UNIVERSITY of GUELPH
LABORATORY SERVICES

Animal Health Laboratory

AHL Newsletter

Canada Post Publications number - 40064673

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In this issue:

Avian bornavirus and proventricular dilation disease

Background Recent findings suggest association of *Avian bornavirus* (ABV) with proventricular dilation disease (PDD). Until now, the diagnosis of PDD could only be confirmed post-mortem, and there has been no easy way to test living birds. It appears that some parrots are 'carriers' of the disease and do not show clinical signs, but can 'shed' the virus to other birds who may become seriously ill. Thanks to collaboration of researchers from the University of California, San Francisco, with the Ontario Veterinary College and the Animal Health Laboratory, **a new RT-PCR screening test that detects ABV genetic material is now available in the Animal Health Laboratory.**

Sample collection and transport

- Collect cloacal or choanal swabs (combined cloacal/choanal swabs are acceptable) in **virus transport medium**. We recommend testing of individual birds.
- Please do not send feces, blood, serum or plasma.
- If samples can reach the lab within 24-48 hours, ship samples chilled (on ice packs) to the lab, otherwise freeze and send later.
- Swabs with virus transport medium can be obtained from commercial suppliers and are also available through the Animal Health Laboratory.
- Please note that shedding may be intermittent, so repeated testing will increase the confidence in a negative status.
- Tests will be run batched, once a week.

For submission forms and shipping instructions, please see the AHL website:
www.ahl.uoguelph.ca

Dr Jim Fairles, Client Services Veterinarian, 519-824-4120 ext 54530 ahlinfo@uoguelph.ca

Removing the tower crane Oct 31- getting closer to the move Aug/2010!

AHL Christmas hours, 2009

- Limited testing services 0900-1700, Dec 24, 29, 30, 31, in both Guelph and Kemptville. Normal services resume Jan 2/10
- Emergency mammalian necropsy and Specimen Reception are available in the Guelph lab on weekends and holidays except for Christmas Day, Dec. 25 (lab is closed) - our specimen drop box is available 24/7.
- Please call if you would like to schedule testing over the holiday period.

Season's Greetings from the staff of the Animal Health Laboratory

Tick ID

The AHL Parasitology lab is able to identify *Ixodes scapularis* (deer tick or blacklegged tick). Suspected ticks can be submitted in a container with ethanol or water (to prevent them from drying out). Please specify if the test is for parasite identification (test code idpr).

For further information, please contact the AHL Parasitology lab, 519 824-4120, ext 54522.

AHL introduces new submission forms!

Our new NCR (no carbon required) submission forms are now being distributed. When you receive the new forms, please discard any old AHL forms that may still be in your clinic.

- All our forms are now one-sided. Details of common tests are available on the back of the form. If a test is not seen on the form, please refer to our User's Guide and Fee Schedule. The Fee Schedule (password protected under "Client login"– call or email for details) and new forms are on our website at: http://www.ahl.uoguelph.ca under "Where and how to submit specimens".
- You will now be able to keep an exact NCR copy of the submission form at your clinic. If you do not need or wish NCR forms, forms are available in a one-page format.
- Because the forms are NCR, we will be using a clinic sticker system in place of custom forms. For those who use customized forms, you should receive sheets of stickers with your forms, with the clinic #, address, phone, fax number included (and veterinarian if desired). Please fill in the veterinarian's name, and any additional information required below the sticker, directly on the form.
- For our large animal practitioners, please fill out any demographic information, including farm municipality, postal code, and/or CCS #. **Demographic information is becoming increasingly important in the surveillance of animal disease**.
- **Owner Unique ID is a** consistent and unique string of letters and numbers to allow computerized cross-referencing of case reports by submitting veterinarians and AHL data systems. **For client accounting and reconciling purposes this is the only identifier passed from the report to the invoice.**

Please feel free to give us your input on the new submission forms as we are trying to make them as user friendly as possible. **Please ask us about customized forms, online access to results, and online submissions as well.** *AHL*

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Its mission is to inform AHL clients and partners about AHL current activities, and laboratory-based animal disease events and disease trends. *Ideas and opinions expressed herein do not necessarily reflect the opinions of the University or the editor.*

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Our continued thanks to all of the non-author AHL clerical, technical, and professional staff who contribute to the generation of results reported in the AHL Newsletter.

AHL immunohistochemistry tests, fall, 2009

Josepha DeLay

Infectious agents

Bovine herpesvirus-1 (BoHV-1, IBRV)
Bovine respiratory syncytial virus (BRSV)
Bovine viral diarrhea virus (BVDV)
Coronavirus:
- Bovine
- Feline (enteric and FIP)
- Canine
- Porcine (TGEV)

Canine distemper virus
Canine parvovirus
Chlamydophila spp
Equine herpesvirus 1 (EHV-1)
Feline panleukopenia virus
Influenza A virus
Leptospira spp
Mycoplasma bovis
Neospora caninum
Porcine circovirus 2 (PCV-2)
Porcine reproductive and respiratory syndrome virus (PRRSV)
Transmissible gastroenteritis virus (TGEV)
Toxoplasma gondii
West Nile virus (polyclonal / monoclonal)

Infectious agent panels

Porcine respiratory panel:
PRRSV, PCV-2, Influenza A virus
Bovine abortion panel:
BVDV, IBRV, *Leptospira* spp

Cell markers

Actin: smooth muscle actin, muscle actin
Calcitonin
CD3
CD79α
CD18 (canine, feline)
CD31
CD61
CK117 (c-kit)
Chromogranin
Cytokeratins: pan CK (AE1/AE3), CK7, CK20, HMW CK
Desmin
Factor VIII-related antigen
GFAP
Hepatocyte paraffin antigen (HepPar 1)
Ki-67
Lambda light chains
Mast cell tryptase
Melan A
MHC II
Neuron-specific enolase (NSE)
PCNA
S100
Synaptophysin
Thyroglobulin
Thyroid transcription factor (TTF)
Uroplakin
Vimentin

For additional information or questions, please contact Dr. Josepha DeLay at jdelay@uoguelph.ca or 519-824-4120 ext. 54576.

Bovine respiratory syncytial virus IHC now available

Josepha DeLay, Murray Hazlett

Testing for BRSV in formalin-fixed lung samples by immunohistochemistry (IHC) is now available at the AHL (Figure 1). Isolation of BRSV and other paramyxoviruses in cell culture is difficult, and diagnosis of BRSV-associated pneumonia in cattle relies on identification of compatible histologic lesions in conjunction with demonstration of viral antigen in tissue. Immunofluorescence (IF) testing for BRSV using frozen lung samples has been offered by the AHL for a number of years. **The new IHC test for this virus is performed using formalin-fixed lung samples, which has the advantage of ease of sample submission for cases shipped to our laboratory by courier**. In a recent validation study, there was 100% agreement between results of IF and IHC tests for BRSV on cattle lung samples.

There are other advantages of IHC over IF testing. The pathologist will often see several areas of lung when first examining the slides and based on lesions present in the different sections, can select the area that is most likely to demonstrate virus. Additionally, the test can be ordered as an afterthought when BRSV was not a consideration in the original submission, long after a case has been finalized if needed. This advantage of retrospective testing also applies to IHC available for other bovine viruses such as *Bovine herpesvirus 1* (IBRV), BVDV, and *Bovine coronavirus*.

BRSV most commonly causes pneumonia in both beef and dairy calves up to 5 months of age as well as in feedlot animals, although cattle of any age may be affected. Between May 2007 and October 2009, 32 cases of bovine pneumonia diagnosed at the AHL had histologic lesions compatible with BRSV infection. BRSV infection was confirmed in 9 of the animals by IF or (more recently) IHC. The remaining 23 cases had either negative test results or did not have fresh / frozen tissues included in the submission for IF testing. Because viral antigen is often detected only in early stages of infection, involvement of BRSV in cases of bovine pneumonia with compatible histologic lesions cannot be completely excluded on the basis of negative test results alone…. *AHL*

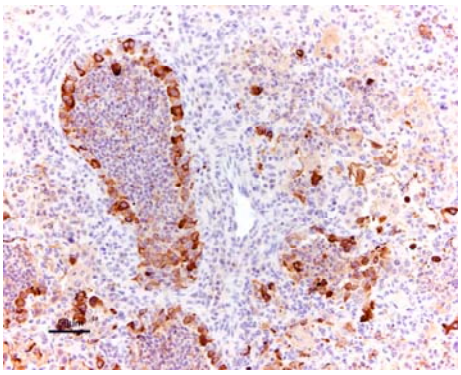

Figure 1. Immunohistochemistry for BRSV antigen, bovine lung. Brown staining indicates the presence of viral antigen in bronchiolar epithelium and in scattered alveolar epithelial cells.

AHL Lab Reports

RUMINANTS

AHL sheep PrP genotyping update

Hugh Cai, Patricia Bell-Rogers, Rebeccah Travis, Jim Fairles

It is well known that the polymorphism (genotype) of the sheep prion protein gene is linked to the resistance of scrapie. The National Scrapie Plan (NSP) for Great Britain listed the following genotypes and their resistance to scrapie (Table 1).

Table 1: PrP genotypes and their resistance to scrapie; adapted from http://svs.mri.sari.ac.uk/nsp.htm

PrP genotypes	Degree of resistance to scrapie	NSP risk group
ARR/ARR	Most resistant	1
ARR/AHQ, ARR/ARH, ARR/ARQ	Resistant	2
ARQ/ARH, ARQ/AHQ, AHQ/AHQ, ARH/ARH, AHQ/ARH, ARQ/ARQ	Have little resistance	3
ARR/VRQ	Susceptible	4
AHQ/VRQ, ARH/VRQ, ARQ/VRQ, VRQ/VRQ	Highly susceptible	5

With the support of the National Survey of Scrapie Genetics in Canadian Purebred Sheep program, the AHL tested 2,368 blood samples from Ontario between December 2005 and December 2008. Our results showed that 69.1% of the genotypes belonged to the NSP type 1 or 2 (resistance) and only 5.4% were NSP type 4 or 5 (susceptible) (Table 2).

Our results also showed that in the last 3 years, the trend in the resistant genotypes (NSP type 1 or 2) in Ontario increased, while the trend in the genotypes with little resistance (NSP type 3) decreased (Table 3). This is likely in part due to breeding selection by sheep producers using the genotyping results.

Although the National Survey of Scrapie Genetics in Canadian Purebred Sheep program has ended, the AHL continues to provide PrP genotyping services. In addition to breed selection and other applications, sheep producers can use the genotyping results to participate in the Scrapie Flock Certification Program (SFCP) administrated by the CFIA. "The objective of the SFCP is to provide owners with the opportunity to have their flocks/herds identified as negligible risk with respect to scrapie. Membership in the certification program provides assurances to potential purchasers of animals that a flock/herd is not infected with scrapie. Export certification of scrapie status for countries with higher zoosanitary requirements for scrapie may be based on enrolment and activities under the SFCP." The statement and more details on SFCP can be found in the CFIA website: http://www.inspection.gc.ca/english/anima/heasan/man/avmmva/avmmva_mod7e.shtml. *AHL*

Table 2. Ontario sheep PrP genotypes

PrP genotype	NSP type	%
ARR/ARQ	2	41.6
ARR/ARR	1	23.9
ARQ/ARQ	3	22.8
ARR/VRQ	4	2.9
ARQ/VRQ	5	2.4
ARR/AHQ	2	1.9
ARR/ARH	2	1.7
ARQ/AHQ	3	0.9
ARQ/ARH	3	0.8
AHQ/AHQ	3	0.6
ARH/ARH	3	0.2
AHQ/ARH	3	0.2
VRQ/VRQ	5	0.1
ARR/AHR	5	0.04
ARR/ARH	2	0.04

Table 3. Changes of major Ontario sheep PrP genotypes from 2006 to 2008 (%)

PrP genotype	NSP type	2006	2007	2008
ARR/ARQ	2	39.0	41.3	42.3
ARR/ARR	1	16.6	26.1	30.2
ARQ/ARQ	3	32.7	27.8	17

Role of *Chlamydophila* spp and *Coxiella burnetii* in caprine and ovine abortions?

For the present, and expected to last until May 2010, **necropsies on aborted lambs and goat kids will be paid for with Animal Health Strategic Investment project funding**, and there will be **no charge to the owner or veterinarian provided the placenta is submitted, either with the fetus (preferable) or alone**. Fetuses submitted without placenta will incur normal charges as listed in the AHL Fee Schedule. *AHL*

Cuffing pneumonia in a calf associated with *Ureaplasma diversum*

Brian Binnington, Tony van Dreumel, Murray Hazlett, Brian MacNaughton

Respiratory disease was an ongoing problem for a year in young Ayrshire heifer calves housed in a barn with older cattle. The herd had been vaccinated with a 10-way modified-live virus and leptospiral vaccine that the calves received at 6 to 8 weeks of age. Vaccinations and antimicrobial treatments were having a limited effect on the disease problem. A 1-day-old bull calf from this herd was colostrum fed but not vaccinated and it was placed with the heifers as a "sentinel" calf. At 26 days of age, this calf was coughing sporadically and at 32 days of age it had wheezing and fluid lung sounds. The calf was euthanized and necropsied.

Portions of the cranial, middle and cranioventral third of the caudal lung lobes had dark red, atelectatic and moderately firm areas. Microscopically, lobules were atelectatic with accumulations of neutrophils and small numbers of macrophages in bronchioles and alveoli. Hypertrophy of bronchiolar epithelium and goblet cells was present in some bronchioles and bronchi. There were prominent accumulations of lymphocytes and small numbers of plasma cells around bronchi, bronchioles, blood vessels and in the mucosa of bronchi (Figure 1). Moderate lymphoplasmacytic tracheitis and rhinitis were present.

No mycoplasma could be isolated, but large numbers of *Ureaplasma diversum* and a few *Pasteurella multocida* were isolated from lung samples. No virus was isolated from lung, thymus, spleen, bronchial lymph node or bronchial and nasal swabs. *Bovine respiratory syncytial virus* and *Bovine parainfluenza virus 3* could not be demonstrated in lung sections by IHC and FA testing.

The term "cuffing pneumonia" has been used to describe bronchopneumonia with prominent expansive peribronchial and peribronchiolar lymphoid hyperplasia. *Mycoplasma bovis* is considered to be a primary bovine lung pathogen, whereas the roles of other *Mycoplasma spp.* and *Ureaplasma diversum* in bovine pneumonia are less certain. Although *Mycoplasma spp.* are more frequently present in cuffing pneumonia, *Ureaplasma diversum* has been demonstrated in pneumonic calves and the experimental inoculation of specific-pathogen-free calves produced histological lesions that are similar to the findings in this sentinel bull calf. *Mycoplasma spp. and Ureaplasma diversum* infections can predispose to bacterial pneumonia with opportunistic bacteria such as *Pasteurella multocida* that was isolated from the lung of this calf.

The inability to demonstrate other significant respiratory viruses, mycoplasmas or bacteria suggests that *Ureaplasma diversum* was a significant pathogen in this sentinel calf. Whether *Ureaplasma diversum* is associated with the ongoing respiratory disease in the herd and to what extent is unclear. *AHL*

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Pirie HM, et al. Mycoplasmas and cuffing pneumonia in a group of calves. Vet Rec 1975;97:345-349.

ter Laak EA, et al. Comparison of pathological signs of disease in specific-pathogen-free calves after inoculation of the respiratory tract with *Ureaplasma diversum* or *Mycoplasma* canis. J Comp Pathol 1993;108:121-132.

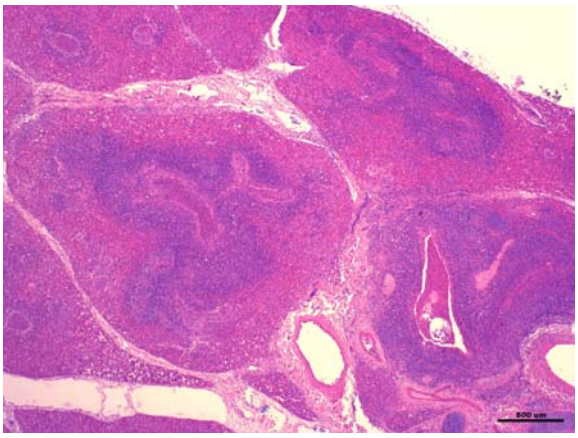

Figure 1. Prominent peribronchiolar lymphocyte accumulations with broncho-alveolitis and atelectasis (H&E)

SWINE

Gastritis in young pigs – a new lesion?

Josepha DeLay, Beverly McEwen

AHL pathologists have identified a recent increase in histologic lesions of gastric inflammation and erosion in young pigs. In a review of porcine cases submitted to the AHL between May 2007 and October 2009, pigs 8 weeks of age or younger were 3.9 times (95% CI, 2.3, 7.6) more likely to have gastric lesions than older pigs. Despite a wide range of testing for infectious agents, **no common etiology has been identified to date**.

Retrospective analysis of data from these cases is underway to correlate age, history, and concurrent disease processes with the gastric lesions. **Practitioners are encouraged to include stomach among formalin-fixed tissues submitted to the AHL for histology from on-farm necropsies**. Updates on this project will follow in future editions of this newsletter. *AHL*

AVIAN/FUR/EXOTIC SPECIES

Cutaneous spirochetosis due to *Treponema paraluiscuniculi* in a rabbit

Marina Brash, Leah Schutt, Christopher Draban, Patricia Turner

A mature female pet brown rabbit with a history of nasal crusting was presented to the referring veterinarian. On physical examination, a focal area of skin on the nasal planum was dry, red, crusting and hemorrhagic (Figure 1). The rabbit appeared otherwise healthy.

Two small skin biopsies from the nasal lesion were submitted to the Animal Health Laboratory for histologic evaluation. There was moderate acanthosis and orthokeratotic hyperkeratosis of the epidermis, with multifocal areas of erosion and ulceration. Within the stratum spinosum, there were multiple areas of spongiosis and scattered dead keratinocytes (Figure 2). Frequent transmigrating heterophils and occasional serocellular crusts containing abundant heterophils were present on the surface of the epidermis (Figure 3). The superficial and deep dermis was congested, edematous and contained multifocal areas of hemorrhage and abundant infiltrating inflammatory cells, including plasma cells, lymphocytes and fewer heterophils.

Differential diagnoses for ulcerative and scabbing lesions on the nose of a rabbit included cutaneous spirochetosis (rabbit syphilis), dermatophytosis, acariasis, myxomatosis, and trauma. Warthin-Starry silver stain of the skin biopsies showed large numbers of long spiral-shaped organisms consistent with *Treponema paraluiscuniculi* within the intracellular spaces of the epidermis and rarely extending into the dermis (Figure 4), confirming the diagnosis of cutaneous spirochetosis or rabbit syphilis caused by *Treponema paraluiscuniculi*, a gram-negative spirochete.

Characteristic lesions of rabbit syphilis include papules, ulcers, and scabs on the genitalia, perianal skin, nose, mouth, and eyelids. Lesions develop 3-6 wk following exposure and are most prominent on the mucocutaneous junctions of the mouth/face and genitalia. The lesions begin as erythema and edema and progress to papules or vesicles, followed by focal ulceration, crusts, and scaling or hyperkeratosis. Mild or subclinical disease is most common, but *Treponema paraluiscuniculi* infection has also been associated with abortion, metritis, and infertility in female rabbits.

Infection with *Treponema paraluiscuniculi* is common in wild hares from many parts of the world, and occasionally occurs in rabbits produced for research facilities. Venereal transmission is typical, however vertical transmission may also play an important role in the spread of disease in enzootically infected colonies. Introduction of subclinically infected carrier rabbits can establish the disease in breeding colonies. Since this rabbit had had no recent contact with other rabbits and no known history of mating, it was inferred that the infection was contracted vertically.

The appearance and distribution of gross lesions of rabbit syphilis are often diagnostic. Diagnosis of *Treponema sp*. in formalin-fixed tissue sections requires Warthin-Starry silver staining in order to visualize the spirochetes. Anti-*Treponema* antibodies are slow to develop, requiring 5-6 wk from the time lesions appear, suggesting serology is not effective for diagnosing active cases.

Rabbit syphilis tends to be self-limiting, with spontaneous regression of lesions usually occurring within several weeks. Natural infections of laboratory rabbits with *Treponema* have been suggested to potentially interfere with dermal and immune-related study protocols. Untreated rabbits can remain seropositive, suggesting a carrier state, possibly in regional lymph nodes. For these reasons, **treatment is the preferred method of control**. The organism is susceptible to procaine penicillin, 40,000 IU IM administered daily for 5-7 days. In this case, the rabbit was treated by the referring veterinarian with a long acting penicillin (Longisil®, Vetoquinol Canada Inc.) and the gross lesions resolved rapidly. *AHL*

Figure 1. Typical exudative and hemorrhagic lesions can be seen on the genitalia, perianal skin, nose, mouth and eyelids (Photo courtesy of Dr. Michael Taylor, Ontario Veterinary College, University of Guelph).

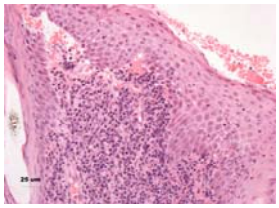

Figure 2. Acanthosis, spongiosis with dermal edema, congestion, focal hemorrhage and lymphoplasmacytic cellular infiltrates.

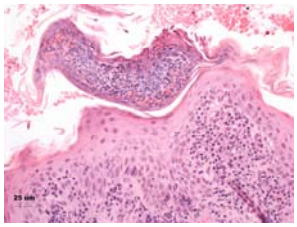

Figure 3: Epidermal acanthosis, orthokeratotic hyperkeratosis and serocellular crusting.

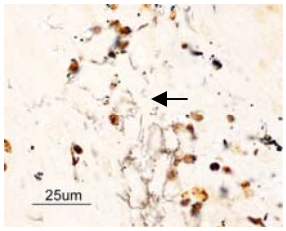

Figure 4. Typical slender long spirochetes (Warthin-Starry stain) are in the epidermis.

HORSES

Alopecia areata like disease in a Belgian horse

Murray Hazlett, Joel Rumney, John Baird

A 3-year-old Belgian stallion was treated initially for a bilateral ocular and nasal discharge with trimethoprim-sulfa for a 2 week period (30mg/kg po SID). Approximately 1 month later, the owner asked that the horse be examined for 1-8 cm irregular coalescing patches of alopecia with hyperpigmentation and scaling that had appeared on the face, neck, and trunk (Figures 1, 2). Mane and tail hairs were unaffected. The horse had some erythema of the anal region and hyperemic buccal mucous membranes. There was a mild bilateral ocular discharge with hyperemia of the conjunctiva. During this time, the stallion was bright and alert, as well as eating and drinking normally, with normal defecation and urination. After another month, skin lesions persisted. Photographs and skin biopsies were submitted to the Animal Health Laboratory for examination.

Microscopically, the most striking change was the presence of large numbers of lymphocytes accompanied by occasional macrophages and eosinophils surrounding the deeper portions of the hair follicles in the affected skin sections (Figure 3). Occasional small accumulations of histiocytes were present and rare multinucleate cells could be found amongst these in foci where there had likely been some disruption of follicles. Hairs in follicles were absent or distorted, and the targeting of the hair bulbs (bulbitis) seemed to be directed at anagen follicles. Mild perivascular mononuclear inflammation was also present in the superficial and middle dermis. The epidermis was mildly hyperplastic with mild orthokeratotic and parakeratotic hyperkeratosis. There was pigmentary incontinence present both in superficial dermis and around occasional follicles. **Lesions seen were felt to be most compatible with a diagnosis of alopecia areata, showing the classical lymphocytic inflammation in and around the hair bulb.**

Alopecia areata is a relatively uncommon disease in horses. In horses, IgG has been shown to be directed toward hair follicle antigens. In some species, including humans, T-cells (CD4 and CD8) are involved. In humans, there is some indication of familial tendency, whereas in horses there is no known breed predisposition.

No curative treatment is currently available. Because of the poor prognosis and the extent of lesions seen, the horse was euthanized. *AHL*

References

Gilhar A, Kalish RS. Alopecia areata: A tissue specific autoimmune disease of the hair follicle. Autoimmunity Reviews 2006;5: 64– 69.

Stannard AA. Alopecia in the horse: an overview. In: Von Tscharner C, Yager JA, Kunkle G. eds. Stannard's Illustrated Equine Dermatology Notes. Veterinary Dermatology 2000;11:191–203.

Tobin DJ, et al. Equine alopecia areata autoantibodies target multiple hair follicle antigens and may alter hair growth. Experimental Dermatol 1998:7;289–297.

Fig 1 – Areas of alopecia around eyes and on face.

Fig 2 – Patchy areas of alopecia on trunk and neck.

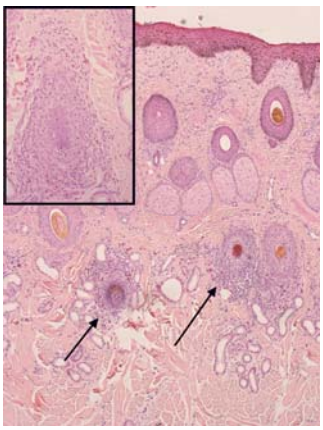

Fig 3 – Histologic section of alopecia areas. Large numbers of inflammatory cells surround follicles, particularly around the bulb (arrows). Close-up of inflamed bulb is shown in inset.

COMPANION ANIMALS

Coagulase-positive *Staphylococcus* spp. in dogs

Durda Slavic

The 2 most frequently recovered coagulase-positive *Staphylococcus* spp. in dogs are *Staphylococcus aureus* and *Staphylococcus pseudintermedius* (formerly *Staphylococcus intermedius*). *S. pseudintermedius* belongs to the recently established *Staphylococcus intermedius* group. Members of this group (*S. pseudintermedius*, *S. intermedius* and *S. delfini*) are very difficult to differentiate biochemically, and additional genetic analysis is needed to classify them to the species level. However, most isolates from dogs are *S. pseudintermedius*, indicating that the source of the isolate can significantly help with the bacterial speciation of *S. intermedius* group.

While *S. pseudintermedius* is a part of the normal bacterial flora of mucosal surfaces in dogs, *S. aureus* is mostly a transient colonizer even though some animals can be permanent carriers. Both bacterial species are also opportunistic pathogens. Pyoderma, otitis, cystitis, wound infections, and surgical site infections are some of the many clinical manifestations caused by *S. pseudintermedius* and less frequently by *S. aureus*. Moreover, there is also a public health concern that is associated with these 2 species. *S. aureus* is a major cause of nosocomial infections in human hospitals, whereas the zoonotic potential of *S. pseudintermedius* is less clear, is most likely underestimated, but is present nonetheless.

In the past, clinical infections of dogs caused by coagulase-positive *Staphylococcus* spp. were relatively simple to treat with β-lactamase-resistant synthetic penicillins and first-generation cephalosporins. This is increasingly changing, however, due to the emergence of isolates resistant to all β-lactam antimicrobials (penicillin, cephalosporins, carbapenems). Coagulase-positive isolates resistant to all β-lactam antimicrobials are classified as methicillin-resistant, either **methicillin-resistant *S. aureus*** (**MRSA**) or **methicillin-resistant *S. pseudintermedius*** (**MRSP**). Both MRSA and MRSP express altered penicillin binding protein (**PBP**) which has low affinity for all β-lactam antimicrobials. PBP is encoded by *mecA* genes located on a staphylococcal chromosomal cassette (SCCmec). This cassette can also carry other resistance genes, or they can be located somewhere else on the chromosome conferring multidrug resistance to some isolates.

When clinical samples are sent to the diagnostic laboratory, the laboratory should be able to differentiate between *S. aureus* and *S. pseudintermidius* (or other coagulase-positive staphylococcal species). **Differentiation between these 2 species is very important for interpretation of susceptibility results for several reasons**. *mecA* gene expression has to be induced in vitro when susceptibility testing is done. Different antimicrobials are used for its expression depending on the bacterial species in question. The use of cefoxitin alone as a predictor of methicillin resistance in *S. pseudintermedius*, more frequently than not, will give false-negative results. Subsequently, the laboratory may report erroneous susceptibility results for some or most antimicrobials belonging to the β-lactam group. In addition, the proper bacterial identification is very important even when the right antimicrobial is used for induction of *mecA*, because the interpretation of the results (S, I, R) will differ depending on the bacterial species.

AHL tests all of clinically significant *Staphylococcus* isolates including *S. pseudintermedius* and *S. aureus* for the presence of the *mecA* gene. **If the *mecA* gene is detected, then all β-lactam antimicrobials are reported as being resistant regardless of in vitro results, since it is well established that clinically they will not be effective.** Therefore to learn if methicillin-resistance is present, a careful look at the susceptibility pattern is warranted.

Currently, the AHL is seeing occasional infections caused by MRSA and increasing numbers of MRSP infections. MRSP isolates can be broadly divided into 2 groups: isolates that are resistant to β-lactam antimicrobials only and isolates that are resistant to other antimicrobial groups such as tetracyclines, potentiated sulfonamides, fluoroquinolones, and some aminoglycosides. Most of the isolates within the second group are still sensitive to amikacin and chloramphenicol. Chloramphenicol-resistant isolates, however, have been detected and it is only a question of time of when resistance to chloramphenicol will become widespread. *AHL*

References

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