NVI is involved in several research projects concerning wildlife health, transmission of diseases between wild and domestic animals and diseases of zoonotic potential. An overview of all these activities, methods and strategies used is given in this presentation.

Poster presentations

P1. Surveillance for zoonotic pathogens in Barcelona periurban wildlife, Spain

J. Millán*, T. Proboste, A.D. Chirife, J. Serra-Cobo, L. León-Vizcaíno, L. Altet

Servei d'Ecopatologia de Fauna Salvatge Wildlife Diseases Research Group / Departament de Medicina i Cirurgia Animals Facultat de Veterinària Universitat Autònoma de Barcelona *syngamustrachea@hotmail.com

The human encroachment in natural environments is one of the causes of the apparent rising in the emergence of zoonotic diseases of wildlife origin. In 2011 we started a research project aimed to detect selected zoonotic pathogens in periurban wildlife inhabiting residential areas in the metropolitan area of Barcelona (Spain) and to determine factors that can affect its prevalence, such as type of habitat (natural vs. residential), host abundance and season. We study two taxonomic groups: micromammals and carnivores. The first group is relevant as reservoir of important zoonotic agents; carnivores can also act as reservoir of some zoonoses but are also interesting as sentinels of others because they prey upon micromammals. Domestic cats and dogs are also being studied as link between humans and wildlife. So far, we have sampled more than 600 micromammals (chiefly *Apodemus sylvaticus*) and about 200 carnivores (including red foxes, genets, pine martens and domestic dogs and cats belonging to inhabitants of residential areas). We have detected infection by pathogens of the genus *Anaplasma*, *Ehrlichia*, *Rickettsia*, *Borrelia*, *Bartonella*, *Coxiella*, *Leishmania*, *Leptospira* and *Salmonella*, many of them of confirmed zoonotic potential. We have not detected yet *Babesia microti* or Hantavirus.

P2. Serological and molecular survey of Aujeszky's disease in wild boar (*Sus scrofa*) in Northern Italy

Chiari M., Canelli E., Zanoni M.G., Cordioli P., Alborali L.G.

IZSLER - Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna "Bruno Ubertini", Brescia, Italy

Aujeszky's disease (AD) is a highly contagious, economically significant disease caused by Pseudorabies virus (PrV), principally affecting wild and domestic pigs. AD can result in trade restrictions from regions where it is endemically present. As a consequence, eradication programs in swine and surveillance programs on wild boar are ongoing. Although reports of PrV transmission from wild boars to domestic pigs are sporadic, success in disease eradication programs in the livestock could be influenced by wildlife reservoirs. Despite wild boar seems to serve as a persistent reservoir for PrV, there is still a lack of data describing the AD epidemiology in free ranging wild boars population. Therefore, the aim of this study was to determinate the temporal dynamic of AD infection in wild boars hunted in an area of North Italy. A total of 2847 sera samples of hunted free-living wild boars were collected from 2006 to 2013 and then tested by a competitive ELISA for AD-gE. Additionally, from 2011 to 2013, 878 wild boar amygdalae were collected and analysed by real time-PCR in order to investigate the presence of PrV in a latent form. In the last decades, an overall increasing of the wild boar populations was registered in the 8 hunting districts here considered. They are located in the footstep mountain in Northern Italy (Province of Brescia, Lombardy), where swine herds are not present. One hundred and nineteen wild boars (4.2%) were seropositive and real time PCR resulted positive in 8 cases (1.22%). Even if the infection is present since 2006, seroprevalences higher than 9.4% have never been detected. PrV is present and persistently circulating in an isolated population, located in an area where swine herds are not present. The obtained data and the absence of pigs farms in the study area could indicate that wild boars can be considered maintenance host for PrV regardless of the presence of pig farms.

P3. Wildlife Disease Monitoring and Surveillance Systems in the Netherlands

Maas¹ M., Gröne² A., H.I.J. Roest³, van der Giessen¹ J.W.B.

¹ National Institute for Public Health and the Environment (RIVM),

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Monitoring and surveillance of (emerging) wildlife diseases is important to receive signals of pathogen circulation of interest for veterinary and public health. Structured overviews of national monitoring and surveillance of wildlife species, including the institutes where this takes place, will be of benefit for (future) national and international collaborations and may be used to identify gaps in the current programs.

We described the current wildlife disease Monitoring and Surveillance Systems (MOSSs) in the Netherlands, illustrated with flow charts per (group of) wildlife species (wild boar, wild carnivores, rodents, cervids, lagomorphs, birds, marine mammals, reptiles/amphibians, fish and bats). Comparison of the Dutch wildlife MOSSs with that at European levels (e.g. APHAEA) and those from countries close to the Netherlands, showed that overall the key animal species, as well as most of the key pathogens, are included in our programs. Though in this general overview no urgent gaps in the MOSSs have been identified, a detailed evaluation per program is recommended, to test if the programs are adequately designed to meet their aims. The current MOSSs could be optimized by additional vector monitoring programs, as well as programs to monitor invasive species and their relevant pathogens. Tissue banks ensure optimal use of the animals and may be extended for some species, e.g. wild boar.

P4. Molecular Typing of *Echinococcus multilocularis* Isolates from Germany

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Alveolar echinococcosis (AE) is a serious zoonosis caused by the tapeworm *Echinococcus multilocularis*. If the disease remains untreated in humans, it may take a lethal course. Infection occurs through ingestion of eggs of *E. multilocularis*. In Europe, the red fox (*Vulpes vulpes*) is the main definitive host of the parasite and rodents serve as intermediate hosts. Other carnivores such as raccoon dogs, (domestic) dogs and cats also represent definitive hosts of the parasite. *E. multilocularis* occurs in the temperate and arctic region of the northern hemisphere.

Since little is known about the genetic diversity of this tapeworm, we set out to study this feature in Brandenburg, Germany, because a large collection of parasite isolates with accompanying information on the infected hosts is available that covers the entire region over a long period of time. Three mitochondrial markers and the microsatellite EmsB are used for genotyping. The PCR products of the mitochondrial targets will be sequenced to find differences between individual tapeworms. Moreover length polymorphisms of the EmsB microsatellites are determined. The respective protocols are harmonized with other groups in Europe to ensure that the results are comparable. It is expected that the typing results and the geographic information on the hosts will allow analyzing the spatial and temporal distribution of different isolates using maps and dendrograms in the course of phylogenetic studies.

We are looking forward to enhance our existing co-operation on the mutual exchange of methods and samples and invite further partners to participate in our project.

P5. Prioritising infectious diseases, their early detection and control scenarios, in the wild ungulate populations of the North-Eastern Italian Alps

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The early detection of a pathogen is aimed at reducing the high-risk period of an epidemic during which the infection can spread uncontrollably. The length of the first high-risk period mainly depends on the awareness and readiness regarding the possible risk. The second high-risk period elapses from the first diagnosis until the enforcement of control measures. The efficacy of the control measures is related to the knowledge of: i) ecology of the involved pathogen; ii) ecology and demography of the host population/s; iii) epidemiology of the pathogen in the susceptible populations. The capacity to interrupt the chain of the infection is represented by the ability to halt the ecological relationships that link the pathogen to the host population.

In the eastern Alps the amount of wildlife, and mainly of ungulates (red deer, roe deer, chamois, ibex, moufflon and wild boar) is consistently increasing. As a consequence, contacts at the wild/domestic animal-human interface have also been increasing, making the circulation of pathogens between the different components of the ecological interface more likely. In such a situation, early detection and the prompt response to the introduction or re-introduction of a pathogen is a pre-requisite of any human and animal health risk management model.

In this context, a project has been undertaken, coordinated by the Istituto Zooprofilattico delle Venezie (IZSVe) and including different institutions for wildlife management of the north-eastern Italian alpine area, with the following key points:

- **prioritization:** assess which infections have the highest risk of being introduced (or re-introduced) at the wild ungulate-domestic animal-human interface, and which of them will be more likely to have clear health implications, according to the following priority criteria: 1) important zoonoses; 2) diseases in wildlife which could compromise national/local control/eradication programmes in livestock and/or have implications in animal and foodstuff national and international trade; 3) diseases important for wildlife conservation, with an impact on wild population dynamics; 4) other diseases;
- **data harmonisation:** collect and integrate relevant data on the possible host populations (wild ungulates), in order to build a spatial data infrastructure as a basic instrument for planning both surveillance and (if this is the case) control/eradication activities;
- **early detection and eradication scenarios:** develop diagnostic tools and surveillance strategies aimed at the early detection of the identified infections, and design of a risk management model for feasible control scenarios.

P6. Salmonella serotypes in wild boars (*Sus scrofa*) hunted in northern Italy

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Salmonella species (spp.) are zoonotic enteric bacteria able to infect humans, livestock and wildlife. *Salmonella* spp. have been sporadically isolated from the intestinal content of wild birds and mammals including white-tailed deer, rabbits and wild boars (*Sus scrofa*). However, little is known about the prevalence and the presence of the different serovars in wildlife.

Over six hunting seasons, the intestinal contents of 2,729 boars hunted in northern Italy were sampled and cultured. *Salmonella* spp. was isolated following the methods reported in "Annex D ISO 6579:2002", mandatory in the implementation of *Salmonella* monitoring and control plan for primary productions. All presumptive *Salmonella* spp. isolates were confirmed using biochemical tests.

Salmonella spp. were isolated from 629 boars (21.06%). The isolates belonged to 48 different serovars classified into three different subspecies of *S. enterica*: *S. enterica* subsp. *enterica* (n=489 strains, 77.75%, 38 serovars), *S. enterica* subsp. *diarizonae* (n=73, 11.60%, 7 serovars) and *S. enterica* subsp. *houtenae* (n=67, 10.65%, 4 serovars). Thirty-eight serovars of *S. enterica* subsp. *enterica* were found, including the human pathogens *S. Typhimurium* and *S. Enteritidis*, as well as *S. Napoli* and *S. Enterica 4,5,12:i:-*), a monophasic variant of *S. Typhimurium*, some of which are emerging serovars both in humans and animals. In addition, the identification of serovars belonging to *S. enterica* subsp. *diarizonae* and *S. enterica* subsp. *houtenae* prove how wild boars may act as healthy carriers of a wide range of *Salmonella* serotypes.

Considering the widespread occurrence of wild boars in Europe and the feeding behaviour (omnivorous scavengers), this specie may be considered a good indicator for environmental presence of *Salmonella* spp. Therefore, the epidemiological role of this species in relation to salmonellosis might be relevant and should be further investigated.

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16	DVM Chiari, Mario mario.chiari@izsler.it	Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna	IZSLER	Via Bianchi 9, 25124 Brescia, Italy	Lecture: "Serosurvey for Schmallenberg Virus in Alpine wild ungulates" Posters "Serological and molecular survey of Aujeszky's disease in wild boar (<i>Sus scrofa</i>) in Northern Italy" "Salmonella serotypes in wild boars (<i>Sus scrofa</i>) hunted in northern Italy"
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