

Ophthalmologic findings associated with *Rhodococcus equi* bronchopneumonia in foals

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RUNNING TITLE: Ocular findings associated with *R equi* in foals

Abstract

Objective: To describe ocular findings associated with *Rhodococcus equi* bronchopneumonia in foals, and to determine whether severity of the ocular lesions is related with outcome. **Animals studied:** Foals diagnosed with *R. equi* infection at the VTH-UAB from January 2002 to December 2017. **Procedure:** *Rhodococcus equi* infection was diagnosed by means of clinical signs, radiographic/ultrasonographic findings, and/or positive culture. In all the foals, a complete ophthalmic examination by a boarded ophthalmologist was performed and ocular signs were recorded and graded (0-4). **Results:** Thirty-nine foals were included in the study, from which 12 showed signs of bilateral anterior uveitis (30.8%). Among these, 3 foals were classified as mildly uveitis-affected (MUA:7.7%) and 9 as severely uveitis-affected (SUA:23.1%). Five SUA foals showed green aqueous flare (5/9;55.5%). Despite the systemic treatment, 9/39 sick foals died (23.1%), being the fatality rate different between groups: SUA (4/9;44.4%), MUA (0/3; 0%) and non-uveitis-affected foals (5/27;18.5%). Among SUA foals, only one with green aqueous flare died (1/5;20%). **Conclusion:** Bilateral anterior uveitis is highly prevalent in foals with *Rhodococcus equi* pneumonia (30.8%). The severity of anterior uveitis might be considered a non-survival prognostic factor and, until proven otherwise, green aqueous flare could be taken as a strongly suggestive ocular sign of the disease. Findings of this study remark the clinical relevance of performing a complete ophthalmic examination in sick foals, in order to help in the diagnosis and prognosis of uveal diseases, as well as to guarantee visual soundness.

Key words: green aqueous flare, prognosis, uveitis, eye, horse

INTRODUCTION

Ocular involvement in systemic diseases of horses is common; however, unless ophthalmic examination is used as a first-approach diagnostic tool, some eye conditions may go unnoticed. Detection and assessment of ocular abnormalities during investigation of horses with systemic disease may provide valuable information on the differential diagnoses, treatment options and prognosis of systemic disorders. Furthermore, timely recognition of ocular manifestations of systemic diseases will permit targeted ophthalmic treatment with potential benefits to limiting ocular pathology and vision loss.¹⁻² Unfortunately, ocular examination is nowadays greatly underused in the diagnosis of systemic diseases in the horse.¹

Reports on ocular manifestations of systemic diseases in foals are few, including neonatal septicemia³, strangles (*Streptococcus equi* infection),^{4,5} adenoviral infection,⁶ equine viral arteritis,⁷ equine herpesvirus-1 infection (EHV-1)⁸ and *Rhodococcus equi* (*R. equi*).⁹⁻¹⁵ To the best of our knowledge, no pathognomonic ocular signs have been described to date for any systemic disease in foals, being impossible to issue the etiologic diagnosis based only on ocular findings.

Rhodococcus equi bronchopneumonia is a well-recognized cause of morbidity and death in foals aged 3 weeks to 6 months.⁷ *R. equi* is a Gram positive facultative intracellular bacterium, ubiquitous in the horse environment and resistant to most disinfectants and dry conditions. This microorganism induces mainly granulomatous bronchopneumonia, but also other extrapulmonary disorders (EPDs), such as abdominal (ulcerative enterotyphlocolitis, intra-abdominal abscesses, intra-abdominal lymphadenitis, peritonitis, pyogranulomatous lesions in liver and kidney), musculoskeletal (septic arthritis, non-septic polysynovitis, osteomyelitis, myositis), hematologic (immune-mediated hemolytic anemia), neurologic (intracranial abscesses, meningitis), dermatologic (cellulitis, subcutaneous abscesses), and ocular.⁹⁻¹⁵ Unfortunately, up to date,

ophthalmologic lesions associated with *R. equi* infection have not been widely described, being limited to anecdotal case reports,^{10,13-15} a consensus statement of the American College of Veterinary Internal Medicine,¹¹ a retrospective study focused on EPDs⁹ and one review paper on general clinical signs.¹⁶ To the authors' knowledge, there are no published studies describing in detail complete ophthalmic examination's findings in foals with *R. equi* bronchopneumonia.

The purpose of the present retrospective clinical study was to further characterize ocular inflammatory findings in foals with *R. equi* bronchopneumonia, when examined by boarded ophthalmologists, and to determine whether the degree of uveal inflammation is related to outcome.

MATERIAL AND METHODS

Case selection

The medical database of the Veterinary Teaching Hospital of the Universitat Autònoma de Barcelona (VTH-UAB) was reviewed to identify foals diagnosed of *Rhodococcus equi* bronchopneumonia that underwent a complete ophthalmic examination, between January 2002 and December 2017. Patients lacking a fundus examination were excluded unless this was impaired due to the presence of severe anterior segment inflammation.

Medical records review

Data extracted from medical records included clinical history, reason/s for initial evaluation at the hospital, signalment, complete ophthalmic examination findings, bloodwork, complementary imaging tests (radiography or ultrasonography) and microbiologic culture, when available.

For the purpose of this study, the diagnosis of *R. equi* infection was defined as a foal aged 3 weeks

to 6 months that met at least two of the following criteria: (1) clinical signs consistent with pneumonia (temperature $>39.5^{\circ}\text{C}$, a respiratory rate $>80/\text{minute}$, coughing, purulent nasal discharge, abnormal lung sounds), (2) white blood cell count $>13.0 \times 10^9/\text{L}$, (3) radiographic or ultrasonography evidence of bronchopneumonia (where abscesses were defined as focal hypoechoic areas of consolidation with a diameter $\geq 1.0\text{ cm}$), and (4) cytological identification of gram-positive coccobacilli in, or isolation of *R. equi* by microbiologic culture from an abscess or tracheobronchial aspirate (TBA).^{9,17}

Complete ophthalmic examination, clinical signs and foals' classification

In all the foals, complete ophthalmic examination was performed in dim light conditions within 24 hours after admission. Menace response, dazzle reflex, palpebral reflex, and pupillary light reflexes were evaluated in every foal. Diffuse transillumination and slit lamp biomicroscopic examination (Kowa SL-15, Kowa Company Ltd., Tokyo, Japan) were completed and findings recorded. Tonometry was performed by applanation (Tonopen XL[®], Mentor, Norwell, MA, USA) or rebound (TonoVet[®], Icare Finland Oy, Helsinki, Finland) tonometers, followed by the application of one drop of tropicamide 2% (Colircusí tropicamida, Alcon Laboratories, Masnou, España) and indirect ophthalmoscopy (Heine Omega 500, Heine, Herrsching, Germany). Direct ophthalmoscopy and fluorescein stain were performed when needed.

Ocular signs were graded as follows: absent (grade 0), mild (grade 1), moderate (grade 2), severe (grade 3) and very severe (grade 4). Exudative anterior uveitis was defined as the presence of aqueous flare, fibrin, hypopion and/or hyphema. Other minor ocular signs, such as corneal edema, decreased intraocular pressure, miosis, keratic precipitates and synechia, were also considered and classified.

Based on the presence and severity of the clinical signs shown, foals were then grouped as

severely uveitis-affected (SUA), mildly uveitis-affected (MUA) or non-uveitis-affected foals. Horses were included in the SUA group when the degree of aqueous flare was 3 or 4, in the MUA group when the degree of aqueous flare was 1 or 2, and in the non-uveitis-affected group when no signs of anterior uveitis were seen.

Treatment and outcome

In addition to the appropriate systemic antimicrobial treatment [combination of a macrolide (erythromycin, azithromycin or clarithromycin) and rifampin, from 3-12 weeks] and ancillary therapies (nonsteroidal anti-inflammatory drugs, provision of adequate nutrition and fluid therapy, oxygen therapy, bronchodilators...), topical treatment for ocular disease was also administered, when needed. Foals with anterior uveitis were treated with topical dexamethasone (4-6 times/day; Maxidex®, Alcon laboratorios, El Masnou, Barcelona, Spain), tropicamide (2-3 times/day; Colircusí tropicamida®, Alcon Laboratorios, El Masnou, Barcelona, Spain) and artificial tears (4-6 times/day; Viscotears®; Bausch & Lomb, Alcobendas, Madrid, Spain).

Complete ophthalmic examinations were performed daily until foals were discharged, humanely euthanized or died due to systemic complications. General and ocular outcomes were recorded. Poor outcome was considered when foals died or were humanely euthanized due to systemic complications of the disease. Eyes were considered as visual when a positive menace response and normal photopic/scotopic maze tests were present.

Statistical analysis

Descriptive statistics was performed using SPSS 22.0 (SPSS®, Chicago, IL, USA). Quantitative variables were described with the mean, minimum, and maximum values, and qualitative variables with the number of animals or eyes, and percentages.

RESULTS

Animals

During the study period, a total of 44 foals were diagnosed with *R. equi* bronchopneumonia, but only 39 met the inclusion criteria. There were 19 fillies and 20 colts of a mean age of 3 months, ranging from 1 to 5 months. Andalusian horses were overrepresented (n=32/39;82.1%). *Rhodococcus equi* bronchopneumonia was diagnosed by means of clinical signs (39/39;100%), ultrasonography findings (24/39;61.5%), radiographic findings (13/39;33.3%) or a positive culture in abscess or TBA (6/39;15.4%).

Ophthalmic findings and foal classification

Bilateral anterior uveitis was present in 12 foals (12/39;30.8%) (Table 1). The condition was exudative in all the animals, being graded as mild in 3 foals (MUA foals 3/39;7.7%) and severe in 9 (SUA foals 9/39;23.1%). In the SUA group (n=9), all the very severely affected foals (5/9) depicted a marked green color of the anterior chamber aqueous flare (5/39;12.8%). Apart from aqueous flare and the fibrin clots [bilaterally observed in the anterior chamber of 3 SUA foals (3/39;7.7%)] there were no other specific signs of anterior uveitis (hyphema, hypopion, keratic precipitates...). Posterior segment examination was precluded in all the SUA foals (9/39;23.1%), in which ocular ultrasonography ruled out severe posterior segment affection. Fundus examination was physiologic in all the MUA foals, showing bilateral peripapillary chorioretinal inflammation in just one non-uveitis-affected foal.

Treatment and outcome

Systemic antibiotic medication was administered in all the sick foals and topical ocular medication in foals affected with uveitis (12/39;30.8%), being drops frequency directly associated to uveitis severity. Despite the systemic treatment, 9 out of the 39 sick foals included in the study died or

were humanely euthanized (9/39;23.1%) (Table 1). The case fatality rate differed significantly between groups, being 18.5% in non-uveitis-affected foals (5/27) and 33.3% in the uveitis-affected foals (4/12). Additionally, all the uveitis-affected foals that died were from the SUA group (4/9;44.4%), while all the foals from the MUA group had good outcome (0/3;0%). The outcome was very similar for foals with green and non-green aqueous flares, showing incidences of 1/4 (20%) and 3/8 (37.5%), respectively. No ocular sequels were observed at discharge in SUA (5/9;55.6%) or MUA (3/3;100%) foals, being all the eyes visual.

DISCUSSION

Ocular signs of systemic diseases have been rarely reported in foals.³⁻¹⁵ *Rhodococcus equi* bronchopneumonia has been sporadically associated to ocular signs in foals,^{10,12-15} but no detailed description of the ocular findings induced by *R. equi* infection has been reported. Although the presence of uveitis is repeatedly named as extrapulmonary disorder (EPD) associated with *R. equi*, this is the first study describing that severe anterior uveitis is highly prevalent in foals affected by *R. equi* infection, characterizing its ocular findings, and suggesting that the severity of uveitis may be considered as a non-survival prognosis factor in sick foals.

Rhodococcus equi infection in foals has been sporadically associated with ocular findings such as anterior uveitis, panophthalmitis and keratouveitis, being anterior uveal affection the most common ocular diagnosis.^{9,10,13-15} In a retrospective study on EPDs in 150 foals with *R. equi* infection, an incidence of 11% was observed for anterior uveitis, placing iridocyclitis as the seventh most common EPD, following diarrhea (33%), immune-mediated polysynovitis (25%), ulcerative enterotyphlocolitis (21%), intra-abdominal abscesses (17%) and abdominal lymphadenitis (17%).¹² Despite both studies being performed in veterinary hospitals with

theoretically similar caseload, the incidence of uveitis in our study was significantly higher (30.8% vs. 11%). This numeric discrepancy could be associated to the fact that in the present study a routine complete eye exam was performed in all the sick foals within 24 hours after admission, whereas in Reuss et al. study, no information was given in regards to this aspect, reason why some cases may have gone unnoticed. The present findings suggest a more important clinical role of uveitis in EPDs of foals with *R. equi* infection. Thus, ophthalmic examination is strongly recommended in foals with a clinical presentation compatible with *R. equi* infection.

Ocular signs in sick foals can be unilateral or bilateral.^{3,10,12-15} Among all the uveitis-affected foals of the study, no unilateral cases were diagnosed, showing all the foals a symmetrical presentation. In all cases, uveitis was characterized by different degrees of aqueous flare, evaluated by means of slit-lamp biomicroscopy. Traditionally, clinical assessment of the aqueous flare has been successfully performed evaluating the refraction of a focus light when passing the anterior chamber. This somehow subjective method of classification has been taught from mentors to residents, reaching a uniform grading scale all over the world. Nevertheless, aqueous flare evaluation can be more objectively performed by using a flare meter, which allows both, determination of the flare and the number of cells present in the aqueous of the anterior chamber.¹⁸ Unfortunately, the diagnostic tool is non-portable and its price does not justify its use in a veterinary hospital. In the present population, the biomicroscopic aqueous flare evaluation depicted that the incidence of severe uveitis was significantly higher (23.1%) than mild-moderate uveitis (7.7%). This finding is not surprising given the importance of the vascular spread in the pathogenesis of *R. equi* dissemination⁷ and the already known weakness of the equine blood-aqueous barrier (BAB).^{19,20} Among the SUA foals (n=9), 3 showed important amounts of fibrin in the anterior chamber of both eyes (OU). The high capacity for fibrin production in foals is

already known, which in severe cases can even fill the complete anterior chamber.^{21,22} If not treated, fibrin can induce corneal or lens sequelae, inducing different degrees of hemeralopia or even blindness. Conversely to previous reports,^{10,12,15} no cases of hyphema, hypopion or keratic precipitates, were observed in the present study.

To the authors' knowledge there are no pathognomonic or indicative ocular signs reported for *R. equi* infection. Because of the necessity of early diagnosis and treatment of *R. equi* bronchopneumonia for achieving a good outcome, reliable and rapidly available tests for diagnosis of infection are desirable. Culture and PCR testing are undoubtedly the gold standard in the diagnosis of *R. equi* infection nowadays but, unfortunately, both diagnosis methods take time and have not been considered as prognosis factors for survival. Green aqueous flare was seen in 5/9 SUA foals. The interpretation of this sign is confusing because none of the foals at the moment of the ophthalmologic examination was under any medication than could explain the green coloration in the anterior chamber, nor this ocular finding has been described previously in the literature. Although, in the present study no cytology of the aqueous humor or histopathology was performed that could elucidate the pathogenic mechanism, an acute ocular inflammation with high concentration of PMNN is strongly suggested. Thus, until proven otherwise, green aqueous flare could be considered as an indicative sign for *R. equi* infection in foals aged 3 weeks to 6 months with clinical signs of bronchopneumonia. Further evaluation is warranted to determine whether other infectious diseases that cause uveitis can also cause green flare.

Unfortunately, the pathogenesis for uveitis associated with *R. equi* infection is unknown, but might include dissemination of bacteria to the eye or immune-mediated mechanisms. Immune-complex deposition is also thought to contribute to other EPDs such as polysinovitis.^{9,12,14,16} Further studies are needed to establish the pathogenic mechanism of the ocular presentation of the

disease.

Intraocular signs have been previously associated with poor prognosis in septic foals, in which not only the presence of uveitis was considered a negative survival factor, but also the degree of intraocular inflammation (the most severe the inflammation was, the poorest prognosis).³ Similarly, extrapulmonary manifestations of *R. equi* have been previously associated with poor prognosis, and more specifically, anterior uveitis has been strongly associated with failure to survive.⁹ The present study not only agrees with *Reuss et al.* paper, but also suggests that the survival prognosis in *R. equi* affected foals varies depending on the severity of intraocular inflammation, being foals with mild uveitis more likely to survive. Due to the reduced number of affected foals, there is not enough sample size to establish a statistical relationship between green aqueous flare and prognosis, but since most of the foals with severe uveitis had green aqueous flare, it can be clinically extrapolated that this ocular finding might be associated with a poor prognosis. The reason for this association has not been elucidated yet. So, the question remains whether uveitis was merely an indicator of other EPDs or whether the association with non-survival was real and uveitis represents a more severe systemic response. The present findings do not support the use of degree of uveitis as the only prognosis factor for survival, but might suggest a poor prognosis of survival for more severely affected foals.

In the present study active chorioretinal inflammation was seen only in one foal (1/39;2.6%), being bilaterally located around the optic nerve head, coinciding with the most permeable point of the blood-retinal barrier (BRB). This low incidence could be explained by the weakness of the BAB to endogenous damage when compared with BRB,²³ by a preference of *R. equi* for the BAB, or by an underestimation of the chorioretinal lesions due to the severe inflammation of the anterior segment in SUA foals (9/39), preventing adequate fundoscopic examination.

Like many retrospective studies, the herein shows some limitations that should be enlightened. Firstly, the diagnosis of *R. equi* was performed based on clinical findings, bloodwork, ultrasonography and radiology in all foals, but microbiologic culture or cytology of TBA fluid was performed only in 15.4% of the cases. These two latter diagnostic techniques, together with the polymerase chain reaction (PCR) testing of TBA fluid to identify the virulent *vapA* gene of *R. equi*, are considered nowadays as the gold standard for the diagnosis of the disease.^{11,24} In daily practice, these advanced diagnosis methods are only recommended when the clinical history and clinical findings do not fully match. Thus, in general these techniques are recommended for cases in which clinical doubts are important and justify the risks of a TBA in a very sick foal. In the present study, the majority of sick foals were from endemic farms with recurrent confirmed cases, and the risk of performing a TBA in foals with grave respiratory distress (dyspnea) were not considered by boarded equine internists. Secondly, the relative low number of cases as well as the fact that only the most severely affected foals are referred to a referral center, should be also taken into consideration.

In conclusion, this is the first study describing in detail the ophthalmologic findings seen in foals with *Rhodococcus equi* bronchopneumonia. The severity of anterior uveitis may be considered a survival prognostic factor and until proven otherwise, green aqueous flare could be considered as a suggestive sign of the disease. Findings of this study remark the clinical relevance of performing a complete ocular examination in sick foals, in order to help in the diagnosis and prognosis of uveal diseases, as well as to guarantee visual soundness.

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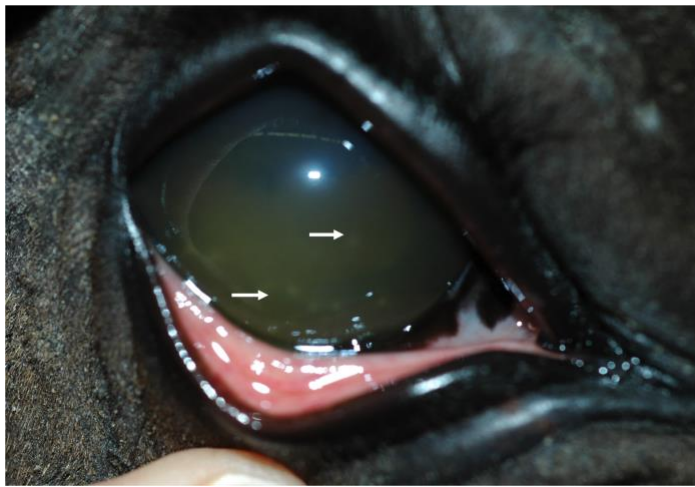
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FIGURE LEGENDS

Figure 1. Severe anterior exudative uveitis in a 3 months-old foal. Fibrin deposits (white arrows) and diffuse green aqueous flare are observed.



Foal's classification (n=39)		Poor outcome (n=9)	
Non-uveitis-affected foals (27 foals; 69.2%)		5/27 non-uveitis affected foals (18.5%)	
Mildly uveitis-affected foals (MUA) (3 foals; 7.7%)		0/3 MUA foals (0%)	
Severely uveitis- affected foals (SUA)	Green aqueous flare (5 foals; 12.8%)	4/12 SUA foals (33.3%)	Green aqueous flare 1/5 (20%)
	No green aqueous flare		No green aqueous flare 3/4

(9 foals; 23.1%)	(4 foals; 10.2%)		(75%)
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336 **Table 1.** Ocular findings and outcomes in foals with *Rhodococcus equi* bronchopneumonia.