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### Keynote speakers' biographies and abstracts

#### Dr René van den Brom

Manager Small ruminant department of Royal GD, Royal GD, Deventer, Netherlands

In 2008, René started working part-time at Royal GD, Deventer, as veterinarian in small ruminant health. Due to the human Q fever outbreak with a suspected relation to dairy goats, a large portfolio of (veterinary) research started with the aim to obtain additional information on *Coxiella burnetii* infections in small ruminants, in which René participated from the beginning. Since 2010, René



works full-time at GD Animal Health, and from that moment on he started writing scientific papers. His external PhD-program started after publication of his first research paper. In 2013, René qualified as specialist in small ruminant health management, and currently is diplomat of the European College of Small Ruminant Health Management (Dip.ECSRHM). In 2015, René received his PhD-degree on his thesis "Veterinary aspects of a Q fever outbreak in the Netherlands between 2005 and 2012". René has an extra interest in infectious diseases in small ruminants. Since 2023, he was involved in the advisory board of the Ministry LNV, regarding the bluetongue serotype 3 outbreak.

# Keynote Address: Bluetongue serotype 3 in the Netherlands; clinical pictures, pathology, spread, impact, preventive measures and anticipation on overwintering

Bluetongue (BT) is an infectious, non-contagious vector borne disease that is caused by bluetongue virus (BTV). Notifiable BTV serotypes 1-24 are transmitted by midges of the genus *Culicoides* spp. BTV causes clinical disease and mortality particularly in sheep, although disease in other ruminant species and camelids occurs. Clinical signs are the result of endothelial damage and as a consequence oedema, haemorrhage, muscular degeneration and necrosis occur. An overview of the BTV serotype 3 outbreak in the Netherlands in 2023 will be presented.

Clinical signs indicating BT were first reported on 3 September 2023. Two veterinary practices located in the middle of the Netherlands notified sheep with clinical signs indicative of BTV on five farms to the Dutch Food and Consumer Product Safety Authority (NVWA). A team of veterinary experts (Royal GD, NVWA and practitioners) visited the farm the next day. Blood samples were taken and submitted for BTV testing to the Dutch reference laboratory Wageningen Bioveterinary Research (WBVR) in Lelystad. BTV infection was confirmed on 5 September 2023 after more than ten years of BT-freedom in the Netherlands. Based on whole genome sequencing, WBVR typed the causative BTV as serotype 3 on 8 September, which was officially confirmed later by the European Union Reference Laboratory (EURL) in Madrid.

Immediately after the detection of BTV-3, presence of BTV infections was retrospectively investigated by testing bulk tank milk samples from cattle of August for the presence of BTV-specific antibodies. Further, serum samples from sheep and goats were investigated by serology. No earlier BTV infections were found, indicating that the index cases were close to the site and time of incursion. In the weeks after the start of the outbreak, BTV-3 spread rapidly over the Netherlands. Unfamiliar with this specific serotype, clinically infected small ruminants were extensively investigated by clinical and pathological examination. Apathy, excessive salivation, erosions of gingivae, and lameness were most frequently noticed. Pathological findings showed severe internal damage to organs as lungs, kidney, and rumen, but also in muscle, joints and coronary band. The impact on the livestock industry was enormous. Especially, in sheep, morbidity and mortality rates were high. The impact on mortality in the small ruminant population was quantified on identification and registration census data from 2020 until the end of 2023.





During this BTV-3 outbreak period in 2023, an additional 55,000 sheep has died compared to the same period in 2020-2022. At flock level a high variety in mortality was observed, with a clear increase in mortality in both flocks that notified BTV-3 or were located in infected areas. In goats, an increased mortality of around 4,000 additional dead goats was observed. However, the association with BTV is more difficult to interpret.

In 2023, BTV-3 was detected and identified in an early phase by the Dutch monitoring and surveillance system. The disease has spread rapidly over all twelve provinces in the Netherlands. To date, BTV-3 has also been confirmed in Belgium, Germany and the UK. Although sheep exhibit the most severe clinical signs and highest mortality rates, also a large impact in other ruminants was observed. After the BTV-8 outbreak, congenital malformations in calves were observed. In 2024, incidentally BTV-3 was shown in aborted foetuses and in one case congenital malformations, related to BTV-3, of the brain were observed in a calf. Given the impact and the assumption that overwintering of the virus is likely, availability of effective and safe vaccine is of utmost importance, and fortunately currently the first BTV-3 vaccines are available in the Netherlands, to avoid further losses to the sheep, cattle and goat population due to BTV-3 in 2024.





#### **Professor Emerita Paula Menzies**

University of Guelph, Canada

Paula Menzies has been a teacher, practitioner, and researcher in sheep and goat health management for over four decades in Ruminant Health Management at the University of Guelph, Canada, where she is now Professor Emerita. Since 2017 she is a member of the Advisory Committee to the WOAH UN FAO Peste des Petits Ruminants Global Eradication Program, with the goal to eradicate this viral disease of sheep and goats from the world by 2030. She is also a Dip.ECSRHM and active on the boards of the ECSRHM, International Sheep Veterinary Association and the International Goat Association.



#### **Keynote Address: Peste des petits ruminants**

Peste des Petits Ruminants, or PPR, is a viral plague of sheep and goats. Caused by a morbillivirus, it is a highly pathogenic and contagious disease. Since its initial discovery in Cote d'Ivoire in 1942, PPR has spread across most of Africa, Middle East, South, Central and East Asia, with more than 70 countries currently infected. It was identified in Europe (Bulgaria) in 2018 showing that PPR has no respect for European borders. Although domestic sheep and goats are the most important species affected, the virus can infect wild ungulates and camels – in some cases causing severe disease.

The disease is devastating. In a naïve flock or herd, it can kill up to 70% of animals with goats and youngstock being more susceptible. The animals exhibit high fevers, diarrhoea and dehydration, oral and nasal erosions, and pneumonia with very high case fatality rates. The virus is spread through animal-to-animal contact either mixing or through a timeline (pastures, vehicles contaminated with virus). Animal movement is one of the most important ways PPR can spread.

In 2015, FAO and WOAH agreed to target PPR for global eradication by 2030. In 2011, rinderpest was successfully eradicated, and – given how critical small ruminants and their products are as a source of income and food for subsistence farmers and more specifically women – eradicating PPR similarly would greatly address many of the UN Sustainable Development Goals. Eradication for each country follows a stepwise plan: Stage 1 (Assessment) where the country identifies its status through surveillance and diagnostic testing of potential PPR outbreaks; Stage 2 (Control) where animal populations are vaccinated – not just in response to outbreaks but also strategic and targeted vaccination designed to protect high-risk populations and stop the spread of PPR through achieving sufficient herd-level immunity. This immunity must be confirmed using post-vaccination monitoring; Stage 3 (Eradication) where surveillance detects the last of PPR and where vaccination ceases. Monitoring for evidence of disease and strong biosecurity measures must continue – particularly where animal movement from PPR infected countries / regions may reintroduce PPR; Stage 4 (Post Eradication), where surveillance and biosecurity continues and all vaccination has ceased as the country prepares to apply for WOAH free-status – the final step.

The Global PPR Eradication programme has moved from Phase 1 to Phase 2, with an updated blueprint for eradication. More emphasis will be placed on identifying and eliminating episystems of infection. Molecular epidemiology will allow for better identification where the virus is originating that feeds those episystems. While PPR eradication is the focus of the programme, control of other high impact (e.g., gastrointestinal parasitism, bluetongue, FMD) and zoonotic diseases (e.g., brucellosis, Rift Valley fever) are also targeted through improving animal health services. This is done throughout the value chain from supplier of veterinary products and supplies, to training of community animal health workers and education of farmers – in particular the most common care givers – women and children. It includes support for high-quality diagnostic laboratory services – both in-country and





regional and vaccine manufacture – including thermotolerant vaccines. It entails Private-Public-Community partnerships.

There are several aspects of PPR that make it an ideal candidate for eradication. As with rinderpest, vaccination once with an attenuated vaccine confers life-long immunity to the animal. Although there are 4 lineages of PPRV, the vaccine appears to be equally effective against all. There is also no carrier-state with PPR; animals once recovered do not shed the virus. While wildlife may contract PPR, there is good evidence that they become infected as a result of spillover from domestic sheep and goats and are not reservoirs themselves.

However, there are many challenges to eradication – most are related to lack of resources (money). The cost of the vaccine itself is relatively inexpensive but the cost of administering the vaccine, collecting samples for surveillance, having suitable diagnostic capacity to properly analyse those samples, combined with all the other administrative aspects – can be a sizable financial burden for some countries. While these are significant barriers, the benefit to that country – in terms of improved food and financial security for the poorest and needlest of their population, will quickly pay back that investment once eradication has been achieved.

The year 2030 is not far away in terms of achieving PPR eradication, but there is a unified will and a cogent plan to meet this goal. The biggest challenge right now is to muster sufficient resources to accomplish eradication. Failure to do so would result in enormous harm to the small ruminant populations of over 70 countries as well as Europe, and to the people who rely on them for their livelihoods.





#### **Professor Jarosław Kaba**

Faculty of Veterinary Medicine, Warsaw University of Life Sciences, Poland

Prof. Jarosław Kaba, Dipl.ECSRHM is a graduate of the Warsaw University of Life Sciences. He graduated from both the Faculty of Animal Breeding and the Faculty of Veterinary Medicine. Since 1992, he has been a research and teaching employee at the Faculty of Veterinary Medicine at the Warsaw University of Life Sciences dealing mostly with clinical and scientific issues of veterinary epidemiology as well as goat diseases. He is the vice-president of the Polish Society of Veterinary Sciences and the vice-president of the European College of Small Ruminant Health Management.



# Keynote Address: Caprine arthritis-encephalitis, caseous lymphadenitis and paratuberculosis in the goat population of Poland

As we have investigated the occurrence of caprine arthritis-encephalitis (CAE), caseous lymphadenitis (CLA) and paratuberculosis (PTB) in Poland for many years, we managed to trace their spread in the goat population of Poland.

The first report of CAE in Poland dates back to serological screening of three goat herds located in the western Poland in 1988–1991. CAE was likely introduced to the Polish goat population in 1980s with goats imported from abroad, perhaps from the Czech Republic, Germany, France, Belgium, and the Netherlands. The first large-scale cross-sectional serological study, conducted in 1996, revealed the presence of SRLV infection in 26% of herds. Two other studies carried out at 5-year intervals revealed a considerable increase of the herd-level seroprevalence to 66% in 2002, and 72% in 2007. A large-scale study was also carried out in 2014-2022. The true herd-level seroprevalence was estimated at 61% (95% confidence interval: 53%–68%).

CLA had been recognized in sheep and goats in Poland long before the first serosurvey carried out in 1996 which confirmed the presence of the disease in 13% of herds. The study conducted in 2002 revealed the presence of infection already in 63% of herds which corresponded to approximately a 5-fold increase. The large-scale study carried out in 2014–2021 confirmed the presence of specific antibodies in 73% (95% credibility interval: 65%–80%) of herds.

There is little data on the occurrence of PTB in the goat population in Poland. The first serosurvey was carried out in 1996. As few as 16% of goat herds tested positive. The next large-scale serosurvey was carried out in 2014–2021. The herd-level prevalence of PTB was estimated at 43% (95% credibility interval: 26%–58%).

No CAE, CLA or PTB control program is in effect in the goat population of Poland. In such a situation, CAE and CLA have spread throughout the population and are now a common. PTB prevalence is much lower. However, given the lack of preventive measures, PTB is expected to spread similarly to the first two diseases soon.





#### **Dr Ciriaco Ligios**

Istituto Zooprofilattico Sperimentale della Sardegna, Italy

I work at the Istituto Zoprofilattico Sperimentale of Sardinia (Italy), a veterinary public health institution that provides technical and scientific services within the framework of national and regional legislation. I am responsible for the histopathology, virology, and bacteriology of animal diseases within the long-term strategic health plan for zoonoses, diffuse viral diseases and prion diseases.

I create my scientific career with studies on prion diseases in small ruminants and emerging threats to animals from arthropod-borne viruses, in particular bluetongue virus, constantly pursuing the One Health aim.



#### Keynote Address: Can scrapie be eradicated? A real experience on an island

Scrapie first appeared in Sardinia (Italy) in 1996. In the following two decades, scrapie has spread throughout the Island with more than 60 outbreaks being notified in 2011. Due to the economic importance of the Sardinian sheep and caprine breeding sector, which has an annual balance sheet of more 500 million euros, this disease has caused great concerns.

The numerous scientific gaps regarding the genetics, epidemiology and pathogenesis of this disease were a further cause for concern at the time, as adequate knowledge is required to develop control strategies for diseases.

This scenario prompted us to organize in vivo experiments to gain better knowledge of the epidemiological and pathological characteristics of scrapie infection in our regional sheep and goat population, including the role of the different polymorphisms of the prion protein gene in modulating scrapie occurrence, the main routes of infection, the strain profile of the circulating prion agent, and the feasibility of breeding selection plans to eradicate the disease.

Among others, we found that the ARQ/ARQ genotype is the most susceptible in Sarda breed sheep, while the ARR/ARR genotype is associated with scrapie resistance. Interestingly, the presence of Valine in 136 position of the prion gene is very rare. In the same breed, the N176K and M137T dimorphisms of the PRNP gene significantly reduce susceptibility to classical scrapie in ARQ/ARQ genotypes. With regard to genetics in goat scrapie, we confirmed that the K222 mutation protects goats even against ovine scrapie isolates, and, for the first time, a possible association of D145 mutation with scrapie resistance. In contrast to the reports on the Valin sheep breed, our studies have shown that in Sardinia, only sheep co-infected with scrapie and Maedi-Visna mastitis transmit infectious prions through milk. Based on this new genetic knowledge, a compulsory selection breeding plan was applied in 2009 in Sardinia, thus allowing a significative decrease of the scrapie incidence in our Island.





#### **Professor Antonio D'Angelo**

University of Turin, Italy

2007 has been awarded the Diploma of the European College of Bovine Health Management (Dip. ECBHM).

2008 has been awarded the Diploma of the European College of Veterinary Neurology (Dip. ECVN).

2017 Full Professor, Department of Veterinary Science – Clinical Section, University of Turin.

2018 – present, Vice Dean, Department of Veterinary Science - University of Turin.

2018 – present, Chairperson of the Research Committee of the Department of Veterinary Science - University of Turin.

Invited speaker at many national/international congresses, seminars and educative courses. Author of more than 100 scientific publications [79 on international peer - reviewed Journals with IF (ISI)].



The neurological examination of small ruminants is fundamentally similar to that of small animals. A thorough neurological examination enables precise neuroanatomical localization and the establishment of a differential diagnosis list. The use of an algorithm helps in reaching these goals.

Evaluating postural and proprioceptive reactions confirms the existence of a neurological problem. Gait evaluation often supplements this information. Once a neurological problem is identified, evaluating spinal reflexes determines if the problem is due to a peripheral nervous system (PNS) or a central nervous system (CNS) lesion.

Generalized spinal hypo/areflexia, the presence of tremors in all four limbs, reluctance to move, or flaccid tetraparesis indicates PNS involvement. If a PNS issue is ruled out, altered mental status, behavior, cranial nerve deficits, or seizures indicate a CNS lesion cranial to the foramen magnum. In the absence of these deficits, the lesion is caudal to the foramen magnum in the spinal cord.

Spinal localizations are identified through spinal reflexes evaluation, in those animals that are presented in lateral recumbency. The presence of neurogenic/disuse atrophy and decreased/normal-increased tonus help distinguishing Low Motor Neuron from Upper Motor Neuron Localizations.

When a problem cranial to the foramen magnum (in the brain) is suspected, a more precise localization has to be achieved. A forebrain lesion will be characterized by normal or slightly altered mental status and gait; abnormal behaviour; seizures; fine tremors and pleurothotonus; abnormal postural reaction/proprioception and decreased/absent menace reaction (on the opposite side to the lesion). A brainstem localization will show an abnormal mental status and gait (hemiparesis/tetraparesis); normal behaviour; abnormal postural reaction/proprioception (on the same side to the lesion); multiple cranial nerves deficits. A cerebellar localization will display normal mental status and behaviour; wide base stance and intentional tremors; abnormal gait (cerebellar ataxia); normal or hyper postural reaction/proprioception; normal cranial nerves with the exception of menace reaction (but the animal is able to see). Finally, a central vestibular localization will be characterized by abnormal mental status; head tilt and wide base stance; vestibular ataxia and hemiparesis (on the same side to the lesion); abnormal cranial nerves (besides those with vestibular function). Normal mental status, absence of hemiparesis and normal cranial nerves (besides those with vestibular function and eventually facial nerve), make it possible to distinguish between a central and a peripheral vestibular syndrome.







Once a neuroanatomical localization is reached, it is crucial to establish a list of differential diagnosis. In this context, using the acronym VITAMIN D (Vascular; Inflammatory; Traumatic; Anomaly; Metabolic/Toxic; Idiopathic; Neoplastic; Degenerative) can be helpful to categorize the patient's history and clinical signs (neuroanatomical localization) into one or more these categories, in descending order of importance.

The most common VITAMIN D categories in the author's field experience are:

#### **INFLAMMATORY:**

Bacterial: the most frequent bacterial inflammatory diseases are Meningitis and Encephalitis, Otitis Media-Media Interna, Listeriosis and *Clostridium perfringens* (mainly types C and D) are the most frequent bacterial inflammatory diseases. Other causes include Brain abscess and Vertebral Suppurative Osteomyelitis-Discospondylitis.

Viral: Lentivirus Encephalitis and more rarely Pseudorabies and Rabies can be found.

Parasitic: Coenurosis (gid) is the most common parasitic inflammatory disease reported, in specific geographic areas.

TRAUMATIC: the most common traumatic diseases include Head Trauma, Spinal cord trauma often associated to Vertebral injury and injury to peripheral nerves.

ANOMALY: Developmental dysfunctions may result from hereditary, environmental, or infectious disorders. The most frequent conditions are Hydrocephalus, Hydranencephaly, Cerebellar Hypoplasia and abiotrophy. Common causes of uterine infections leading to these anomalies are Blue Tongue Virus, Border Disease Virus, BVD virus, Akabane Virus and Schmallenberg Virus.

METABOLIC/NUTRITIONAL/TOXIC: conditions in this category include Polioencephalomalacia or cerebrocortical necrosis, Hepatic Encephalopathy, Hypomagnesaemia, Hypocalcaemia, Copper deficiency (Enzootic swayback), Pregnancy Toxemia, Tetanus and Floppy Kids syndrome. Among intoxications, Lead poisoning, Sodium salt poisoning, Water intoxication and Toxic plants are the most common.

DEGENERATIVE: Scrapie is the most common degenerative disease in small ruminants.





### **Oral presentation abstracts**

#### Oral presentation 1: Treatment guidelines for BTV3 (Margit Groenevelt)

Diergeneeskundig Centrum Zuid-Oost Drenthe, The Netherlands

Co-authors: Fiona Lovatt, Rachael Tarlinton

During the outbreak in 2023 in The Netherlands it quickly became apparent that the BTV-3 strain that was circulating was highly pathogenic in sheep. Flock morbidity and mortality varied enormously (0-100%), mainly depending on the proximity of the flock to the centre of the outbreak and the exact date of first clinical signs. The closer to the centre of the outbreak and the earlier in the year affected mostly meant higher levels of both morbidity and mortality. However, individual case mortality rate was estimated to be between 50-70% for any sheep that showed clinical signs. After a few weeks it also became apparent that surviving animals often developed secondary problems and as such long-term prognosis of sheep was very poor. Therefore, during the later months of the outbreak early euthanasia rather than treatment was increasingly considered and applied on welfare grounds.

However, individual treatment should still be considered and discussed. Unfortunately, there are limited reports of appropriate treatment for BTV, although a single clinical case series of 35 hospitalised sheep in the USA (with an 83% survival rate) summarises the types of treatment regime that have been tried, and treatment guidelines used in The Netherlands are comparable.

Basic husbandry and nursing care should be as for any downer cow or sheep with easily available water and multiple feed options, soft non-slip bedding with the expectation to regularly lift and turn recumbent patients, whilst carefully checking for signs of bed-sores or hoof sloughing. Sheep being nursed will need to have close contact with companions to avoid further unnecessary distress.

The mainstay of therapy for clinical cases in both the USA study and in The Netherlands in the current BTV3 outbreaks are:

- Anti-inflammatories and analgesics to decrease pain, fever and immune mediated damage to the vascular endothelium see details below.
- Supportive fluid therapy (both orally or intravenous fluids with variety of crystalloid fluids) to maintain hydration and blood volume.
- Thiamine or multiplex B vitamin supplementation may be helpful to support appetite and prevent polioencephalomalacia.
- Antimicrobial cover (penicillin or oxytetracycline) for secondary infections caused by complications
  attributed to the hematogenous spread of bacteria through the dysfunction of the normal gut lining causing
  septicaemia or septic arthritis.

Early in the Dutch outbreak, farmers and vets also tried using antihistamine preparations to treat clinical cases; however, this stopped as soon as it became clear that there was no benefit from this treatment.

In both the USA series of cases and initially in The Netherlands, most animals received non-steroidal anti-inflammatory drugs (NSAIDs), such as flunixin meglumine or meloxicam at an initial dose of meloxicam 1mg/kgBW (1ml/20kg), followed by a lower once daily dose of 0.5mg/kgBW (1ml/40kg). Meloxicam treatment for cattle was suggested to be 0.5mg/ml for the first day, followed by 0.25mg/ml on subsequent days and in camelids, 1mg/kg was advised every other day due to its longer half-life. However, postmortem examinations of sheep in The Netherlands revealed severe tubular degeneration in the kidneys as a common phenomenon even in early cases or in animals that had died within 24 hours. The use of NSAIDs is contraindicated in animals with severely decreased kidney





function or in a state of dehydration or both. It was considered most likely that all BTV-3 cases have some decreased kidney function and the vast majority will be dehydrated. For these reasons, advice in The Netherlands has changed to emphasise the need for fluids, to suggest reduced dosages of NSAIDs and often to advise the use of oral paracetamol instead.

Although there are limited reports of the pharmacokinetics and effectiveness of the analgesic effects of paracetamol in sheep, and a risk of hepatotoxicity if prolonged treatment; there are reports of its use in sheep and goats in the literature. In The Netherlands, the dose of 10-20mg/kg three times daily was advised for sheep.

If this authorised paracetamol is not available in sufficient quantities to meet demand, then practitioners may consider prescribing human paracetamol under the cascade. This was necessary in The Netherlands, with advice suggesting a 500mg tablet should be crushed and administered three times a day per sheep. Note that access to the human preparation would be under veterinary prescription.

Treatment with dexamethasone (0.06mg/kg once daily for up to 10 days) early in clinical disease for severely affected animals, such as those with high temperature, swollen heads and respiratory distress, could also be considered. The rationale behind this is the same principle as the use of dexamethasone (6mg daily for up to 10 days) in humans with severe COVID-19, reducing the severe 'cytokine storm' and immune mediated damage in severe COVID-19 pneumonia cases. However, it is important to note that dexamethasone will also induce parturition or abortion in pregnant ruminants (at a dose of 8mg in ewes). Additionally, there have been no veterinary controlled trials to examine its efficacy in BTV treatment and prolonged corticosteroid treatments are commonly contraindicated for viral infections, so its use should be approached with caution.

Supportive therapy should focus on providing easily accessible and good quality food and water. In The Netherlands, most farmers used a 'buffet' for the sick animals with multiple choices. Most foodstuffs included were silage, hay, fresh grass, muesli, soaked concentrates, willow and even apple sauce. Although oral fluid supplementation was used, due to the damage seen in the oesophagus it was considered to also be damaging to drench sick animals multiple times a day. Low volume drench fluids such as propylenglycol were used more often.

Although a lot of different treatment regimens were used, tried, advised and discarded, no single treatment seemed to have been most successful. Due to the severity of the disease and the primary damage it does to the sheep, the therapies described above will only get you so far. Care needs to be taken to keep assessing the prognosis of the individual animal and to always keep euthanasia an option.





#### Oral presentation 2: Use of an autologous vaccine against bluetongue virus (Martin Ganter)

Clinic für Swine and Small Ruminants, University of Veterinary Medicine Hannover, Germany

Co-authors: A. Zabel, N. Ossowski

The German Standing Committee on Veterinary Vaccines (StIKo-Vet) has the opinion that the production and use of autologous vaccines against Bluetongue virus 3 is essential due to the lack of licensed vaccines. As there were in spring 2024 no ad hoc approved vaccines for the current BTV 3 variant in Germany, an autologous vaccine was considered to fight the anticipated epidemic on the basis of Article 2 (3) of Regulation (EU) 2019/6 for "the treatment of one or more animals in a herd with a confirmed epidemiological link" for the herds in the BTV 3 positive federal states.

A confirmed epidemiological link was postulated on the basis of the:

- 1. genetically largely identical virus isolates from the outbreaks in the Netherlands, North Rhine-Westphalia (NRW), and Lower Saxony (LS), and
- 2. the unlimited spread of the mosquitoes during the main flight period.

The manufacturers of autologous vaccines in Germany were asked to produce a vaccine based on the existing isolates from NRW and LS. In the end, only SAN Group Biotech Germany GmbH showed interest and willingness to produce such a vaccine.

As a result, the Ministries for Agriculture in NRW and LS authorised the use of the autogenous vaccine ANIVAC BTV3 produced by SAN Group Biotech Germany GmbH.

The autogenous BTV 3 vaccine was used for the first time on 5 April 2024 in 61 animals in our Clinic. From 8 April on 1534 animals (1266 sheep; 89 goats, 175 cattle, and 4 alpacas) were vaccinated in 11 herds by us. In the clinic the first sheep showed elevated body temperature at 12 April followed by 6 other sheep until 21 April. No clinical side effects were seen in the 61 animals. On 19 April, severe clinical diseases occurred in sheep flock "S" on pasture, and on 21 April in a second flock "U" housed indoor. They showed severe clinical signs with a high number of sheep with fever up to 41.5°C. The symptoms were apathy, oedema at the heads and lips, impaired sensitivity at the lips, salivation, ulcers in the mouth, laminitis, uveitis, and necrosis at the teats. In a group of cows the average milk yield decreased from 37.2 Liters at day 6 post vaccination to 28 L at day 16 p.vac. Nearly normal average milk yield was reached after one month p.vac. In sheep, it was obvious that mothers of twins and multiples were the most clinically affected. Until the end of May from the 1595 vaccinated animals 291 were clinically affected, 14 died, and 11 were euthanized due to animal welfare reasons. From the 190 animals tested by PCR for BTV 3 a total of 73 reacted positive.





Oral presentation 4: Retrospective analysis of submissions to the world reference laboratory for foot-and-mouth disease: what can these data tell us about the role of small ruminants in disease epidemiology? (Rheinallt Jones)

School of Biodiversity, One Health and Veterinary Medicine, College of Medical, Veterinary and Life Sciences. University of Glasgow, UK

Co-Authors: Donald P. King, Valentina Busin

Introduction. Epidemiological studies can be useful to understand the spatiotemporal dynamics of foot-and-mouth disease virus (FMDV). Data collected by FMD Reference Laboratories provide an incomplete picture and are often biased towards large ruminants testing due to their high value and overt clinical presentation of disease. Studies on small ruminants, are limited, which may lead to an under-appreciation of their epidemiological importance in the spread (and therefore control) of FMD. To elucidate the spatiotemporal and species distribution, data from samples submitted between 1958 and 2022 to the FAO World Reference Laboratory for Foot-and-Mouth Disease (WRLFMD) were analyzed. Analysis concentrated on defining the contribution of small ruminants to these laboratory data, with the aim of improving our understand of their contribution to monitoring strategies and inform future approaches of their inclusion in monitoring and control strategies.

Material and methods. Analysis and visualization were made using R version 4.2.0. Categorical variables analysed were: i) Family; ii) Genus; iii) Group (Large Ruminant, Small Ruminant, Pigs, Wildlife or Not defined); iv) Test result (Positive, Negative, Not defined); v) FMDV Serotype; vi) Continent; vii) Country. Summary count data were presented as median counts with inter quartile ranges (IQR). Summary proportional data were presented as a mean value with standard deviation. A one-way chi square test was used to assess differences between factor occurrence within a categorical variable.

Results. The total number of samples included in the analysis was 32802. The median number of samples collected was 444/year (IQR: 259). When samples were classified into domesticated species groups, the most represented were Large Ruminant (n = 15021), followed by Small Ruminant (n = 1972), Pigs (n = 1486) and Wildlife (n = 294). Within the domesticated groups, 73.0% of Pigs and 72.2% of Large Ruminant samples were FMDV positive. Whilst Small Ruminant samples had significantly fewer FMDV positive samples, at only 30.0% (p < 0.0001). All 7 FMDV serotypes were identified from Large Ruminants samples, 5 serotypes (A, Asia 1, O, SAT1 and SAT2) from Small Ruminant samples and all but serotype SAT3 were identified for Pig samples. Samples were submitted from a total of 144 countries. Across all years, Africa and Asia submitted most of the samples. Nepal (n = 1065), Islamic republic of Iran (n = 974), and Pakistan (n = 949) were the countries with the highest Large Ruminant submissions. Whereas, Cyprus (n = 268), Ireland (n = 221) and Saudi Arabia (n = 186) submitted the most Small Ruminant submissions. Finally, the highest Pigs submissions were from Hong Kong SAR (n = 522), Viet Nam (n = 190) and the Philippines (n = 168).

Discussion and conclusions. A few interesting insights have emerged from these analyses, where there have been regular submission of approximately 500 samples a year, mainly from Africa and Asia. As expected, Large Ruminants provided the highest number of submissions, however Small Ruminant represented the second highest number. Regarding the outcome of testing, it was interesting to note that a high proportion of samples were negative, especially from Small Ruminants. Based on this analysis, the yearly number of samples submitted to the WRLFMD might need to be revisited, to target FMDV serotype pools and high-risk countries. Species allocation should also be considered. Finally, the WRLFMD role in for confirmatory testing should be considered especially as regards to the high negative Small Ruminant submissions.





#### Oral presentation 5: Natural and experimental EHDV-8 infection in Sarda sheep (Davide Pintus)

Istituto Zooprofilattico Sperimentale della Sardegna, Sassari, Italy

Co-Authors: Elisabetta Coradduzza, Maria Giovanna Cancedda, Massimo Spedicato, Angela Maria Rocchigiani, Roberto Bechere, Simona Macciocu, Annalisa Oggiano, Simone Pulsoni, Paola Madrau, Giulia Franzoni, Ottavio Portanti, Angelo Ruiu, Barbara Bonfini, Giovanni Savini, Ciriaco Ligios, Giantonella Puggioni.

Epizootic hemorrhagic disease (EHD) is a vector-borne viral disease of ruminants transmitted by *Culicoides* spp. and caused by the epizootic haemorrhagic disease virus (EHDV). EHDV belongs to the *Orbivirus* genus, *Sedoreoviridae* family. To date seven serotypes have been officially recognized.

Clinical signs of EHD have historically been observed in deer and less frequently in cattle, while the susceptibility and the role of sheep in transmitting EHDV is still under debate.

In October 2022, EHDV-8 affected 3 cattle farms in the southern part of Sardinia (Italy), this was the first evidence of EHDV in Europe. In early November 2022, blood samples were taken from animals belonging to farms epidemiologically linked to the EHD outbreak, to assess the presence of antibodies and viral RNA. Surprisingly, within a flock of 125 healthy sheep, out of 85 animals sampled 74 (87.1%) tested positive for EHDV antibodies, and 73 (85.1%) tested positive for EHDV RNA. After 18 months, 55% of the sampled animals of the flock still had antibodies against EHDV-8, with titre ranging from 1:10 to 1:640.

Given these results, to understand the epidemiological role of sheep and the dynamics of EHDV-8 infection, three Sarda breed sheep were inoculated intravenously with naturally EHDV-8 infected bovine blood. These sheep underwent virologic and serologic testing before being sacrificed 11 days post-infection. No clinical signs were observed in any of the three sheep during the trial. However, EHDV-8 RNA was detected in all three sheep by real time RT-PCR assay from the day 3 to day 11 post-infection and the virus was isolated in cells culture from day 3 to day 9 post-infection. Antibodies against EHDV-8 were detected in only one sheep at 10 days post-infection. At the necroscopy, EHDV-8 RNA was found in several organs of all sheep, especially in spleen and lungs, where abundant viral mRNA was detected by in situ hybridization. These results suggest that sheep may play an effective role in the spread of EHDV-8 among susceptible ruminant species.

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#### Oral presentation 6: Tick-borne infections in small ruminants – the zoonotic potential (Snorre Stuen)

University of Life Sciences, Faculty of Veterinary Medicine, Department of Production Animal Clinical Sciences, Norway

Ticks and tick-borne diseases occur worldwide. Millions of ticks are spread annually by migrating birds and movement of mammalians between countries and continents, making the possibility for tick-borne infections to be established in new geographical areas. The ongoing climatic changes may have a huge effect on their distribution.

The effects of ticks and tick-borne diseases are a barrier to development in the livestock industry in different parts of the world due to high economic losses caused by disease outbreaks, mortalities, damage of hides and poor production. The most important tick-borne pathogens associated with small ruminants are within the genera *Anaplasma*, *Babesia*, *Ehrlichia*, *Theileria*, *Flavivirus* and *Nairovirus* causing variable clinical signs, from subclinical, acute, fatal to chronic wasting conditions.

In this context, several tick-borne infections affecting small ruminants have a zoonotic potential, especially pathogens in the bacterial genera *Anaplasma / Ehrlichia*, but also viral diseases such as Louping ill (LI), Nairobi sheep disease (NSD) and Crimean Congo haemorrhagic fever (CCHF) may cause challenges in man. NSD-virus generates a serious infection with high mortality in small ruminants. Few human cases have been reported, but the virus is transmitted by tick species having a potential for a world-wide distribution. In contrast, CCHF-virus, transmitted mainly by *Hyalomma* ticks, causes only subclinical infection with transient viraemia in small ruminants, but may give rise to severe infection with high fatality rates in human.





# Oral presentation 7: Epidemiological study of *Leishmania* seroprevalence in sheep population of Aragón, Spain (Pablo Jose Quílez)

Animal Pathology Department, Instituto Agroalimentario de Aragón, Universidad de Zaragoza, Spain

Co-Authors: Héctor Ruiz, Delia Lacasta, Teresa Navarro, Diana Marteles, Aurora Ortín, Marta Ruiz de Arcaute, Juan José Ramos, Antonio Fernández, Sergio Villanueva-Saz

Leishmaniosis, a parasitic disease caused by protozoa of the genus *Leishmania*, is a significant health concern in various mammalian hosts. Dogs are the main reservoir for human infections. In Europe, *Leishmania infantum* is the main responsible *Leishmania* species causing the infection. Recent evidences suggest that it is possible to detect seropositive sheep in endemic areas with *L. infantum* infection. Given the absence of epidemiological studies on sheep infection in the Aragon Autonomous Community, the aim of this study was to determine the seroprevalence of Leishmaniosis in a population of sheep from Aragon Region (Spain) including all representative areas of the region.

Based on an expected seroprevalence of 9.27% (the sheep seroprevalence in Spain), an accepted 5% deviation from the true prevalence and a confidence level of 95%, the sample size necessary to estimate the seroprevalence was calculated to be 130 animals. A total of 727 sheep were sampled from October 2022 to April 2024. The samples were tested for seropositivity using enzyme-linked immunosorbent assay (ELISA).

Out of these, 20 animals tested positive for *Leishmania* antibodies, indicating a seroprevalence rate of approximately 2.75%. Most of the seropositive samples (17/20) were detected during sandfly period activity (from March to October).

The presence of *Leishmania* antibodies in sheep suggests a possible exposure to the parasite, which could have significant implications for animal health and productivity. The relatively low seroprevalence rate observed in this study could be indicative of sporadic transmission or a well-contained infection within the population. Nevertheless, it highlights the need for further investigation into the potential reservoirs and vectors of *Leishmania* in the region, as well as the environmental and management factors contributing to the exposure of sheep to this parasite.

Given that leishmaniosis is typically associated with dogs and humans, the detection of seropositive sheep emphasizes the importance of a One Health approach to disease surveillance and control. Understanding the epidemiology of Leishmaniosis in sheep can provide insights into the broader dynamics of the disease, including cross-species transmission and the role of wildlife and livestock in the maintenance of the parasite. Further studies should aim to elucidate the transmission dynamics, identify potential risk factors, and develop effective control strategies to address this emerging health issue in sheep populations.





#### Oral presentation 8: All ruminant lentiviruses genetic diversity in Italy: a 2019-2022 Snapshot (Paola Gobbi)

Istituto Zooprofilattico Sperimentale dell'Umbria e delle Marche (IZSUM), Italy

Co-Authors: Luca Tassoni, Elena Tinelli, Eleonora Scoccia, Mario Orrico, Cecilia Righi, Sara Mrabet, Carmen Maresca, Monica Giammarioli, Francesco Feliziani, Maria Serena Beato

Small Ruminant Lentiviruses (SRLVs) are retroviruses that infect goats and sheep, causing extremely diversified clinical manifestations. They include VISNA-MAEDI and CAEV and are characterised by a great genetic variability. SRLVs are worldwide distributed. Control and eradication measures are not mandatory in the majority of countries, including Italy. SRLVs are classified into five genotypes (A-E) and several sub-genotypes (A1-24; B1-5; E1-2). Full understanding of SRLV epidemiology is still a matter of research, and genetic characterization remains a powerful tool to understand the complexity of such infection in small ruminants. A previous study, surveying the genetic landscape of SRLV in Italy, suggested a high genetic variability according to the geographic area and identified potential new sub-genotypes; three genotypes and fourteen sub-genotypes were identified so far in Italy. The present study aims to update the genetic characterization and phylogenetic analysis of SRLV collected from 16 out of 21 Italian regions, between 2019 and 2022. During this period, 213 sheep and goat farms were subjected to SRLV diagnosis, of which 69 were positive and 144 negatives. In 2019 55/89 farms resulted positive, in 2020 35/50, in 2021 33/42, in 2022 27/40. A total of 363 samples were tested, of which 112 resulted positive. The gag-pol conserved gene fragment was the target of the nested PCR test, generating an amplicon of about 800 bp. Proviral DNA or RNA extraction was performed from a variety of matrices, including somatic cells, buffy coat, joint fluids, and organ homogenates. Maximum Likelihood (ML) phylogenetic analysis was conducted on gag-pol sequences of 68 samples from the period 01.10.2019-31.12.2022 and three samples from 2023, totaling 71 sequenced samples. A high genetic diversity was identified during the study period, confirming previously described data for SRLV in Italy, with a higher prevalence of B genotype in both sheep and goat farms. We confirm the presence of subgenotypes B1, B2 and B3, as well as subgenotypes A9, A11, A19, A20 and A24. At the same time, we identified ambiguities in the observed phylogenetic tree topology for A24 and A9 subgenotypes, which could call into question previous classifications. We also identified some samples classified as A9 that were previously identified as A19. Further and more in-depth studies are needed, such as sequencing of the complete gag gene and full genome with the aim to monitor the genetic SRLV diversity and support development of proper diagnostic methods.





# Oral presentation 9: Herd-level seroprevalence of SRLV infection in Carpathian goat population of Romania (Adrian Valentin Potarniche)

University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, Romania

Co-Authors: C. Cerbu, D. Olah, E. Ungureanu, M. Czopowicz, M. Mickiewicz, A. Moroz-Fik, K. Biernacka, D. Nadolu, A.H. Anghel, J. Kaba

Background. Caprine arthritis-encephalitis (CAE) is a contagious, life-long, and progressive disease of goats caused by a small ruminant lentivirus (SRLV). CAE constitutes a substantial economic problem, mainly in dairy goat herds due to its negative impact on animal welfare and productivity as well as the quality of milk. CAE monitoring programs are generally based on serological testing since the detection of antibodies is an accurate indicator of the infection. However, no surveillance programs, either voluntary or mandatory, are in place in Romania so the epidemiological situation of SRLV infection in this country remains unknown. Control programs based on blood sampling are laborious and expensive, however bulk-tank milk (BTM) testing offers a quick, convenient, and stress-free alternative for monitoring the epidemiological situation at the herd-level. Therefore, in this study, we tried to determine the herd-level seroprevalence of SRLV infection in the Carpathian goat population of Romania using BTM.

Materials and methods. The study was conducted between 2022 and 2023. In total, 277 BTM samples were randomly collected from herds located in all four macro-regions of Romania: Ro1 – 48 BTMs (17.3% of 277), Ro2 – 115 (41.5%), Ro3 – 86 (31.0%) and Ro4 – 28 (10.1%). The size of tested herds ranged from 11 to 691 adult animals with the median (interquartile range) of 158 (99–226). BTM samples were collected right after morning milking into Falcon tubes (50 ml), directly from the tank and tested using a commercial ELISA kit (ID Screen® MVV/CAEV Indirect, IDVet – Innovative Diagnostics, Grabels, France) following the manufacturer's instructions. The herds were classified as seropositive if the optical density of BTM (ODBTM) was more than the cut-off value of 0.15, and the within-herd seroprevalence was considered >50% if ODBTM was >3.0.

Results. Based on the criteria mentioned above 140 herds tested positive which corresponded to the herd-level seroprevalence of 50.5% (95% confidence interval from 44.7% to 56.4%). Only two goat herds appeared to have the within-herd seroprevalence >50%. Seropositive herds were significantly larger than seronegative herds (p=0.003), with the median (interquartile range) number of adult goats of 173 (111–256) and 124 (90–206), respectively.

Conclusions. Our findings showed that the SRLV infection is widespread in the Carpathian goat population in Romania. BTM testing appears to be a convenient method for crude evaluation of CAE herd-level prevalence, especially in countries like Romania where the epidemiological situation of the disease is unknown and control programs are missing.





#### Oral presentation 10: Epidemiological study on the prevalence of Anaplasma ovis in Aragón, Spain (Héctor Ruiz)

Veterinary Faculty, University of Zaragoza, Spain

Co-Authors: Marta Ruiz de Arcaute, José María González, Pablo Quílez, Marina Pomar, Cristina Baselga, José Luis Arnal, Delia Lacasta

In recent decades, the emergence of vector-borne diseases in the ruminant livestock sector in the European Union has become particularly relevant. According to official data from the Ministry, Spain has 14,452,585 heads of sheep, with the Aragon region representing 10.31% (1.490.062) of the total. In 2014, the first clinical outbreak of ovine anaplasmosis was diagnosed in Aragon, severely affecting a significant number of flocks. Since then, multiple cases have been diagnosed by the Ruminant Clinical Service of the University of Zaragoza (SCRUM), which could be associated with both the expansion and colonisation of new areas by the biological vector of the disease, ticks of the Ixodidae family and the increased temperatures associated with climate change, a factor that has favoured the conditions of these vectors.

For this reason, taking advantage of the specific characteristics of the territory of the Autonomous Community of Aragon (47,697 km2), an epidemiological study was proposed to determine the real distribution and extent of *A. ovis* infection throughout the region. To achieve this, considering it is a vector-borne disease, the different climatic zones and key aspects that can determine the population of vectors were analysed. The territory was grouped into three zones according to their climatic characteristics previously described by the Aragon Climatic Atlas: zone 1 (steppe and continental Mediterranean dry climate); zone 2 (cold and warm continental sub-Mediterranean climate); and zone 3 (humid sub-Mediterranean climate and Mediterranean-continental transition climate). The ovine census present in each climatic zone was also considered. Finally, a total of 68 farms were selected, with censuses ranging from 35 to 10,000 sheep, and distributed by the three climatic zones defined previously.

All farms followed traditional semi-intensive or extensive grazing systems, allowing the flocks access to pasture for long periods and exposing them to the biological vectors. Once the farms were chosen, blood samples with EDTA (5ml) were taken from 10 adult animals (>2.5 years) randomly selected. Simultaneously, an epidemiological survey was conducted with the farmer to collect data such as census, breed, grazing areas, feeding, treatments, etc. Individualised samples from each animal were distributed in 1.5ml Eppendorf tubes and stored individually at -20°C. They were later analysed in pools of 5 animals (2 per farm) using a quantitative real-time PCR with the EXOone *Anaplasma ovis* kit (EXOPOL S.L.).

All farms except one were positive in both pools (67/68: 98.53%). The only negative farm was an educational farm located in the city of Zaragoza with less than 50 ewes, dedicated to environmental education, which always graze in the same fenced fields, with limiting access to wildlife. No clinical signs were detected in the flocks. No statistical differences were found between farms or climatic zones.

The results indicate the presence of *Anaplasma ovis* in practically all flocks, regardless of the analysed climatic zones. These data suggest a widespread distribution of the infection throughout the territory and contact with the vector.





# Oral presentation 11: Genetic types and subtypes of SRLV circulating in Polish, Lithuanian, and Hungarian goat populations (Michał Czopowicz)

Warsaw University of Life Sciences-SGGW, Poland

Co-Authors: Saulius Petkevičius, Laszlo Ózsvári, Giuseppe Bertoni, Carlos E. Abril, Snorre Stuen, Jarosław Kaba, Patricia Klibavičė, Krisztina Bárdos, Agata Moroz-Fik, Kinga Biernacka, Marcin Mickiewicz, Zofia Nowek, Iwona Markowska-Daniel, Emilia Bagnicka.

Introduction: Caprine arthritis-encephalitis (CAE) is caused by small ruminant lentivirus (SRLV). SRLV is classified into 5 genotypes (A-E) of which A and B are widespread. Genotypes A and B are subdivided into 27 and 5 genetic subtypes, respectively. There are limited data on SRLV genotypes and subtypes circulating in the central Europe.

Aim: To genetically characterize SRLV strains responsible for CAE in 3 central-European countries.

Materials and Methods: This multicenter study was carried out in years 2021–2022 in Poland, Lithuania, and Hungary. Goat herds seropositive for SRLV infection were selected: 20 herds in Poland, 10 in Lithuania, and 10 in Hungary. Blood was collected from seropositive goats in these herds, and DNA from leukocyte pellets was tested using a 2-stage real-time nested PCR (RT-nPCR) which distinguishes between genotype A and B. Then, the Sanger sequencing was performed using a 200 bp-long fragment of the Long Terminal Region (LTR) and the gag gene sequence (LTR-gag) located within the RT-nPCR target sequence.

Results: Both SRLV genotypes were identified in all the 3 countries.

In Poland, SRLV infection was confirmed in 20/20 herds. Genotype A was found in 18/20 herds (90%; CI 95%: 70%–97%) and SRLV strains were classified as A2, A2/3, and A19 subtypes. Genotype B was found in 8/20 herds (40%; CI 95%: 22%–61%) and SRLV strains were equally often B1 and B2 subtypes. Concomitant infection with genotypes A and B was detected in 6/20 herds (30%; CI 95%: 15%–52%).

In Lithuania, SRLV infection was confirmed in 9/10 herds: genotype A was found in 5/9 herds each (56%; CI 95%: 27%–81%) and SRLV strains were classified as A1 and A2. Genotype B was also found in 5/9 herds each (56%; CI 95%: 27%–81%) and all SRLV strains were classified as B1. A concomitant infection was detected in 1/9 herd (11%; CI 95%: 2%–44%).

In Hungary, SRLV infection was confirmed in 10/10 herds. Genotype A with was found in 7/10 herds (70%; CI 95%: 40%–89%) and SRLV strains were classified as A1, A2, A4, and A18. Genotype B was found in 7/10 herds (70%; CI 95%: 40%–89%) with predominating B1 subtype. Concomitant infection was detected in 4/10 herds (40%; CI 95%: 17%–69%).

Conclusion: Both SRLV genotypes and several subtypes are responsible for CAE in Polish, Lithuanian, and Hungarian goat populations.

Funding: The study was financed by the grant no. ICRAD/I/CAE-RAPID/02/2021.





# Oral presentation 12: *Streptococcus ovis* in small ruminants: an underestimated pathogen? (Sebastian Alessandro Mignacca)

Department of Agriculture, Food and the Marine - Pathology Division, Celbridge, Ireland

Co-Authors: Aine O'Doherty, Michael Finglas, Colm Brady, John Moriarty, Cosme Sánchez-Miguel

Introduction. Pathogenic streptococci can cause suppurative disease and sepsis in humans and livestock. In sheep, *Streptococcus ovis* is the aetiological agent of "sheep septicaemic streptococcosis", an acute, febrile and septic zoonosis.

S. ovis was described for the first time in 2001, and to date, little is known regarding its pathology.

This work aimed to describe the pathological findings in small ruminant carcasses with *S. ovis* infection through routine surveillance within the Department of Agriculture, Food and the Marine (DAFM) - State Veterinary Laboratory (Ireland) - from January 2018 to May 2024.

Materials and Methods. During small ruminant post-mortem examinations (PME) (12.661 sheep; 527 goats), selected tissues underwent bacteriological culture based on standard laboratory protocols. Suspected streptococci isolates were confirmed using MALDI-TOF MS (Bruker Daltronik GmbH, Bremen, Germany) according to the manufacturer's instructions. Streptococci strains confirmed as *Streptococcus* spp through routine cultures and API tests were excluded from this study. The *S. ovis* cases were reviewed.

Results. *S. ovis* was identified in 24 animals (23 sheep; 1 goat) belonging to both sexes, and, a part from a pair of ovine foetuses (twins), ages ranged between four weeks to five years of age, with an average of 1.9 years. These carcasses were submitted from 22 different flocks throughout Ireland. Eleven out of 24 cases (45.8%) demonstrated varying pulmonary lesions. In 4 cases (16.7%), *S. ovis* was isolated from subacute-chronic necrosuppurative (oro)-pharyngeal fistulae or phlegmons, likely consequential to dosing gun injuries, affecting surrounding tissues and eroding the main cervical blood vessels resulting in fatal haemorrhages. Another three ewes had additional focal pharyngeal abscessation. The gastro-enteric tract was involved in 3 cases where abomasitits and enteritis were described. In two cases, purulent ascites and abdominal abscessations were observed. *S. ovis* was diagnosed as the causative agent of abortion in twin ovine foetuses. One ewe had an associated purulent mastitis. Finally, in two instances, *S. ovis* was cultured from the liver, but the cause of death was not reached due to autolysis. Thirteen carcasses (54.2%) also demonstrated lesions consistent with sepsis, and 15 animals (62.5%) had concurrent disease (usually parasitism). Seven cases (29.2%) were observed in summer (August and September), of which six demonstrated pulmonary pathologies; all the other cases were observed from late autumn till early spring. Sporadically, other bacterial pathogens were also associated.

Discussion and Conclusion. Suppurative lesions were the more frequent finding. The respiratory system was the main affected; therefore, *S. ovis* should be considered where respiratory pathology is present. Although *S. ovis* could represent a primary pathogen in sheep, concurrent diseases (parasitism), environmental stressors (winter) or injuries (drenching or bolus dosing) could predispose to the disease or increase its pathogenicity. The higher summer cases of pneumonia could be associated with exposure to higher temperatures and frequent rain in recently sheared sheep. The single isolate in a goat could reflect the lower number of animals in this species in Ireland. Finally, the authors believe that *S. ovis* infection, alone or in association with other bacteria, and its effects may be underestimated.

Acknowledgements: Private veterinarian practitioners for providing information and assistance, and DAFM staff in Central and Regional Veterinary Laboratories for collecting data, undertaking post-mortem examinations, sample testing and reporting.





# Oral presentation 13: Q-Net-Assess: Improved molecular surveillance and assessment of host adaptation and virulence of *Coxiella burnetii* in Europe (René van den Brom)

Royal GD, Deventer, The Netherlands

Co-Authors: Tom McNeilly

Q fever is an almost ubiquitous zoonosis caused by *Coxiella burnetii*, which is able to infect several animal species, as well as humans. Cattle, sheep and goats are the primary animal reservoirs. In small ruminants, infections are mostly without clinical symptoms, however, abortions and stillbirths can occur, mainly during late pregnancy. Shedding of *C. burnetii* occurs in faeces, milk and, mostly, in placental membranes and birth fluids. During parturition of infected small ruminants, bacteria from birth products become aerosolised. Transmission to humans mainly happens through inhalation of contaminated aerosols.

Whole genome sequencing (WGS) has revolutionised molecular epidemiology and surveillance of many zoonotic pathogens. Our understanding of the extent to which Q fever manifestation depends on C. burnetii genotype is very limited. Due to variable and non-standardized methods, current typing provides little information. WGS offers an alternative to this, as it is easy to standardize and provides comprehensive genetic information. Currently, only a few full-genome sequences of Coxiellae have been published, most of which are limited to old laboratory isolates. This is due to the often-difficult isolation of the pathogens from field samples. Recently, a pan-European consortium, with experts in Q fever diagnostics and surveillance as well as genomics, has been established with the aim of collating C. burnetii positive samples from a wide range of hosts like livestock, wildlife and humans, with accurate associated clinical meta-data. Isolation methods will be optimized within the project to obtain new and archived isolates for WGS to generate a comprehensive database of annotated C. burnetii genomes, and phenotypic data from the field and in-vitro cellular assays as proxies of in vivo virulence. Based on phylogenetic analyses, strains representing distinct genomic groups will be selected and phenotypically characterized in cell culture and whole blood infection models. The combination of geno- and phenotypic data should allow the identification of molecular determinants of host range and virulence. An important aim of this project is to create a pan-European framework for future molecular surveillance of C. burnetii. These opportunities in surveillance and advanced techniques will most likely bring source tracing of *C. burnetii* infections to the next level.





#### Oral presentation 14: Variation of semen quality of rams and bucks during the year (Nina Ossowski)

Clinic for Swine and Small Ruminants, Forensic Medicine and Ambulatory Service, University of Veterinary Medicine Hannover, Germany

Co-Authors: Martin, Ganter, Clinic for Swine and Small Ruminants, Forensic Medicine and Ambulatory Service, University of Veterinary Medicine Hannover, Germany

Introduction. Small ruminants are classically considered as short day breeders. As the length of daylight decreases, the willingness to mate increases. This hormonally controlled behavior is based on the fact that lambs are born in spring with a sufficient food supply.

In some breeds, seasonality has been bred out through targeted breeding. A distinction can now be made between breeds with seasonal and aseasonal reproductive cycles. In aseasonal breeds mating is performed throughout the whole year whereas in seasonal breeds mating is mainly performed in autumn and winter.

In practical small ruminant husbandry, seasonal breeds predominantly give birth in the barn in winter and early spring, while the continous lambing of aseasonal breeds mainly takes place on pasture. Aseasonality plays an important role especially in dairy farming. Only by aseasonal lambing all year-round dairy farms can supply milk throughout the whole year.

Heat stress in summer can result in reduced sperm quality, especially in aseasonal breeds. In this study, the sperm quality of seasonal and aseasonal breeds was examined throughout different seasons of the year.

Material and Methods. Data were collected between August 2017 and August 2023 at the EU-approved artificial insemination centre (Al-centre) for sheep and goats at the University of Veterinary Medicine Hannover (D-KBSZ-003-EWG).

The rams and bucks housed at Hannover University are routinely bred at least once a week throughout the whole year.

Each ejaculate collected is examined immediately after collection in our in-house laboratory. Examination primarily includes a visual check, volume (ml), progressive motility (%), density (mill/ml) and total sperm count (bill).

In the following, the quality parameters of seasonal and aseasonal sheep and goat breeds are compared over the course of the year. One buck/ram per breed was observed.

Results. Ejaculate volume of the AG was significantly higher in spring than in winter. In summer, on the other hand, the volume of the EFMS was higher than in the rest of the year. Regarding progressive motility, there were no significant differences between breeds and seasons. Density and total sperm count were subjects to high animal-specific variation throughout the year and were highest across breeds in different seasons.

Discussion. Unlike comparable studies, there were no significant differences in semen quality of seasonal and aseasonal breeds throughout the year in this study. This can be mainly explained by the constant temperature and lightening system in the Al-centre even in summer. In addition, rams become shorn twice a year to avoid heat stress. Moreover, only males with a consistently good libido and excellent sperm quality are used for semen collection.

On farm fertility of seasonal breeds is significantly reduced in spring and summer. This condition affects both males and females equally, so that mating often only occurs from late summer onwards. Even in aseasonal breeds, a slightly reduced reproductive performance can be observed over the hot season, although this is much less noticeable than in seasonal breeds.





Oral presentation 15: Influence of dystocia on lamb mortality and postpartal ewe health compared to uneventful vaginal delivery and planned caesarean section (Katja Voigt)

Clinic for Ruminants, LMU Munich, Germany

Co-Authors: Mara Theisges, Viktoria Balasopoulou, Frank Weber, Holm Zerbe, Yury Zablotski

Introduction. Dystocia has been attributed to a significant percentage of peripartal ewe deaths and is also a major contributor to perinatal lamb mortality. Field data are however commonly confounded by management, husbandry and underlying flock health issues and frequently lack a control group. The true influence of dystocia is therefore difficult to assess from such data. In the field, it is frequently the quality or (in)adequacy of dystocia management and treatments which are decisive for survival or alleviation of suffering.

The true impact of dystocia on ewe and lamb mortality and post-partal ewe health can therefore only be assessed in healthy animals under best practice conditions, and ideally in comparison not only to uneventful vaginal deliveries but also in comparison to planned caesarean sections, a delivery technique which avoids any potential negative impacts of a vaginal birth process on lamb viability. In addition, the influence of planned caesarean sections on post-partal ewe health was assessed in healthy animals in comparison to uneventful vaginal parturitions and well-managed dystocia in order to shed light on any potential effects of surgical delivery on ewe health in the absence of any concurrent health issues.

Methods. Lambing records (between 2009 and 2023) of a closely-monitored university sheep flock of a high health status and free from infectious abortions were thus evaluated to assess the influence of dystocia (n=75) on lamb mortality and post-partal ewe health compared to uneventful parturition (n=283) and elective caesarean sections (n=218).

Univariable Bayesian logistic regressions were initially performed for the four response variables stillbirth, perinatal lamb mortality, retained foetal membranes and post-partal fever episodes and their corresponding predictors (the mode of delivery and potential confounders). The predictors were subsequently ranked for their relative importance by means of p value and Chi Square Statistics as an effect size. Final multivariable models were selected by the automated brute-force model selection method with multimodel inference for variable importance using the glmulti package in R. The brute-force approach provided the list of models ranked by the Akaike information criterion (AIC), where the model with the lowest AIC contained the most informative combination of predictors.

Results. Parturition-related post-partal ewe mortality was 0.2% (1/576 lambings). Overall stillbirth and perinatal mortality rates of 3.4% and 7.1% were observed (n=980 lambs). The mode of delivery (p<0.001) and lamb birth weight (p<0.001) were the most influential factors for stillbirth, with increased odds following dystocia and for lambs with low birth weights. Low birth weights (p<0.001) were also the most influential factor for perinatal mortality, but increased odds were also associated with dystocia. Retained placenta was the most influential risk factor for post-partal fever episodes (p<0.001), but the mode of delivery also showed a tendency (p=0.058), with increased odds following dystocia.

Discussion and Conclusions. The high ewe and lamb mortality rates frequently associated with dystocia under field conditions are unnecessary and can be avoided by good management. Parturition-related ewe mortality was 10 to 20-fold lower than published field estimates. However, dystocia remains an important influential factor for lamb mortality even under best practice conditions. Planned caesarean sections in sheep are associated with excellent clinical recovery and ewe and lamb survival.





## Oral presentation 16: Video surveillance and artificial intelligence in ovine parturition management (Vera Gebhard)

Clinic for Ruminants with Ambulatory and Herd Health Services, LMU Munich, Germany

Co-Authors: Häfner Dominik, Magana Gets, Vandeweijer Denise, Zerbe Holm, Voigt Katja

Introduction. Dystocia is an important factor contributing to lamb and ewe mortality. Early intervention is crucial to reduce losses and to avoid unnecessary suffering. Staff shortages or extensive husbandry systems are however often a hindrance to round-the-clock observation. Recent studies have therefore investigated whether a prediction of the time of parturition is possible using accelerometer data and Global Navigation Satellite System (GNSS)-sensors. Behavioural changes detected by these methods were however insufficient to accurately predict the onset on labour, and individual on-animal sensors are expensive in large flocks, may be lost or distract the animals. This study therefore aims to explore the use of computer vision and artificial intelligence (AI) algorithms in monitoring sheep flocks at lambing time, with the aim of developing a parturition alert system to enable timely intervention.

Methods. Video data was collected on four co-operating farms by 24h video surveillance during lambing. At first, the videos were annotated using bounding boxes to detect sheep and lambs, and later for key points on prominent body parts such as head and limbs, which can be reliably recognized. These key points were used to develop and compute a sheep skeleton fitting the six body parts head, torso and four legs. This enables the tracking of initially simple, skeleton-based movements. To further develop the 3D model, this skeleton was refined, and motion sequences were animated using blender software. This allows three-dimensional information to be obtained from the initially two-dimensional video data in order to provide the program with information on these behaviours from any perspective. The behaviours used for this were standing, lying, walking, pawing, circling, contractions, raising the head, and extending the hind legs to the side in a lying position.

In addition, the recorded births were analyzed regarding the frequency of parturition-related behaviours and events during the entire recorded birth period. The behaviours used for this were pawing, circling, abdominal contractions, raising the head, extending the hind legs to the side, licking, flehmen, lying down and standing back up, amniotic sac visible, amniotic fluid visible, parts of lamb visible.

Results. The average percentage share of behaviours and events in the total observed duration of parturitions was as follows for pawing: 0,4%, circling: 10,2%, contractions: 20,1%, raising the head: 9,1%, extending the hind legs to the side: 6%, licking: 0,5%, flehmen: 1%, amniotic sac visible: 0,4%, amniotic fluid visible: 1,5%, parts of lamb visible: 5,7% and the animals showed an average of 6,5 times of lying down and standing back up. These results form the basis for refined AI algorithms. Good progress has already been made in recognizing simple behaviours and tracking the individual animals. Work on refining the models for recognizing complex movement patterns is ongoing.





# Oral presentation 17: Investigation of the prevalence of subclinical mastitis and its dynamics between lactations in meat producing sheep (Alberto Luque-Castro)

University of Edinburgh, Royal (Dick) School of Veterinary Studies and The Roslin Institute, Easter Bush Veterinary Centre, Roslin, UK

Co-Authors: Neil Sargison, Alastair Macrae, Rob Kelly

Cases of clinical mastitis in meat-producing sheep are reported to peak at the first week post-lambing and at 3 to 4 weeks of lactation. Known risk factors such as low milk yield due to poor ewe nutrition, or litter size which lead to lamb-induced damage to the teat allowing in infection would explain the incidence peak at 3-4 weeks of lactation. However, it seems unlikely that these factors would have enough time to result in teat damage from newborn lambs causing clinical mastitis within the first days of lactation. The intramammary infection that leads to clinical mastitis at birth or early lactation may be associated with an unresolved subclinical infection from the previous lactation or a new infection during the dry period.

This study aims to investigate whether ewes without gross lesions to the udder are subclinically infected at weaning, whether these infections self-cured or become chronic and whether new infections are acquired during the dry period.

In a Scottish lowland indoor lambing flock, udder half milk samples (n=780) were collected from meat-producing breeding ewes at weaning and lambing (n=190 and n=213, respectively). Milk samples were analysed for somatic cell counts (SCC) and the literature cut-off of 500,000 cells/ml was used for interpretation. Data on body condition, age, litter size and udder palpable defects was also recorded. Aseptically udder half milk samples were collected and frozen for further bacterial culture and isolate identification with matrix-assisted laser desorption ionization (MALDITOF).

At weaning (n=190), the average SCC was 644,000 cells/ml (95% CI: 491,000-769,000 cells/ml) and 39.5% of ewes had at least one udder half above the subclinical cut-off value. At lambing (n=213), the average SCC was 1295,000 cells/ml (95% CI: 968,000-1623,000 cells/ml) and 39.9% of ewes had at least one udder half above the subclinical cut-off value. Looking at the variation of SCC of the ewes with results at both sampling times (n=107), 39,2% had SCC below the cut-off at weaning and lambing at least in one udder half, 20.6% had SCC above the cut off at least in one half at weaning and low SCC at lambing and 16,8% had SCC below the cut off at weaning and at least one udder half with high SCC at lambing.

This study highlights the potential incidence of subclinical mastitis in a meat-producing sheep flock. Bacteriology and further analysis looking into the relationship of SCC between udder halves and potential risk factors are ongoing.





# Oral presentation 18: A Case Study – Using a Brix refractometer to assess lamb serum total protein in the investigation of colibacillosis on a commercial sheep farm (Phillippa Page)

Flock Health Ltd, UK

Co-Authors: Luke Johnson, Joseph Angell, Fiona Lovatt

Introduction. Neonatal lamb bacterial infections on UK sheep farms were historically managed by use of prophylactic antibiotics. This practice has reduced significantly to the extent that by 2022 there were no oral products authorised in the UK for use in neonatal lambs. Increasingly Brix refractometers are used on UK farms to assess colostrum quality with data from 1295 Welsh ewes indicating associations between colostrum quality, diet and ewe body condition (BCS).

Repeated incidents of clinical colibacillosis ('watery mouth disease') on a 350 ewe-commercial sheep where the shepherd was conscientiously testing the colostrum of every ewe, led to the clinical decision to monitor levels of lamb serum total protein as an indication of whether sufficient passive transfer of immunoglobulins was taking place.

This study took the opportunity to use these clinical data from lambing 2023 to assess factors other than colostrum quality that may affect the incidence and risk of watery mouth causing increased lamb mortality on a commercial farm in the UK.

Materials and Methods. During the lambing period, veterinary visits were made on four occasions to blood-sampled all lambs that were 24-48 hours old. One hundred and three lambs were sampled from the jugular vein into a serum vacutainer tube (red top). The levels of total protein in the serum were measured using a Brix refractometer. All lambs born were weighed and ear tagged. Further data collated from all animals included dam BCS, breed, age and colostrum quality recorded by the farmer using the Brix refractometer. The timing of colostrum first suckled was also recorded in the following ranges <2 hours, 2-6 hours and >6 hours from birth.

All lambs were weighed at 12 weeks old for calculation of daily liveweight gain to 12 weeks and fate outcomes were recorded (i.e. sold to slaughter, retained in flock, or died).

Analysis and Results. Initial analysis used a serum total protein cut off value of 8.6% on the Brix though since collection of these data, optimal thresholds for IgG levels measured by digital Brix refractometry of both ewe colostrum and lamb serum have been published and further consideration is being given to the results.

Logistic regression has considered risk factors with the incidence of colibacillosis, mortality, lamb serum total protein at 24-48 hours old and growth to 12-weeks old as outcomes. Preliminary analysis suggests that the timing of first suckle of colostrum significantly affects the serum total protein measurement of the lamb.





# Oral presentation 19: A randomised controlled trial to compare the use of a novel product with strong iodine to protect navel and ear tag sites of neonatal lambs (Fiona Lovatt)

Flock Health Ltd and University of Nottingham, UK

Co-Authors: Phillipa Page

Background - In the UK, ovine *Streptococcus dysgalactiae* bacteria is the most common cause of infectious arthritis (joint ill) in newborn lambs with a previously reported prevalence of up to 50% of lambs and a subsequent mortality of 20% of cases. Joint ill and navel ill infections in newborn lambs and calves are currently managed prophylactically, typically by the application of strong iodine to the umbilicus area, and sometimes ear tag sites, via dip-immersion or aerosol spray.

The aim is to fully desiccate the umbilicus whilst providing antiseptic activity against surface dwelling bacteria. In addition, lambs may be given prophylactic antibiotic injection in the first few days of life to prevent such infections: however, routine prophylaxis is no longer considered an appropriate use of antibiotics. There is actually very little evidence for the effectiveness of iodine as a preventative in joint ill and, indeed, studies with calves have reported no fewer cases of external umbilical infections in calves' navel-dipped with 7% iodine tincture compared with those not treated.

Concerns were raised in 2022 over production issues and, consequently reduced global availability of iodine causing prices to quadruple. In response to these concerns, NoBACZ Healthcare developed a long-acting adhesive solution comprising alcohol, shellac and transition metal ions.

Methods & Results - A pilot study of the use of NoBACZ Navel was undertaken on 440 neonatal lambs and 30 calves on five different farms in 2023; there were no cases of joint ill recorded and positive farmer reports of usability.

A further randomised controlled trial to compare outcomes for lambs treated with strong iodine versus NoBACZ Navel was undertaken at 2023/24 lambing for 12 flocks that were already electronically recording their data. Data were gathered from over 2600 lambs with respect to disease prevalence, mortality and growth rates to 56 days. These numbers of cases are sufficient, in terms of power, to demonstrate equivalence (primary outcome) or superiority (secondary outcome) versus the effectiveness of strong iodine.

Summary - Should there be further issues with the global supply of iodine, it is important for ruminant practitioners to have confidence in the effectiveness of an equivalent, and potentially superior, product.





#### Oral presentation 20: Transmission of Reindeer Chronic Wasting Disease to sheep (Lars August Folkman)

Section for Small Ruminant Research and Herd Health, Institute of Production Animal Clinical Sciences, Norwegian University of Life Science, Norway

Co-Authors: Erez Harpaz, Federico Cazzaniga, Sylvie Benestad, Tram Thu Vuong, Linh Tran, Michael Tranulis, Arild Espenes, Fabio Moda, Cecilie Ersdal

Chronic wasting disease (CWD) is a prion disease in cervids. These disorders are fatal neurodegenerative diseases, caused by the deforming of the normal prion protein PrPC to the pathologic form PrPSc. CWD was first discovered in Colorado, United States (US) in 1967 and has since been reported in Canada, South-Korea and Fennoscandia. The first Norwegian case was detected in 2016 in a reindeer in the mountain region Nordfjella. CWD in reindeer is a contagious disease similar to scrapie in sheep, and prions are found in peripheral tissues as well as in the central nervous system (CNS). Later, sporadic cases of CWD have been found in moose and red deer. In these cases, prions seem to be confined to the CNS. In the US, sheep have been experimentally infected with CWD from mule deer and white-tailed deer. In vitro, rodent studies have revealed that American and Norwegian CWD are composed of different strains, and it is uncertain whether the American results are extrapolatable. It is therefore important to further investigate the transmissibility of Norwegian CWD to sheep.

Six newborn Rygja sheep with scrapie susceptible genotype VRQ/VRQ were orally inoculated with brain and spleen from CWD-positive reindeer, and six 2.5-3.5 month old VRQ/VRQ lambs were inoculated intracerebrally with a CWD-infected brain homogenate. Two sheep were orally mock inoculated to serve as controls. Three of the orally inoculated and two of the intracerebrally inoculated animals were euthanized before the study endpoint due to intercurrent disease. The animals were neurologically examined regularly, and blood, feces and rectal biopsies were sampled throughout the study. The study endpoint was 72 months for the oral group and 42 months for the intracerebral animals. At the end of the study the sheep were necropsied, and tissues collected for analysis using classical methods (histopathology, Immunohistochemistry, ELISA and Western Blot) and ultra-sensitive methods (Protein Misfolding Cyclic Amplification and Real Time Quaking Induced Conversion) for prions.

The concept of species barriers describes how prions cause low attack rates and slow propagation when transmitted between distant species, followed by more aggressive disease and attack rates with subsequent passages. It is therefore hypothesized that classical methods may not be able to detect infection on first passage, while highly sensitive methods may. At the conference, we will present results from both the intracerebral and the per oral transmission study.





# Oral presentation 21: MRI changes observed in a case of atypical scrapie in a 7-year-old Herdwick ewe (Sander Prins)

University of Glasgow, UK

Co-Authors: Kim Hamer, Ana Cloquell, John Spiropoulos, Neil Sargison, Piet Vellema

Background. Atypical scrapie is a transmissible spongiform encephalopathy that is rarely diagnosed in alive animals. In March 2022, a seven-year-old Herdwick ewe was referred to the Scottish Centre for Production Animal Health and Food Safety at the University of Glasgow because of circling behavior and ill thrift.

The ewe had a low body condition score, was obtunded, with a wide-base stance of the pelvic limbs, and was circling to the left.

Methods and results. Haematologic, biochemical, and cerebrospinal fluid analyses were unremarkable, but postmortem magnetic resonance imaging (MRI) findings were consistent with diffuse, bilateral, and symmetrical atrophy of the forebrain and ventriculomegaly. The clinical signs, the involvement of an individual older ewe, and the MRI results led to the clinical diagnosis of scrapie. Immunohistochemistry on the fixed brain, performed by the UK Animal and Plant Health Agency, showed deposits of PrPSc, which is a specific disease marker of transmissible spongiform encephalopathies, mainly in the cerebellum and at lower concentrations in the cerebrum and obex; consistent with the diagnosis of atypical scrapie.

Discussion. MRI findings in a sheep with atypical scrapie have not previously been described, to our knowledge. Scrapie should be included in the list of clinical differential diagnoses when veterinarians are presented with sheep showing progressive neurologic signs of several weeks' duration.





# Oral presentation 22: The long road to diagnosis of the causes of weight loss and mortality in a sheep flock (Aija Mālniece)

Faculty of Veterinary Medicine, Latvia University of Life Sciences and Technologies, Latvia

Co-Authors: Martin Ganter

The Sheep and Goat Health Service of the University of Veterinary Medicine Hannover, Germany was asked for help by a shepherd who had established a new flock for landscape protection on an 800 h rewetting of a peat bog project. The flock was established in July 2023. The owner bought 500 sheep, including 400 White Polled Heath ewes, 70 Horned White Heath ewes and 30 Bentheimer Landrace sheep from three different origins. Sheep spend most of the year on the pasture and were housed for lambing in a new barn. Three months after the purchasing, 2 ewes died and the whole flock started to lose weight. From October to December 2023, all sheep were dewormed once per month with different anthelmintics (ivermectin, doramectin, followed by moxidectin). Pooled FECs after deworming indicated a possible resistance to macrocyclic lactones. All sheep were dewormed with monopantel in January 2024. Fenbendazole in drinking water was used to treat cestodes. In search of other causes of weight loss, liver tissue examination for Se and Cu levels were performed and revealed deficiency of both - Se and Cu (Se 0,118 mg/kg FS and Cu 5 mg/kg FS). All animals were treated with 50 ml of 2% CuSO4 solution orally, twice at 1 month interval, and sodium selenite and vitamin E solutions were injected. Despite these efforts, the White Heath sheep became increasingly emaciated, apathetic and were dying at regular intervals. By 02.05.2024. a total of 80 sheep had died.

During the farm visit in May 2024, one hundred animals left in the barn were inspected. Mineral feed consumption per animal per day was calculated as an average of 3.5 g. The ewes had a low BCS. Three sheep were in sternal or lateral recumbency and were extremely emaciated (BCS =1) and depressed, one had diarrhea. Another sheep had severe dyspnea, coarse crackles and abscess of the left parotid lymph node, BCS =1. Blood samples for paratuberculosis antibody and caseous lymphadenitis (CLA) detection were taken from these animals and they were euthanized. Sheep with respiratory distress was taken for necropsy to test for ovine pulmonary adenomatosis (OPA) using PCR from a lung sample. Two other White Heath ewes had dental pathologies (sharp tooth edges, very long incisors, missing teeth) and these animals were over 8 years old by dental assessment.

Preliminary results. Paratuberculosis was confirmed by serology in 2 out of 5 samples. Individual FEC were still high in some sheep. Deficiencies of Se and Cu were present due to inadequate dietary intake of minerals. CLA was confirmed by postmortem, culture and serology. OPA was confirmed PCR positive.

Conclusion. The diagnosis of Johne's disease can be obscured by other health problems such as parasitosis, trace element deficiencies, coexistence of other infectious diseases, and can be highly prevalent in certain sheep breeds.





# Oral presentation 23: Prevalence of gastrointestinal and lung parasites in the goat population of Poland (Zofia Nowek)

Division of Veterinary Epidemiology and Economics, Institute of Veterinary Medicine, Warsaw University of Life Sciences-SGGW, Warsaw, Poland

Co-Authors: Marcin Mickiewicz, Kinga Biernacka, Adrian Valentin Potârniche, Michał Czopowicz, Agata Moroz-Fik, Tomasz Nalbert, Olga Szaluś-Jordanow, Paweł Górski, Iwona Markowska-Daniel, Jarosław Kaba

Background. Numerous genera and species of parasites of the alimentary and respiratory tracts affect small ruminant populations worldwide and cause considerable losses in production and farming. However, knowledge of their prevalence in the Polish small ruminant population is still limited. Poland is one of the largest European countries, spanning an area of 312,696 km² divided into 16 administrative provinces known as voivodeships. The goat population in Poland currently counts approximately 100,000 heads and continues to grow. This study aimed to investigate the prevalence of gastrointestinal and lung parasites in the goat population of Poland.

Materials and methods. Fecal samples were collected in 2015-2024 from 195 goat herds (including 2,667 individual animals) scattered over the entire country. All samples were examined using the modified McMaster method, centrifugal sedimentation, and the Baermann technique. Additionally, coproculture of the gastrointestinal nematodes (GIN) was prepared from all samples by mixing collected faeces and pooling them in a single sample. After 7-day incubation the larvae were extracted by Baermannization and 100 subsequent strongyle larvae of the third type (L3) were identified based on their morphological features at light microscope (400× magnification). The genera or species of L3 were identified at the herd level.

Results and discussion. The most prevalent parasites were GINs of the strongyle type present in 189 herds (97%) and 2,216 individual animals (83%). Among the GINs, *Haemonchus contortus* (171 herds; 88%) and *Trichostrongylus* spp. (164 herds; 84%) were most common, followed by *Teladorsagia* (*Ostertagia*) *circumcincta* (92 herds; 47%), *Oesophagostomum* spp. (74 herds; 38%), and *Nematodirus* spp. (12 herds; 6%). Furthermore, *Strongyloides papillosus* was found in 193 animals (7%) from 49 herds (25%), *Moniezia* spp. in 16 animals (0.5%) from 12 herds (6%), *Skrjabinema* spp. in 15 goats (0.5%) from 14 herds, and *Trichuris* spp. in 35 animals (1%) from 25 herds (13%). From among the protozoa, only *Eimeria* spp. were detected – the infection was present in 1,238 goats (46%) from 140 herds (72%). The only parasite of the respiratory tract found in the study was the nematode *Muellerius capillaris*, present in 151 herds (77%) and 1,226 goats (46%). The least common parasite found in our study was *Fasciola hepatica*, confirmed only in 1 goat (0.03%).

Conclusion. The goat population in Poland is most often affected by GINs belonging to the *Trichostrongyloidea* family (*H. contortus*, *Trichostrongylus* spp. and *Teladorsagia*(*Ostertagia*) *circumcincta*). These nematodes are responsible for most parasite-related problems in goats in Poland. Among the lung parasites, *M. capillaris* is the most common. Moreover, *F. hepatica* is an extremely rare parasite of the gastrointestinal tract in the goat population of Poland, so the health risk associated with this parasite is negligible.

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# Oral presentation 24: Gut-health evaluation in small ruminants: a multidisciplinary approach to optimize animal health and performance (Maria Teresa Capucchio)

Department of Veterinary Sciences, University of Turin, Italy

Co-Authors: Talal Hassan, Muhammad Irfan Malik, Muhammad Adnan Arif, Elena Diaz Vicuna, Lara Rastello, Laura Gasco, Claudio Forte, Manuela Renna

Health of gastro-intestinal tract (GIT) is defined as the absence/avoidance/prevention of gastrointestinal diseases to maintain animal health, welfare, and performance. In fact, digestion and nutrient absorption are strictly linked to the maintenance of gut-health, and any impairment of the GIT functionality may affect animal performance, increase the susceptibility to diseases and consequently lead to an overall increase in production cost. Gut-health depends on a delicate balance between the host, intestinal environment, and feed. Changes in diet composition may modify GIT activity, leading to a decrease in nutrient digestion and absorption. The maintenance of an optimal gut-health relies on an adequate gut morphology, gut mucin expression, local immune response, and gut microbiota. Gut morphometry and histological assessment are crucial for evaluating the presence of any intestinal mucosal damage, representing the "physical mucosal barrier" that separates the lumen from the underlying sterile tissue, thus avoiding bacterial translocation and assuring adequate nutrient absorption. In small ruminants, the reticulorumen is the major site for microbial fermentation of feed and hence, the production of volatile fatty acids (VFAs), which provide approximately 70-75% of the total metabolizable energy, but also prevent the invasion of rumen microbes and/or toxins. The luminal surface of the rumen wall is covered with papillae that increase the surface area for absorption of VFAs. It has been known for many decades that ruminal papillae change in response to dietary inputs, therefore morphometry and histological assessment of the rumen represent good indicators of the ruminal health. Histomorphological studies of small ruminant GIT health need to consider rumen and gut parameters, with special attention to area of absorption, dimensions of papillae and villi, and wall integrity (mucosa, submucosa, and muscle layers). Ruminal VFAs and microbiota should also be evaluated, as dietary changes can alter the microbial population and modify VFAs production which may lead to a worsening of animal performance or making the animal more susceptible to metabolic disorders. The authors describe the modalities for the assessment of GIT health in small ruminants, drawing on their research experience, stressing the importance of a comprehensive and multidisciplinary approach. Dietary manipulations using functional compounds (e.g., polyphenols, chitins) or novel ingredients (e.g., hazelnut skins) can improve ruminant health by modulating rumen microbiota, potentially increasing ruminal papillae length, and reducing epithelial keratinization. This approach may also mitigate intestinal inflammation commonly seen in intensively reared animals fed on soy-based diets (mononuclear and eosinophilic inflammation), by decreasing its severity or favoring a change in immune response (only mononuclear cells); suggesting a potential decrease of allergic immunostimulants. The observed results suggest that functional compounds/novel ingredients in small ruminants feeding show no negative effects on histomorphometry and allow the maintenance of the regular function of the gastro-intestinal barrier. Future studies should explore their use alongside conventional diets to optimize animal performance and health.





### Oral presentation 25: Evaluating kid rearing practices on dairy goat farms using data tools (Eveline Dijkstra)

Department of Small Ruminant Health, Royal GD, Deventer, the Netherlands

Co-Authors: Henriëtte Brouwer-Middelesch, Tara de Haan, Nienke Snijders-van den Burgwal, René van den Brom, Inge Santman-Berends

Modern dairy goat farming is increasingly aiming for sustainable farming methods, including young stock rearing. Optimal young stock rearing benefits from animal health and welfare and therefore contributes to a more sustainable herd. To evaluate the rearing process, producers require insight in their kid rearing results. Data regarding mortality and antimicrobial usage, which are available for all Dutch dairy goats, provide the opportunity to define key performance indicators to assess young stock health and welfare in dairy goats. Additionally, for each indicator the Dutch average can be defined with which the results of individual goat herds can be compared.

This study aimed to develop a data tool to help dairy goat farmers evaluate their kid rearing practices and compare it to a benchmark based on key performance indicators, related to mortality and antimicrobial usage. Indicators were developed using routinely collected census data of 395 dairy goat herds. Based on the quality of Identification and Registration data, i.e. accuracy and completeness of the data, three groups of dairy goat herds were distinguished (good, average, and poor quality). Four mortality indicators were defined: mortality risk of neonatal kids, and mortality rates of postnatal, preweaning and postweaning kids. Annually mortality risks and rates were determined for each farm. Animal daily doses (ADD) over the rearing period were determined for each farm based on national registered medicine prescriptions and the average number of kids. To verify whether results of the data tool were indicative for the actual rearing situation, a validation study was conducted on 27 volunteering farms. Participating farms provided information on their rearing practises through a survey. Additionally, participating farmers were visited by a small ruminant veterinarian for an objective scoring of animal health and rearing conditions.

Results showed that recorded kid mortality was significantly higher in herds that were classified as herds with good quality of registration among all four mortality indicators. A benchmark value for antimicrobial usage could be determined based on antibiotic prescription data of 88% of all Dutch dairy goat herds. Participating farms in the validation study had a good (15 farms) or average (12 farms) quality of registration. Overall, on farms with a lower health score, more kids with signs of disease were seen compared to farms with a higher health score. However, since participation in this study was subject to voluntary submission, no farms participated with severely impaired rearing quality.

This study provided insight into kid mortality in Dutch dairy goat herds and showed that it is possible to monitor the quality of kid rearing based on routinely collected data. Knowledge about their rearing performances will support farmers in effectively improving kid rearing.





# Oral presentation 26: Integration of Smart Flock Management Systems in Sheep Reproduction: A Precision Livestock Approach (Koray Tekin)

Ankara University, Faculty of Veterinary Medicine, Reproduction and Artificial Insemination Department, Turkey

Co-Authors: Burak Yılmaz

Efficient sheep handling systems are essential in modern agriculture, offering significant reductions in time and labor while enhancing productivity. This study, involving 206 Merino sheep, focused on integrating advanced technologies, RFID ear tags and reader, sheep handling system, automatic weighing, and Bluetooth data converter, to streamline sheep management. The collected data including age, breed, body condition score, live weight, health status, estrus behavior, breeding records, pregnancy, and estimated delivery time. A subset of 60 females underwent a synchronization protocol with progesterone sponges for 11 days, followed by 500 IU PMSG administration, and controlled natural mating. Our findings indicated notable differences in estrus duration (P<0.001), significant correlations between body condition score and estrus duration (P<0.05), as well as a strong positive correlation between ram service capacity and pregnancy rates (Rs=0.9, P<0.037), and an inverse correlation between sperm concentration and pregnancy rates (Rs=-0.9, P<0.037). The gestation period prediction accuracy was within 3.23 ± 2.47 days (P=0.002), demonstrating the precision of our model in estimating lambing dates. Additionally, integrating alligators into routine sheep management practices led to a marked reduction in total management hours and labor required, from 4.89 hours ( $\pm$  2.60) to 2.24 hours ( $\pm$  1.23) and from 5.15 ( $\pm$  1.68) to 3.43 ( $\pm$  0.84) in labor, respectively, both being statistically significant (p < 0.00001). These results highlight the transformative impact of combining innovative technologies and non-traditional methods in sheep breeding and management, paving the way for more efficient, sustainable, and economically viable agricultural practices.





# Oral presentation 27: Assessing lung consolidation in goats using different ultrasonographic techniques (Giuliano Borriello)

Department of Veterinary Sciences, Clinical section, University of Turin, Italy

Co-Authors: Flaminia, Valentini, Giulia, Cagnotti, Maria Teresa, Capucchio, Sara, Ferrini, Simona, Zoppi, Antonio, D'Angelo, Claudio, Bellino

Goats are often affected by respiratory diseases and, despite ultrasonography can assess lung consolidation in several species, it is rarely used in these animals. So, this study aimed to evaluate the efficacy of on-farm lung ultrasonography in healthy and sick goats. Twenty-seven goats scheduled for slaughtering underwent to a complete clinical examination and lung ultrasonography. The ultrasonographic examinations were performed on manually restrained, not-sedated goats in quadrupedal stance using convex (1-8 MHz) and gynecological linear transducer (5-10 MHz) before and after shaving both the lung areas (3rd to 12th intercostal spaces). A solution of 70% isopropyl alcohol was applied as transducer agent and an assistant operator timed each procedure. To localize the consolidations, eight quadrants per animal were obtained and labelled alphabetically on the right side from A to D, on the left from E to H. Each one was classified on the basis of the presence or absence of lung consolidation and by maximum consolidation 'depth (4-point scale: 0 healthy; 1 lesion depth <1 cm; 2 lesion depth between 1 and 3 cm; 3 lesion depth >3 cm). After the goats were submitted to post-mortem examination, the sensitivity and specificity of ultrasonography examinations and Cohen's κ coefficient were calculated. Differences in the percentage of consolidation distribution were assessed using the McNemar test. In eighteen (66%) goats, chronic lung consolidations were found at post-mortem examination. Each ultrasonographic examination took less than 5 minutes, on average, to complete and no significant differences were found between the techniques. Sensitivity (83%-89%), specificity (100%) and Cohen's κ coefficient (0.67-0.72) values were high with all the ultrasonographic techniques and a lower (p≤0.01) percentage of class 1 lesions were found at ultrasonography compared to the postmortem examination. Nevertheless, both transducers effectively detected lung consolidation deeper than 1 cm.

In conclusion, ultrasonography seems an effective tool for lung examination in goats with chronic pneumonia. The ultrasonographic examination using the linear or the convex probes without shaving the hair could be a promising tool for the on-field diagnosis of pneumonia. However, further research on larger sample sizes are necessary to validate these findings.





### Poster session 1

Biosecurity: washing and disinfection - role of boots as a carrier of pathogens on sheep farms (Fabio Rampin)

Huvepharma NV

Co-Authors: Nicoletta D'Avino, Massacci Francesca, Michele Tentellini, Paola Papa, Alessandro Necci

Biosecurity refers to all those measures implemented to avoid the entry of a specific disease into an unaffected population, as well as limiting its spread if already present. This trial had the objective of determining the degree of contamination of the litter by some specifically identified etiological agents (bacteria, parasites and protozoa) and an evaluation of the possibility that farm workers can act as passive vectors through contaminated footwear.

Ten farms with a history of *Salmonella abortus ovis* outbreaks were selected and boots were walked through the litter in a defined area using a grid pattern before environmental swabs corresponding to 25 g of faeces each were taken. Samples came directly from boots after contact with the litter (boots L), from boots that had been pressure washed with water only (boots W), or from boots that were pressure washed and then disinfected (boots P). Then, samples were sown on selective media for the detection of *Salmonella abortus ovis*, *E. coli*/fecal coliforms, *Staphylococcus* spp./Coagulase negative *Staphylococci*, fecal streptococci and *E. coli* ESBL/AmpC. A "parasitological" boot sample was also taken and analysed for a FLOTAC quantitative parasitological examination, and compared with pools of faeces from pens in each sample unit.

Out of the 90 samples collected, none were positive for *Salmonella* spp., 8 out of 90 samples (9%) tested positive for *E. coli* ESBL/AmpC (7 from boots L and 1 from boots W). The antibiotic resistance profile showed resistance to ampicillin, cefotaxime, ceftazidime, ciprofloxacin, ceftazidime, tetracycline and cefepime in all isolates. Molecular phylogroup analysis showed that the most representative phylogroup was the Clade I phylogroup. The remaining strains tested belonged to phylum group A. For the other bacteria tested, the average in CFU/g and other descriptive statistics were reported for each boot treatment; the data show very low charge values from the samples of boots treated with disinfectant (boot P), somewhat higher values in boots treated only with water (boot W) while very high concentrations in boots 'as is' from the bedding (boot L).

In the 'parasitological' boots, *Eimeria spp*, Gastrointestinal nematodes, Bronchopulmonary nematodes, *Strongyloides spp.*, *Dicrocoelium dendriticum*, *Tenia*, *Trichuris spp.*, *Nematodirus spp.* were detected.

Biosecurity is certainly an indispensable element in the livestock sector. Aetiological agents of different natures often recognise common and overlapping pathways of spread, so that the control of a single risk is expressed in multiple control actions. This basic principle has been amply demonstrated by this work. Footwear is one of the main pathways for the spread of pathogens both within different farm areas, and between different farms when not properly managed. It is evident that applying a protocol for footwear cleaning and disinfection, or alternatively using disposable footwear, is an action that is simple to perform but undeniably effective.





#### Epidermolysis bullosa in a goat kid – A suspicious case (Nina Ossowski)

Clinic for Swine and Small Ruminants, Forensic Medicine and Ambulatory Service, University of Veterinary Medicine Hannover, Germany

Co-Authors: Inga Nägler, Malgorza Ciurkiewicz, Martin Ganter

Introduction. Epidermolysis bullosa (EB) is caused by a gene defect in humans and several animal species. EB is characterized by vesicular detachment of the epidermis (erosions and ulcerations on the gums, palate, lips, tongue and distal limbs). In veterinary medicine, a distinction is made between 3 forms. In human medicine, these forms are subdivided into subtypes. In German Black Headed Moutons an autosomal recessive gene defect is known. In goats the causative mutation is unknown.

Material and Methods. A goat kid was referred to our clinic. Since birth it showed skin lesions on the limbs with blistering and lameness. Anamnesis reported that in recent years, sporadically kids with identical symptoms were born on the same farm. Most of these kids died within the first days or were euthanized due to severe symptoms. Occasionally, the lesions heal spontaneously. The kid was pre-treated with NSAID's and antibiotics, but its condition worsened progressively. Skin biopsies were taken for further diagnostics. Despite intensive treatment, the kid had to be euthanized after 24 days, and a full post mortem investigation was then conducted.

Results. The skin on the ears, ventral chest and limbs showed crusty-scaly changes, the coronet rim and ankle showed a massive serous to purulent blistering. Microbiological tests indicated an unspecific secondary bacterial infection. Histopathology revealed pustular-purulent dermatitis with orthokeratotic hyperkeratosis. The skin of the ears, chest and distal limbs was highly affected by vesicular to erosive-ulcerative changes. Similar changes were seen on the mucosa of the palate, tongue, gums and esophagus. Visually intact mucosa could be detached by manipulation. Histological examination showed multifocal to confluent, severe erosive-ulcerative dermatitis with associated purulent inflammation, serocellular crusts, intracorneal pustules and superficial bacteria and fungi. The macroscopically less affected areas showed multifocal subepidermal clefting, partly with accumulation of protein-rich exudate. PAS reaction showed the basal membrane above the cleft formation. Similar findings were found in the tongue, gum and esophagus.

Discussion and Conclusion. Subepidermal clefting below the basal membrane provides the suspicion of epidermolysis bullosa dystrophica (EBD). In goats, EBD was described as an autosomal recessive gene defect. In this case, further investigations were initiated to identify the underlying genetic defect.





# Extensive unilateral facial and pharyngeal mass in a 2.5-year-old goat one month after extirpation of the right bulbus (Viktoria Balasopoulou)

Clinic for ruminants with ambulatory and herd health services, LMU Munich, Germany

Co-Authors: Elisabeth Pernpeintner, Simone Schabmeyer, Marcel Suchowski, Katja Voigt

Neoplastic lesions are not uncommon in goats. In a retrospective study from the USA, 102 tumours from 100 goats were examined. The most commonly diagnosed neoplasias were lymphomas, squamous cell carcinomas, and thymomas. The present case involves a 2.5-year-old goat, which was presented at the Clinic for Ruminants of the LMU in January 2024 due to a severe swelling and prolapse of the third eyelid, a severe corneal lesion with corneal opacity, and right-sided blindness. The owner suspected a fighting injury. Additionally, there was moderate exophthalmos, interpreted as a consequence of panophthalmitis. Since the eye was severely damaged, a bulbus extirpation was performed. Postoperatively, a haematoma developed at the temporal area and ventral to the orbit, which, however, had organized well by 14 days post-operation, when the animal was discharged from hospital in good general condition. One month later, it was re-presented with an extensive right-sided facial mass. General examination revealed emaciation (BCS 1/5) and elevated body temperature (40.0°C). The mass was protruding from the area of the right orbit, with the overlying skin intact, and extended to the right cheek, where it measured approximately 6 cm in diameter. The mandible showed a shift to the left, and the animal had difficulty in feed intake. Purulent discharge emerged from a fistula in the temporal corner of the orbita. A strong odour was noticeable from the mouth and slight purulent nasal discharge was also noticed. Following sedation, exploration of the oral cavity was undertaken, and radiographic as well as ultrasonographic examinations were performed to establish the extent of the mass. During oral exploration, a cauliflower-like tissue proliferation of about 7 cm was visible on the right side, spreading both buccally and lingually. Ultrasonographically, a solid tissue in the area of the right orbit was depicted. Suspicions of neoplasm or marked granulation tissue formation were raised. Due to the severe clinical findings, poor prognosis and markedly reduced general condition, the goat was euthanized. During the postmortem examination, a neoplastic mass was found on the right half of the head expanding from the extirpated bulbus area through the maxillary sinus into the oral cavity. Histologically an infiltrating malignant predominantly spindle-shaped tumour cell population was identified. Amongst others, rhabdo-/leiomyosarcoma, fibrosarcoma, or a peripheral nerve sheath tumour were considered as differential diagnoses. Finally, after immunohistochemical examination of the tissue, a Vimentin und alpha-smooth-muscle-Actin positive result was obtained, leading to a definite tumor differentiation and the diagnosis of a leiomyosarcoma.





#### Septic tendosynovitis in a breeding ram and isolation of Mycoplasma arginine (Eva Roden)

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Co-Authors: Irene Schairer, Viktoria Balasopoulou, Katja Voigt, Marcel Suchowski, Joachim Spergser, Julia M. Riehm

Introduction. In the European Union, highly pathogenic *Mycoplasma* species in small ruminants are mainly found in Mediterranean countries. Therefore, and due to the relatively elaborate diagnostic methods and low animal value, *Mycoplasma* species may currently be underdiagnosed in Germany. Nonetheless, *Mycoplasma mycoides* subspecies *capri* has recently been described in a German goat herd.

*Mycoplasma arginini* is frequently isolated from the respiratory and genital tract of healthy and diseased small ruminants. Nonetheless, until recently, *M. arginini* has not been considered a primary pathogen but a contributor to disease, exacerbating clinical signs in mixed bacterial infections. However, recent research results indicate that it may be more pathogenic than widely believed.

Other than in cattle, septic tendosynovitis seems to be a rare condition in sheep. Despite extensive literature research, no description of septic tendosynovitis in sheep has been found.

Case description. A Bavarian Alpine breeding ram was submitted to the Clinic for Ruminants, LMU Munich, due to persistent pneumonia with high fever and significant weight loss. Upon submission, besides the pneumonia, the ram exhibited a high-degree lameness on all four legs. All digital flexor tendon sheaths were fluid-filled to a high degree. Furthermore, the ram showed a mild brisket sore (hairless area with little crusts and no exudation).

Ultrasound examination and aspiration of synovia from the tendon sheaths led to the suspected diagnosis of septic fibrinous tendosynovitis. Due to the painful physical condition of the animal, the owner consented to euthanasia on the day of submission.

The post-mortem examination at the Bavarian Health and Food Safety Authority in Oberschleissheim revealed a chronically-active, fibrinosuppurative inflammation of the digital flexor tendon sheaths on all four legs. Additionally, a distinctive chronically-active, necrotizing suppurative to abscess-forming soft tissue inflammation was identified in the sternal region. Haemolytic *Escherichia coli* was isolated from tissue samples of the lungs, scrotum, digital flexor tendon sheaths and soft tissue covering the sternum. Furthermore, *Klebsiella oxytoca* colonisation of the lung and sternal soft tissue was identified. *Trueperella pyogenes* was isolated from samples of the sternal soft tissue. Additionally, mycoplasma was cultured from samples of the lung and digital flexor tendon sheaths. The isolates were sent to the Institute of Microbiology at the University of Veterinary Medicine in Vienna for identification. In both cultures, *M. arginini* was identified by MALDI-TOF mass spectrometry. Genome sequencing and characterization was performed using Nanopore and Illumina sequencing followed by hybrid assembly. Both genomes carried a prophage which has been associated with arthritogenesis in hosts infected with closely related *Mycoplasma* species (e.g., *M. arthritidis*) before.

Discussion. To the authors' knowledge, this is the first description of septic tendosynovitis in sheep and also the first isolation of *M. arginini* from tendon sheaths in small ruminants. It is important to stress that the contribution of *M. arginini* to the diagnosed tendosynovitis cannot be conclusively clarified. Nonetheless, this case report highlights the importance of a thorough diagnostic approach and draws attention to the possibly underestimated contribution of *Mycoplasma* species to small ruminant diseases.





# Gastrointestinal and pulmonary parasites in small ruminants in northeast Portugal – What's up doc?! A case herd study (Delia Lacasta)

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The breeding of small ruminants in northeast Portugal is largely based on a traditional production system, mainly on semi-extensive grazing and direct use of existing natural resources.

Gastrointestinal and pulmonary parasite infections are particularly important, as they can have impact on animal health and welfare, as well as on productivity, and therefore be responsible of economic losses including: delayed growth, weight loss, decreased fertility, and also rejections at slaughterhouse.

To act on the prophylaxis, one first should have a detailed knowledge of the parasite species involved, as well as their prevalence, in order to plan prevention and control strategies.

Faecal samples were analysed to monitor egg burdens and prevalence of gastrointestinal and pulmonary parasites in two autochthonous sheep and goat breed herds, with similar farm management, and sharing the same pastures.

Coprological qualitative and quantitative analysis (flotation, natural sedimentation, mini-Flotac and Baermann technique) were performed in two periods, February and April 2024, on fifteen animals in each flock.

The egg/oocyst genera morphologically identified were: trichostrongyle-type, *Nematodirus* spp., *Moniezia* spp., *Trichuris* spp. and *Eimeria* spp. Trematode eggs were not identified.

The mean egg per gram (EPG) values of trichostrongyle-type ranged from 25 to 67 among the sheep flock, and 62 to 90 among the goat herd.

The mean oocyst per gram (OPG) of *Eimeria* spp. varied greatly in the two flocks, ranging from  $0.46 \times 102$  to  $3.11 \times 102$  among the sheep flock, and  $4.74 \times 102$  to  $1.45 \times 104$  among the goat herd.

Fecal examination by the Baermann technique, revealed the overall prevalence of lungworm infection to be, in the goat herd, 21.4% to 33,3%, whereas the sheep samples were negative. The specie found were larvae from *Muellerius capillaris*.

This work aims to continue to: determine the gastrointestinal and pulmonary parasite profile of small ruminants along the year seasons; identify differences in parasite population in the two species, and assess the anthelmintic efficacy and possible resistances.

The development and implementation of innovative, refined and sustainable approaches to worm control, targeted at the appropriate regional scale, is a prerequisite for reducing the helminth infection burden in small ruminant livestock production, and therefore increase the productivity and profitability, and also contribute to the preservation of natural resources.





#### Changes in seasonal patterns of Haemonchus contortus infections in Northern Belgium (Eva Van Mael)

Animal Health Care Flanders, Belgium

Co-Authors: Willem Van Praet, Stefaan Ribbens, Nermin Caliskan, Emily Rolly

Worldwide, all grazing sheep are exposed to gastrointestinal nematode infections. *Haemonchus contortus* is one of the most important and common worm species in small ruminants and camelids in Belgium. In 2023 more than 42% of all the small ruminant necropsy cases at Animal Health Care Flanders showed parasite infestation, mainly *H. contortus*. This is an increase on the previous year with 3%. Adult worms nest in the abomasum, where they feed on blood sucked from the stomach wall, resulting in anemia. In severe cases, sheep farmers are surprised by sudden deaths.

H. contortus is originally a tropical worm species, so problems in the past usually occurred from June to September-October when temperatures are higher. In recent years, however, we have seen an extension of this period in Northern Belgium. Necropsy data show us that Haemonchus victims coming in from April or even earlier to deep into autumn or winter, are no longer an exception. The number of months in which we see cases increases every year. This is probably the result of increased resistance of H. contortus to anthelmintics used on the one hand, and milder winters on the other, which means that pasture infestation remains a real danger for a longer period of time. Sensibilisation of sheep farmers on this topic is very important and one of the core tasks of Animal Health Care Flanders.





### Salmonellosis in dairy goat farming; development of an accreditation programme (Eveline Dijkstra)

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Salmonellosis is an infrequently diagnosed disease in small ruminants in the Netherlands. Nevertheless, in 2016, high morbidity and mortality were observed amongst neonatal dairy goat kids due to an outbreak on two related farms caused by *Salmonella enterica* subsp. *enterica* serovar *Typhimurium*. Shortly after, salmonellosis was detected in some of the producers' family members, caused by the same MLVA-type of *S. typhimurium*. Descriptions of outbreaks of salmonellosis in humans derived from goats as source are rare. In collaboration with the dairy goat industry, a comprehensive study was initiated to investigate the prevalence of salmonellosis within the dairy goat sector, to identify potential risk factors and ways to control salmonellosis on dairy goat farms.

In 2021, all dairy goat farms in the Netherlands (n=403) were encouraged to participate in a prevalence study by taking repeated dust and faecal-contaminated bedding samples for culture of rearing facilities at week three and six of the kidding period. In addition, bedding samples were taken of trailers at the three largest small ruminant abattoirs after delivering young stock. Of 35 volunteering dairy goat farms of which the *Salmonella* spp. infection history was known, bulk milk samples from March, April, May, July and August 2023 were screened for the presence of antibodies against Salmonella spp. serotype B/D LPS, using an ELISA. In addition, with permission of milk processors, single bulk milk samples from April of 318 dairy goat farms were anonymously tested for antibodies.

Of the approached farms, 52% (N=209) participated in the prevalence study, of which 148 farms delivered the requested two samples within a lambing period and 61 farms participated just once. *Salmonella* spp. were detected on seven farms. Positive *Salmonella* spp. cultures were found on all abattoirs in seven of 37 pooled samples. Samples from farms with (recent) salmonella infections or using an autovaccine had higher S/P-values in the antibody ELISA, compared to farms with an unknown salmonella infections (median test, P<0.001).

Based on the participation of more than half of the Dutch dairy goat industry, a prevalence was found of 4%. Th The prevalence of salmonellosis amongst dairy goat farms is similar to the prevalence of salmonellosis in Dutch dairy cattle. Apparently, presence of *Salmonella* spp. in goats does not necessarily lead to a clinical outbreak, so it is expected that *Salmonella* spp. are more common on farms than previously thought. In addition, we found that a *Salmonella* BD-ELISA can contribute to distinguish dairy goat farms with or without salmonella infections.

Nevertheless, further research on the antibody dynamics in goats after salmonella infection and the relation between individual antibody responses and bulk milk antibody levels will be necessary to refine the cut-off value and provide more insight into the implication of the ELISA for goat bulk milk in an accreditation scheme.





# ddPCR assay for rapid identification and quantification of *Haemonchus contortus* eggs in small ruminant feces (Barbara Moroni)

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Co-authors: Zoccola R., Zoppi S., Goria M

Introduction. Identification and quantification of gastrointestinal nematodes (GIN) of small ruminants is of paramount importance for a correct and timely treatment. Nevertheless, traditional copromicroscopical methods based on egg count and larval culture of GIN are time-consuming and need the expertise of specialized operators for the morphological identification of larvae. Novel droplet digital PCR (ddPCR)-based applications have been recently validated to identify and quantify mixtures of genomic DNA extracted from different species of adult worms (Elmahalawy et al., 2018. Vet par 261, 1-8), although in-field applications of ddPCR should be tested to quantify parasitic egg-derived DNA in the feces, rather than in cultured adult worms. The aim of this pilot study was to test a new ddPCR protocol for *Haemonchus contortus* eggs collected from sheep and goat feces with symptoms compatible with haemonchosis in Piedmont (Italy).

Material and methods. First, *H. contortus* was isolated and morphologically identified from a goat that was necropsied at the Istituto Zooprofilattico Sperimentale in Turin. Then, from the same animal, feces were collected, and parasitic load was quantified using a concentration McMaster technique with a sensibility of 100 EPG (Roepstorff et al., 1998. FAO Animal Health Manual No. 3. Food and Agriculture Organization of the United Nations, Rome, pp. 51–56). Two solutions were obtained from McMaster chambers with 5 and 10 eggs, respectively, DNA was extracted from both using the QIAamp Fast DNA Stool Mini Kit (QIAGEN Inc.) and serial dilutions were prepared. All the dilutions were tested in triplicate and the amount of DNA from strongylides and from *H. contortus* was quantified in multiplex ddPCR with two different sets of primers and probes as described by Elmahalawy et al., 2018. As a positive reaction control was used an extract obtained from an adult parasite morphologically identified as belonging to the genus *Haemonchus*. Then, the same protocol was tested in feces of animals that presented symptoms compatible with haemonchosis, including severe anaemia.

Results and discussion. Preliminary tests correctly detected the presence of the target DNA in extracts in both the solutions (5 and 10 eggs), but only at the lowest dilution. Both assays tested (ITS2 in the ribosomal DNA gene array present in any strongylid nematode and the internal transcribed spacer region 2 of the ribosomal RNA gene array of *Heamonchus*) worked correctly with good repeatability of results. The copy number detected with the two ddPCR assays did not correspond proportionally to the number of eggs in the starting matrices. This result is probably due to a non-optimised extraction procedure. The results of this pilot study indicate that ddPCR can be regarded as a useful diagnostic tool to identify *Haemonchus* spp. in small ruminant feces, bypassing time-consuming and laborious techniques such as larval culture. Preliminary results on the quantification of the egg-load were not successful, although this will be object of further investigation.





#### Poster session 2

#### Efficacy of paromomycin for the prevention and treatment of cryptosporidiosis in lambs (Fabio Rampin)

Huvepharma NV

Co-Authors: Ylenia Abbate, Nicoletta D'Avino, Pilar De Ferrari, Andrea Felici, Marco Gobbi, Orazio Paoluzzi, Michele Tentellini

Cryptosporidiosis is caused by protozoa of the genus *Cryptosporidium* which affects several animal species including humans. In lambs, *Cryptosporidium parvum* is believed to be the most pathogenic species and is a major cause of neonatal enteric disease.

In this trial, 40 newborn Comisana and Sarda lambs, both female and male, were randomly distributed into two groups. Lambs in Group A (treatment) were treated with an oral administration of 50 mg/kg of paromomycin sulphate once per day from day 5 to day 9. Lambs in Group B (placebo) received an orally administered placebo of 50 mg/kg once per day from day 5 to day 9.

The experimental protocol began with weighing each lamb at birth (day 0). Faecal material was taken directly from the rectal ampulla of each animal on days 2, 4, 7, 10, 14, 21 and 28, and processed using the modified Telemann-Myagawa copromicroscopic technique for the detection of *Cryptosporidium* spp. oocysts, and using the SmartStripsTM Cryptosporidium-BIO K403 immunochromatographic technique.

The aim of the study was to assess the efficacy of paromomycin for the treatment and prevention of parasitosis, by means of the following measurements:

- assessing the difference in weight between the two groups
- analysing the percentage of animals affected by diarrhoea
- testing the correlation between the presence of diarrhoea and positivity to *Cryptosporidium* spp.

The animals were weighed again on day 28 to check whether the treatment with paromomycin sulphate had influenced weight gain. The results revealed that average weight was 1.1 kg higher in the Group A lambs compared to the lambs in Group B.

When analysing the *Cryptosporidium* spp. positivity rate, 31.1% of the lambs in Group A tested positive compared to 100% of the lambs in the placebo group. In Group B, a peak of positive animals was observed on days 7, 10 and 14 at 52.6%, 84.2% and 57.6%, respectively.

The percentage of animals with diarrhoea during the test were 31.2% in Group A and 42.1% in Group B. The presence of diarrhoea was positively correlated in coprological tests, and was more often associated with positivity to *Cryptosporidium* spp. in Group B than in Group A.

The preventive use of paromomycin sulphate significantly reduced morbidity of the disease, playing a key role in controlling symptoms and reducing the excretion of *Cryptosporidium* spp. In addition, its use had a positive effect on weight gain in the treated subjects.





### Innovative use of ultrasound in andrology and gynecology of small ruminants (Calogero Stelletta)

Department of Animal Medicine Production and Health, University of Padova, Italy

Co-authors: Koray Tekin, Daskin Ali

In farm animals, the use of ultrasound has become the gold standard of diagnostic imaging of the reproductive system. In small ruminants, recent approaches have considered an improvement in the accuracy and precision of the breeding soundness evaluation (BSE) thanks to the use of ultrasound for the evaluation of accessory glands. The overall study of the male reproductive system certainly improves the ability to treat post-collection ejaculate to increase the time of use of inseminating doses. In ewes and does the use of ultrasound has improved in terms of image resolution, type and position of probes. High technological content can be highlighted when assisted reproduction techniques are applied. The use of the echo-color technique seems to be the best option to evaluate the functional quality of the corpus luteum at the field level. Numerous other data relating to the integration of classically applied assisted reproduction techniques with the innovation of ultrasound technique are necessary.





# Effects of elective caesarean sections in healthy near-term ewes on subsequent reproductive performance (Katja Voigt)

Clinic for Ruminants, LMU Munich, Germany

Co-Authors: Mara Theisges, Yury Zablotski, Frank Weber, Holm Zerbe

Introduction. Post-surgical reproductive performance following ovine caesarean section has not been well studied. In clinical cases undergoing surgical delivery due to dystocia, the frequent presence of underlying health issues, the potential additional effects of dystocia, the difficulty of obtaining reliable long-term follow-up data and the owners' frequent decision of avoiding re-breeding for fear of future problems lead to major difficulties in studying any potential effects of caesarean section on subsequent breeding outcomes. This information is however important for farmers and vets to make evidence-based decisions as to the economic value of caesarean sections, and to guide culling or re-breeding decisions.

Methods. To assess any direct effects of surgical delivery in the absence of confounders such as dystocia or underlying diseases, we studied elective surgery performed in healthy animals for teaching purposes. Four hundred and eleven paired breeding records following vaginal delivery (n=233), elective caesarean section (n=122), and subsequent further vaginal deliveries in animals with a history of one prior elective caesarean operation (n=56) were evaluated retrospectively. From the available data, subsequent reproductive performance was measured as subsequent conception (based on the cumulative outcome of permitted mating attempts), the number of synchronization/mating attempts leading to this pregnancy, the subsequent litter size, number of liveborn lambs, stillbirth and perinatal mortality rates, lamb birth weights and the incidence of premature foetal death during subsequent pregnancy as measured by mummification and abortion. Statistical analyses were performed in R (version 4.3.1 (2023-06-16)). Uni- and multivariable Bayesian logistic regressions were applied for binary outcomes (conception, stillbirth, perinatal mortality, foetal death), and Bayesian linear regressions with Gaussian distribution family were used for numeric outcomes (mating attempts, litter size, number of live born lambs, birth weight).

Results. The overall subsequent pregnancy rate was 95%. Multivariable statistical analyses did not reveal any significant influence of planned caesarean surgery on subsequent conception, stillbirth, perinatal lamb mortality, lamb birth weights or the incidence of premature foetal death (measured by mummification and abortion). A significant reduction in litter size was however seen in the first pregnancy immediately following a surgical delivery in comparison to animals which had previously only delivered vaginally (p=0.001), but litter size returned to precaesarean levels in further follow-up pregnancies in animals with a history of one elective caesarean section (p=0.436). An increased number of mating attempts was primarily associated with increasing ewe age (p=0.025), but an immediately preceding caesarean section also showed significance (p=0.033).

Discussion and Conclusions. Subsequent long-term reproductive performance of sheep following elective caesarean section was excellent. A temporary reduction in litter size in the pregnancy immediately following surgical delivery is economically acceptable. The particular circumstances of the studied flock in relation to oestrus synchronization, year-round breeding and very short natural mating periods, if applied, are likely to have made an increase in the number of mating attempts come to light - it is however likely that these differences are not clinically relevant in field situations involving natural, seasonal breeding and mating periods covering at least two full oestrus cycles. The clinical and economical significance of this observed effect is therefore debatable.

The results fully encourage the use of caesarean section in ovine obstetrics, and the observed excellent post-surgical reproductive outcomes support retention for breeding.





#### Productive and metabolic profile of "Elite milking ewes" (Jose Maria Bello Dronda)

Nutreco Animal Nutrition Iberia, Spain

Co-Authors: Gustavo Gonzalez, Alejandra Valentin

Introduction. Genetic improvement on dairy sheep farms provides animals with high productive yields. High levels of production can produce metabolic disorders that should be prevented through proper nutrition. This study tries to find out if the feeding strategies of this type of animals, based on the usual nutritional models, are suitable under practical conditions. The objective of this work is to compare the results of two types of sheep (Elite, and High Production -HP-) belonging to the same farm and feeding group, regarding productive parameters and metabolic and health indicators throughout 4 periods set up per lactation (prepartum -PREP-, early lactation -EL-, medium lactation -ML-, and late lactation -LL-).

Material and methods. A total of 18 multiparous lacaune ewes were randomized into two groups of 9 animals each (Elite, with previous lactation of more than 650 liters) and HP (more productive ewes of the farm but without becoming Elite). Individual production data (milk production -MY-, percentage of fat -MF-, protein in milk -MP-, milk fat and protein production -KgF+P-), and body status data (body condition score -BCS-, live weight -BW-) were taken. In addition, some milk and blood parameters, as indicators of metabolic and health status were analyzed (somatic cells account -SCA-, transaminases AST- and -ALT-, beta hydroxybutyrate -BHBT-, non-esterified fatty acids -NEFA-, glucose -GLU-, and calcium -Ca-) during each of the 4 periods stablished (PRE, 7 to 13 days before lambing; EL, 3 to 38 days after lambing; ML, 49-100 days after lambing; and LL, 123 to 162 days postpartum). Statistical analysis was performed, both by groups of animals and by periods (ANOVA). Besides, the correlations between the parameters of animal status and metabolic and health parameters and the productive results were also calculated.

Results. The average of MY obtained was 3.28 liters/day in the Elite group and 2.59 in the HP one. Significant differences between the two groups of animals (p<0.001) in MY and KgF+P were found. The comparison between periods showed significant differences (p<0.001) in BCS, MY, MF, MP, KgF+P, ALT, GLU, NEFA. Although the differences by groups found in our study were expected, in the case of ALT (in all periods within physiological levels), we need to do more investigations. The most correlated parameter with MY was ALT (0.463, p<0.05).

Conclusion. There are no differences between animals in terms of metabolic and health indicators and milk quality, consequently we can conclude that our feeding strategies are suitably implemented in the practical conditions studied.





### A case of increased incidence of subclinical and clinical mastitis in Lacaune dairy sheep (Sander Prins)

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Co-Authors: Kim Hamer, Hélène Baudel, Neil Sargison, Piet Vellema, Dominique Bergonier

Background: In a flock of 80 Lacaune dairy ewes, an increased number of subclinical mastitis cases was observed at the start of the lactation period in 2023: 19 out of 80 ewes were observed positive with the California Mastitis Test (CMT). Two out of eight cultures of milk taken from affected ewes revealed the presence of coagulase-negative *Staphyloccus* spp. As the farmer was familiar with subclinical and clinical mastitis in previous lactation periods, he started vaccination against *Staphylococcus aureus* before the 2023 lactation period and introduced the CMT to detect possible cases of (sub)clinical mastitis earlier.

Methods: To investigate the increased incidence of subclinical mastitis, a farm visit was carried out to gain an overview of the farm and to observe milking technique and udder hygiene. Milk recording data from the 2022 lactation period was used to obtain a general overview of the flock's somatic cell count level.

Results: Observations during the farm visit revealed that gloves were not worn during milking and that the cups were not rinsed after milking an ewe with mastitis. The positioning of the teat cups during the cleaning cycle of the milking parlour was such that water remained in the cups until the next milking. Udder dermatitis was present in 40-50% of the ewes and was treated during milking. It was also observed that the lambs suckled on their mothers until weaning.

Discussion: The increased incidence of subclinical and clinical mastitis in this flock was most likely caused by deficiencies in milking technique, inadequate udder hygiene and environmental factors identified during the farm visit. Therefore, recommendations were made to address the main deficiencies: 1) Wear gloves and sanitise the teats before attaching the cluster. Change the position of the cup during parlour cleaning to avoid water residues in the cups. 2) Stop treating udder dermatitis during milking to prevent it from spreading in the parlour. 3) Since complete separation of lambs from their mothers does not seem possible, we advised to feed the lambs additional milk replacer to prevent aggressive drinking by the mothers when the lambs need more milk.

Although vaccination may support an approach in some cases, milking technique, milking hygiene, and environmental factors need to be considered before relying completely on vaccination in controlling (sub)clinical mastitis.





A topical anaesthetic/antiseptic formulation for the treatment of ecthyma in experimentally induced or naturally affected lambs. Effect on the concentration of serum amyloid A (Aurora Ortin)

Universidad de Zaragoza, Spain

Co-Authors: Sergio Villanueva-Saz, Antonio Fernández, Lucía Rodriguez, Alex Gómez, Pablo Quilez, Enrique Castells, Teresa Navarro, Juan José Ramos

Contagious ecthyma (CE), caused by the orf virus (ORFV), is a highly contagious eruptive skin infection of sheep and goats, which mainly affects young animals. Vaccines are not available in Spain, and treatment is based on standard hygiene practices and antibiotics to control secondary infections, increasing the risk of antimicrobial resistance. Alternative treatments are needed to improve the welfare of affected animals and control the spread of the disease. This study evaluated the effect of a topical anaesthetic/antiseptic formulation, Multi-Solfen® (MS, Dechra, UK), on the concentration of the major acute phase protein serum amyloid A (SAA) in naturally and experimentally ORFV infected lambs.

Fifty 15-day-old Lacaune male lambs from a CE free farm were intradermally inoculated with ORFV, and randomly divided into two equal groups, control group (group C) and group T treated with MS, sprayed on the lesions, on two occasions: one week and 11 days after the infection. Blood samples without anticoagulant were collected from all the lambs prior to the treatment (T0) and 2 (T2), 7 (T7) and 14 days (T14) after the first dose of MS. The concentration of SAA was assessed using a sandwich ELISA kit (PHASE TM SAA Assay, Tridelta Development Ltd, Ireland). Data were statistically analysed using IBM SPSS statistics version 26.0 software. In the control group, SAA concentration increased over time, reaching the highest value at T14, which was significantly higher than at T2 and T0 (p = 0.030; p = 0.013). However, group T peaked at T7, and significant differences between this sampling time and the others were not detected. In addition, SAA concentration at T7 and T14 was lower in this group than in the control group, being significantly lower at T14 (p = 0.041). These results point to treatment with MS reduced the SAA response, which peaked down earlier.

Taking into account these promising results, a new experiment was carried out on a sheep farm affected by a CE outbreak. One hundred Lacaune lambs 25-30 days old with CE lesions were selected and randomly divided into two groups, the control group (group C) and group T, which was treated with MS on 3 occasions, with an interval of 3 days between treatments. Blood samples were collected from 12 randomly selected lambs of each group prior to the treatment (T0), 10 days (T10) and 20 days (T20) after the first MS administration. SAA concentration was determined, and data were statistically analysed, as above. Significant differences in the concentration of SAA over time were not registered in group C, whereas in the group treated with MS, values at T10 and T20 were significantly lower than at T0 (p = 0.030 in both). Significant differences between the groups were not detected, although at T10 and T20, a statistical trend towards a lower value was observed in group T. Further research with a higher number of animals is needed to confirm these results. Still, they suggest that reduction of SAA response is also possible in natural CE outbreaks by treatment with MS.





### Computer tomography to diagnose upper respiratory tract disorders in sheep (Pablo Jose Quilez)

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The accurate diagnosis of upper respiratory tract diseases (URD) in sheep is complex due to the similarity of clinical signs of the different pathological disorders. The differential diagnosis of URD in Spain includes the following main disorders: oestrosis, enzootic nasal adenocarcinoma (ENA), chronic proliferative rhinitis (CPR) and obstructive rhinopathy due to pithomycotoxicosis. Computed tomography (CT) is an innovative diagnostic imaging technique that offers enormous diagnostic advantages that are clouded by its high cost. Although CT scanning is currently used primarily for research purposes, its use will probably expand in the future.

In this study, CT scans of the heads of 22 sheep clinically diagnosed with URD were evaluated. A complete post-mortem study was subsequently carried out on all of them. A total of 9 sheep with CPR, 6 with ENA, and 7 with obstructive rhinopathy were examined. In addition, *Oestrus ovis* larvae were found in three animals; however, oestrosis was not considered the primary disease. CT scans were performed under total anaesthesia using a Bivro two-slice CT scanner from General Electric Healthcare, in the case of ENA, a second CT scan with intravenous contrast was performed. After scanning, images were analysed using RadiAnt DICOM Viewer 4.6.9 with bone and soft tissue filters, and airway algorithm, and reports were generated.

In CPR, an increase in tissue density was observed in one or both ventral nasal turbinates. In mild cases, slight inflammation is usually appreciated, which gradually increases until it occupies the entire nasal cavity, compressing adjacent structures. Applying the airway algorithm, which displays blue airflow, it was observed that the absence of air in the cavity hindered breathing flow. The ENA cases showed a proliferation of soft tissue, observed as a denser tissue enhancing post-contrast located at the ethmoidal turbinate level, which can be unilateral or bilateral. In severe cases, this tissue caused lysis and destruction of adjacent bones, as well as exophthalmos, as a consequence of the displacement produced by the tumour on nearby structures. With the airway algorithm, it was observed that the space that should be occupied by air appears absent or reduced due to tumour proliferation. The image observed in cases of obstructive rhinopathy showed an increase in tissue density bilaterally located only at the most rostral level of the nasal cavity, the alar portion. When applying the airway algorithm, the absence of air presence was also observed. Finally, clear images of oestrus larvae can only be obtained in the final stage (L3), being less evident in earlier stages.

CT stands out for its ability to create 3D reconstructions and visualise airways with different filters or algorithm, improving the assessment of lesion severity. Although it offers advantages, such as better tissue visualisation and avoidance of structure overlap, it has disadvantages such as the need for anaesthesia and high costs. Currently, its use in sheep is primarily limited to research, but it could become a more common diagnostic tool in farm veterinary practice in the future, underscoring the importance of understanding its interpretation.





# Condemnations of lamb carcasses due to jaundice. What could the seasonal increase of recent years be associated with? (Héctor Ruiz)

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The genus *Anaplasma* is becoming an increasingly important pathogen in Europe. Specifically, ovine anaplasmosis, caused by *Anaplasma ovis*, is a concern in the Mediterranean basin. Until 2020, ovine anaplasmosis had only been diagnosed in adult sheep, causing nonspecific clinical signs. However, in 2020, an outbreak of ovine anaplasmosis was diagnosed for the first time in fattening lambs. During this outbreak, after the slaughter of 462 lambs from different affected farms, 34.84% were condemned due to a marked jaundice in the carcasses. Although the affected animals did not show clinical signs prior to slaughter, blood was taken, and severe regenerative anaemia, as well as a positive PCR with a high bacterial load of *A. ovis* was determined. This situation repeated the following year, with a jaundiced condemnation rate reaching nearly 40% in some batches of lambs of affected farms between May and August.

In order to assess the real incidence of condemnations due to jaundice, data from 2 fattening farms were analysed for the period 2018-2023. This involved the analysis of a total of 479,610 lambs, with carcasses weighing between 8 and 12.5 kg and aged between 2 and 3 months. The data were analysed individually by fattening farm, and seasonal factors were studied monthly. The criteria analysed were feedlot, slaughter date, condemnation or not, and cause. IBM SPSS Statistics v.24 software (IBM, United States) was used for the statistical analysis.

Out of the 479,610 lambs analysed, 945 (0.20%) were condemned due to jaundice during the studied period. Condemnations due to jaundice were the main cause of carcass condemnation, representing 35.51% of the total of them during the period. Although in 2018, condemnations due to jaundice represented only 14.54% of the total complete condemnations, in 2020, this cause accounted for 43.94%, a trend that continued in the following years: 55.08% in 2021, 44.94% in 2022, and 38.43% in 2023, being the most important cause of complete carcass condemnations.

Additionally, a clear seasonal factor was observed during the study, with peaks in condemnations between May and August of each year. Significant differences (p <0.05) were observed compared to the rate during the rest of the year, except in 2018, where there was barely an increase throughout the year. Similarly, significant differences were observed between feedlots. In feedlot A, carcass condemnations due to jaundice represented 0.27% of slaughtered lambs, while in feedlot B, they accounted for 0.16%, resulting in 1.67 times more risk of condemned carcasses due to jaundice in A than in B (p< 0.001). However, the importance of jaundice as a cause of carcass condemnation behaved inversely. In feedlot A, it represented 25.54%, while in B, it was 52.06% (RR: 3.17; p <0.001).

Although molecular studies to detect the presence of *A. ovis* in lambs were not possible, the seasonal increase in condemnations due to jaundice during spring and summer seems to be associated with peaks of maximum tick activity.





#### Poster session 3

Classical Scrapie in sheep: possible diagnosis *intra-vitam* through the detection of pathological prion marker in milk samples by RT-QuIC Assay (Alessandra Favole)

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Classical Scrapie in sheep and goats, like bovine spongiform encephalopathy (BSE) and human Creutzfeldt–Jakob disease (CJD), belongs to the transmissible spongiform encephalopathies (TSEs), a group of fatal and neurodegenerative disorders, also defined as "prion diseases". These diseases are characterized by the conversion of the normal prion protein (PrPC) into a partially protease-resistant isoform (PrPSc), which is therefore used as a diagnostic marker for the disease. PrPSc is widely distributed throughout the central nervous system (CNS), lymphoreticular system (LRS) tissues, and body fluids in both clinically and preclinically Scrapie-affected sheep.

One of the major challenges in managing TSEs in animals is the lack of validated and sensitive *intra-vitam* assays. The high sensitivity of Real-Time Quaking-Induced Conversion (RT-QuIC) in amplifying PrPSc across various biological matrices suggests that it is an ideal method for this purpose. However, it remains ineffective on blood and impractical for cerebrospinal fluid (CSF) due to collection challenges at the flock level.

Given the established involvement of milk in prion transmission, the study aimed to develop RT-QuIC conditions for the detection of PrPSc in milk obtained from naturally Scrapie-infected sheep.

The analysis demonstrated that two rPrP substrates (rHa 90-231 and BV 23-231) can sensitively detect Scrapie PrPSc spiked in diluted milk. Specifically, rHa PrP 90-231 exhibited rapid reactions with lag phases comparable to reactions seeded with Scrapie brain homogenates. Furthermore, the precipitation protocol using an isopropanol/butanol solution enabled the detection of seeding activity associated with the presence of PrPSc in RT-QuIC tests, with a latency phase of 20-30 hours when applied to 10 mL of individual milk samples collected from 2/2 Scrapie naturally infected sheep.

These data confirm the secretion of prions within milk during the early stages of disease progression and a role for milk in prion transmission. Furthermore, the application of RT-QuIC to milk samples offers a non-invasive methodology to detect scrapie during preclinical/subclinical disease.





#### An Investigation into a Classical Scrapie outbreak in the United Kingdom in 2019 (Katrina McCrory)

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Classical Scrapie (CS) is a Transmissible Spongiform Encephalopathy affecting sheep and goats. CS is caused by PrPSc, which are a misfolded conformer of the natural prion protein (PrPC). The PrPC sequence modulates the susceptibility or resistance of sheep to CS, specifically the codons in positions 136, 154 and 171 where the following haplotypes have been identified: ARR, AHQ, ARH, ARQ and VRQ. The ARR haplotype confers resistance whilst the VRQ haplotype confers susceptibility to CS. The approach that was adopted in the UK under the National Scrapie Plan (NSP) was to breed for resistant genotypes (ARR/ARR, ARR/ARQ, ARR/AHQ and ARR/ARH) and to eliminate the most susceptible ones (ARQ/VRQ and VRQ/VRQ). The NSP was implemented from 2001 to 2009. This greatly reduced the prevalence of CS in the UK. Genotypes were assigned an 'NSP Risk Type' on a scale from 1-5, where 1 was least susceptible to CS and 5 was most susceptible.

Here we report a case of a CS outbreak in 2019, after the measures were implemented, in a flock with no previous history of CS. CS was detected in a fallen stock sheep following statutory testing (ELISA, Immunohistochemistry and Western Blot analysis), and retrospective farm enquiry showed CS clinical signs noted by the farmers, including tremors, bruxism, incoordination, loss of body condition and separation from the flock. The PrP genotype of that case was ARH/VRQ.

The owners opted to have their flock culled, with the medulla at the level of the obex being sampled for statutory BioRad ELISA testing in all animals, totalling 827. Additionally, 365 heads were harvested from licensed abattoirs and sent to APHA Weybridge for additional sampling of lymphoreticular tissues (retro pharyngeal lymph nodes, palatine tonsils and nictitating membranes), and genotyping using brainstem material. These included animals aged between 1 and 12 years. After the initial positive, a further 8 animals were diagnosed with CS (4 from the group that had the head sampled, 1 brought in as fallen stock to a regional laboratory, and 3 found positive from the statutory testing of the caudal medulla after slaughtering at a licenced abattoir), seven of these were ARQ/VRQ, and one was ARH/VRQ. The genotypes of the remaining CS negative animals in the cull included also NSP risk group 4 and 5 genotypes, particularly 1 VRQ homozygous, 54 VRQ heterozygous and 39 ARQ/ARQ sheep.

The way CS was introduced in the farm is unclear, as before the index case there had been no reports of CS in the premises dating back to 1993. However, although most of the replacement animals were bred on farm, there were occasions where animals were bought from markets or strayed from nearby farms into the flock. Regardless, the lack of genetic selection for CS resistant genotypes allowed a progressive increase in the prevalence of susceptible genotypes, which caused the spread of the causative agent of CS once a susceptible animal was infected with it, leading to an outbreak of the disease.

Mouse bioassays have been initiated to identify the CS prions strain(s) and are ongoing.





#### Preclinical diagnosis of classical Scrapie PrPSc in ARQ/ARQ experimental sheep (Ledi Pite)

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Classical Scrapie is a chronic, neurodegenerative, fatal disease of sheep and goats caused by prion protein Scrapie (PrPSc). The study aimed to provide an understanding of the preclinical diagnosis of classical Scrapie in ARQ/ARQ experimental sheep, which could contribute to the control of the disease spread and the potential zoonotic risk, acquiring strategic importance of animal and public health.

Fourteen ovine aged 14 days of ARQ/ARQ genotypes were included in a two-year pilot study. Twelve animals were challenged orally with ovine brain tissue homogenate positive for classical Scrapie by Western Blot technique. Two animals were kept as a control group. The ovine were subjected to a monthly blood sampling, and to a cerebrospinal fluid sampling every three months. In addition, an exhaustive clinical and neurological examination was performed until the clinical disease endpoint. The serum samples were tested with the ultra-sensitive Single Molecule Array (SiMoA). The cerebrospinal fluid (CSF) samples were tested with the ultra-sensitive Single Molecule Array (SiMoA) and the Real-Time Quaking-Induced Conversion (RT-QuIC). Medulla oblongata was tested with IDEXX HerdChek BSE-Scrapie Rapid Test, and the nervous tissues were tested with TeSeE™ Western Blot (TeSeE™ WB) and Immunohistochemical method (IHC) at the clinical disease endpoint.

Multilevel regression models explored the relationship between the concentration of Neurofilament Light protein (NfL) in the serum and CSF detected by SiMoA since the challenge time. Regardless of the challenging condition, the physiological effect of the disease appears non-negligible. The NfL concentration in CSF in the Scrapie group is statistically significant, with an increasing trend up to 21 months of incubation, after which there is a non-significant decreasing trend. The Receiver Operating Characteristic Curves (ROCs) that classify the animal as positive or negative from the infection, based on the experimental challenge condition as a gold standard, identified an adequate NfL concentration cut-off value for CSF. The correctly classified samples are 91.67% for the NfL concentration ≥ 318 pg/mL with Se=100% and Sp=50%, AUC=55%. The CSF of two out of six animals in the Scrapie group tested positive using RT-QuIC at a late incubation stage before the clinical disease endpoint. The eight animals that reached the clinical disease endpoint were positive by IDEXX HerdChek BSE-Scrapie Rapid Test, TeSeE™ WB, and IHC. After the challenge, they developed Scrapie typical clinical signs at a mean of 729 days (N = 8, range 655–793 days). The six animals who died of other causes at different times after the challenge (mean 144 days, range 34 – 290 days) were negative by IDEXX HerdChek BSE-Scrapie Rapid Test, TeSeE™ WB, and IHC.

The preclinical diagnosis of PrP Sc remains to be further explored in order to contribute to the disease surveillance and control.

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### Smart grazing – The use of sheep as "natural weed controller" in growing maize (Johanna Meilwes)

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Introduction. In many countries sheep are part of agricultural crop rotation systems. In Germany, the use of sheep in arable farming has become less important.

Especially, organic maize cultivations are challenging for every farmer as the use of herbicides is not allowed. In early vegetation stages a high competition between weeds and maize is a serious problem. Therefore, the most important plant protection measure is mechanical weeding like weed harrowing and row hoeing. But after row closing (80cm growth height) it is not possible to drive through the maize fields by machines anymore.

In this Project sheep were used at this point to continue the weed control in the growing maize culture.

Material and Methods. In two consecutive years (2021/2022) sheep were kept in three different maize fields in Hannover. In each of these fields there were two grazing areas of 50x50 metres. During the "grazing" period the areas were inspected weekly, and a botanical evaluation documented which plants were fed and whether and how the maize was damaged by the sheep. The areas grazed by the sheep were compared to ungrazed areas.

Results. Sheep grazing behaviour varied in the different fields which showed a very varied weed growth in mass and in diversity.

Main weeds were: Echinochloa crus-galli, Chenopodium album, Persicaria maculosa, Datura, Solanum nigrum and Fallopia convolvulus. The sheep mainly showed an intake of: Echinochloa crus-galli, Chenopodium album and different kinds of Persicaria maculosa.

The growth of weed between the rows was not only reduced by grazing. Additionally, weeds were reduced as the sheep trampled the plants growing there.

Sheep only showed interest in the lowest maize leaves, but not in leaves or cobs above their nose level (110 cm). Single maize plants were bent by the movement of the sheep.

Conclusion. Weed control only worked when weed diversity was high and the plants nutritionally attractive. When the number of weeds was low, weed control failed and maize plants became attractive to the sheep. In this case there was considerable damage of maize culture.

The eating of the lower leaves is not a problem for maize plants, it might even have a positive effect. In other botanical species, the removal of lower leaves is common because it promotes the so-called apical dominance and increases the productivity of the upper plant parts.

The sheep showed a good general state of health and increasing weight.

It seems that the use of sheep as a "natural weed controller" could be an interesting approach in sustainable farming. First field studies reveal promising results. The effectiveness with regard to weed elimination while sparing the corn crop depends on various factors. More field studies are needed to evaluate these factors.

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#### Seroepidemiological study of small ruminant lentivirus in 3 regions of Portugal (Delia Lacasta)

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Introduction. Small ruminant lentivirus disease (SRLV) is a disease that affects sheep and goats, formerly known as Maedi-Visna and Caprine Arthritis and Encephalitis, respectively. Several studies have shown that SRLVs are present all over the world. This is a persistent infection, causing chronic, insidious and progressive disease. For diagnosis, laboratory methods are essential. In Portugal, the production of small ruminants is of economic and social importance, and there are very different production systems. The objective of this work was to study the seroprevalence of SRLV and the associated risk factors in 3 regions of Portugal.

Material and methods. This SRLV seroprevalence study was carried out in sheep and goat herds from 3 different regions of Portugal (Trás-os-Montes; Centro; Alentejo). Herds were randomly selected, and producers were invited to enroll in the study. Based on the total number of animals in each herd, 14 to 19 blood samples were randomly collected from different animals. A commercial indirect ELISA test (ID Screen® MVV/CAEV Indirect) was performed on each sample with the aim of detecting the infection. Each herd was considered positive with at least one seropositive animal.

Results. We obtained samples from 94 herds, of which 35 herds were from Trás-os-Montes region, 28 from the Central region, and 31 from the Alentejo region. Regarding the laboratory results obtained, the seroprevalence was 67.02%. By region, 30 (85.71%) positive herds were obtained in Trás-os-Montes region, 23 (82.14%) in the Central region, and 10 (32.26%) in the Alentejo region.

Finally, and in relation to the results obtained per animal, we collected 1653 samples, of which 606 were in the Trásos-Montes region, 494 were in the Central region and 553 were in the Alentejo region. The individual seroprevalence was 36.12%. By region, 257 (42.41%) positive samples were obtained in Trás-os-Montes region, 231 (46.76%) in the Central region and 109 (19.71) in the Alentejo region.

Conclusions. We concluded that there is a high seroprevalence of SRLV in herds of small ruminants in Portugal. The high individual and herd seroprevalence in Trás-os-Montes and Central regions stands out in comparison to the much lower results obtained in the Alentejo region. This fact may be due to the predominance of the extensive production method in the latter region. This reinforces the importance of carrying out serological screening tests for the early detection of diseases in small ruminant herds. Carrying out epidemiological questionnaires with the aim of identifying possible risk factors is also a key factor. This knowledge allows the implementation of effective preventive measures. Biosafety measures must be promoted and implemented to reduce viral transmission, with a view to reducing the prevalence of this disease. We understand that, in this sense, government authorities must promote and audit voluntary control and eradication programs to eradicate this disease in sheep herds in all the regions of Portugal.





#### Long lasting infection with Anaplasma ovis in sheep (Héctor Ruiz)

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Ovine anaplasmosis is an emerging vector-borne disease in Europe caused by *Anaplasma ovis*. The infection has spread quickly in recent years, causing moderate to severe outbreaks in sheep flocks. Ovine anaplasmosis is characterised by unspecific clinical signs such as weakness, anorexia, weight loss, anaemia and pallor of mucous membranes. However, sometimes *A. ovis* is silently transmitted without causing apparent clinical signs.

The disease has quickly spread in Europe, as has been demonstrated by the high number of recent outbreaks reported. This spread has been associated with global warming and climate change, which favour tick's life cycle. However, the long persistence of *A. ovis* infection has also been hypothesised as another fundamental aspect, although it has never been scientifically proven. The present work intends to analyse whether animals infected by *A. ovis* are really permanently infected if proper antibiotic treatment is not applied. In order to demonstrate this hypothesis, 11 ewes were analysed during their whole productive life (between 4 to 6 years of life).

Eight out of eleven of the analysed animals were *A. ovis*-positive at the beginning of the experiment, either by experimental or natural infection. Between 2015 and 2017, five of these animals were intravenously inoculated with a single dose of 30ml of whole blood mixed of two naturally infected ewes. Simultaneously, three positive animals belonging to naturally infected herds close to our facilities in Zaragoza were included in the study to follow up on the development of the natural infection. In addition, three clinically healthy and *A. ovis*-negative ewes of the faculty flock were included as a control group. All the animals were kept indoors throughout the experiment, without access to pasture, living with other teaching flock animals in the Zaragoza Veterinary Faculty facilities. No ticks were detected in the animals throughout the experiment.

Clinical follow-up was performed monthly on all animals throughout the duration of the study (between 4 to 6 years), and blood samples with anticoagulant (EDTA) were collected every six months. The specific detection of *A. ovis* was carried out by using the commercial kit EXOone *Anaplasma ovis* (EXOPOL S.L.).

All animals that started the study being *A. ovis* positive continued to be positive until the end of the experiment, and all the negative animals were negative during the analysed period. Initially, the ewes showed clinical signs. However, throughout the duration of the study did not show them again. No clinical signs associated with ovine anaplasmosis were observed in the animals during the study. Different degrees of fluctuation were observed, both in naturally and experimentally infected animals, showing the development of the bacterial load throughout the analysed period.

Our results indicated that animals affected by ovine anaplasmosis can be permanently infected without demonstrating clinical signs for up to 6 years. In natural conditions, these animals can act as reservoirs of the disease, favouring the spread of the infection. However, this aspect can help to reach endemicity in the flock due to the natural immunity and the high intraherd-prevalence levels.





#### Minisatellites analysis of MAP field isolates from goats in Northern Italy (Anita Filippi)

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Background. Paratuberculosis is a chronic incurable bacterial infection diffused worldwide in domestic and wild ruminants. The disease is caused by *Mycobacterium avium* subspecies *paratuberculosis* (MAP) and has a deep impact on animal welfare and production, and it has also some impact on public health due to its potential role as a zoonotic agent. Paratuberculosis has been included in the new European animal health legislation as a disease for which surveillance in ruminants is mandatory. Recently, the Italian Reference Centre for Paratuberculosis, according to the Italian Ministry of Health, updated the Guidelines for the adoption of Control Plans and for the assignment of health status to herds by including small ruminants (goats and sheep).

Notably, goats appear to be particularly susceptible to paratuberculosis and the detection and control of the disease is crucial to avoid the introduction and the spread in flocks, particularly in some developing countries and other intensive goat breeding areas.

The diffusion of the disease appears to be underestimated in many countries, but paratuberculosis in goats has been world widely reported. In Europe, was reported an inter-herd prevalence of infected goat herds over 20%, confirmed as well by Italian available surveys. Understanding the genetic variability of MAP field isolates is a key point in the epidemiological investigation so, aim of this study, was to explore the variability of MAP field isolates from goats in Northern Italy by minisatellite analysis, in order to check if there are difference among the MAP isolates circulating in goats. This method doesn't allow reaching very powerful discriminating indexes but provide useful information with the advantage of being cheap.

Methods. The characteristics of MAP field isolates from goats were investigated via a combination of eight Minisatellite loci (VNTR X3, VNTR 3, VNTR 7, VNTR 10, VNTR 25, VNTR 32, VNTR 47, VNTR 292), according to a scheme already proposed. Isolates were from the same macro area (Northern Italy) and have been isolates from 2015 to 2019.

Results. All 23 MAP goat isolates typed belonged to the Type II, the type most diffused in cattle and in other animals and already reported circulating in the Northern Italy.

Based on minisatellite loci, two main allelic profiles have emerged: 18 isolates belonged to INMV1, which is one of the most diffused profiles worldwide, both in cattle and in other species; while 3 isolates belonged to INMV2, another subtype that is widespread worldwide; finally, the other two isolates belonged to INMV3 and INMV5 subtype, already described in the Alps.

Conclusion. Overall, our data underlined a certain degree of heterogenicity among the MAP populations circulating in Italian goats; a more powerful investigation by WGS approach could elucidate the dynamic and origins of infection, data which are pivotal for the implementation of strategies for prevention and control of the disease.