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Investigations into the occurrence of serum amyloid A in the equine eye

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Summary: The acute-phase protein serum amyloid A (SAA) is a very sensitive marker for inflammation in horses and humans. Due to its quick reaction profile in serum, it can be used to diagnose and monitor inflammatory processes. The aim of this study was to establish SAA's involvement in equine ocular diseases and evaluate its use as a diagnostic tool. Consequently, SAA was determined by ELISA in 220 intraocular samples from 151 horses (90 samples of aqueous humour: AH, 130 samples of vitreous body: VB) and 86 serum samples. The intraocular samples were obtained by paracentesis, vitrectomy or after euthanasia. All eyes were categorized according to ophthalmological findings into the following groups before taking the samples: controls, equine recurrent uveitis (ERU), glaucoma, glaucoma and ERU, Leopard-Piebald Uveitis, miscellaneous (e.g. keratitis, tumour), healthy eyes whose partner eye suffers from ERU and ERU-like symptoms. Additionally, 27 samples of eyes suffering from ERU were examined retrospectively to evaluate the SAA as a prognostic marker for the development of secondary glaucoma after suffering from ERU. The SAA concentrations in healthy eyes and eyes that exhibited non-uveitic related changes were below the test's detection limit ($< 0.1 \mu g/ml$). The SAA concentrations in eyes suffering from uveitis (ERU and Leopard-Piebald Uveitis) were significantly increased, both in AH and VB. Acutely inflamed eyes contained significantly more SAA than those without signs of inflammation. Concentrations of SAA were significantly increased in eyes where an intraocular infection with leptospira could be verified compared to eyes without leptospiral infections. The amount of SAA found in intraocular fluids correlated greatly and significantly positive with the level of the intraocular microscopic agglutination test (MAT) titre for leptospira. The space of time that had passed between the last uveitis attack and the sampling of the eye (in days) did not correlate with the SAA levels in AH and there was only a poor, negative significant correlation for levels in VB. Correlation of the SAA levels in serum and AH was weak and not significantly positive, whereas SAA levels in serum and VB did not correlate at all. The SAA concentrations in eyes suffering from glaucoma were below the detection limit in AH, and slightly elevated in VB. By the time of vitrectomy, there was no significant difference in the SAA concentrations in eyes that developed and did not develop glaucoma secondarily to ERU. It was demonstrated that SAA is participating in intraocular inflammation. Unlike in serum, increased amounts of the protein can be detected in intraocular fluids long after the inflammatory stimulus has subsided. Bacterial infection of the inner eye by leptospira spp. seems to trigger considerably greater amounts of SAA than non-infectious eye diseases. The concentration of SAA correlated strongly with the intraocular antibody titre against leptospira, thus, it seems that SAA is relevant in fighting the Gram-negative bacteria. Due to the low (AH) or non-existent (VB) correlation between SAA in serum and the eye, it is perceived that the protein might, at least in part, be produced locally. Consequently, using the SAA serum concentration as a diagnostic tool for eye diseases is, therefore, not possible. As has been shown, SAA levels are elevated in eyes suffering from ERU, Leopard-Piebald Uveitis and Glaucoma. Those increased concentrations might contribute to the amyloid residues considered responsible for a malfunctioning aqueous outflow and an increased intraocular pressure. Since there is no significant difference in SAA concentrations in eyes suffering from ERU that did or did not develop secondary glaucoma, the protein cannot be used as a prognostic tool for the development of this disease.

Keywords: horse, equine, ERU, Amyloid A, inflammation, marker, ophthalmology

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Introduction

The acute-phase protein serum amyloid A (SAA) is considered to be a very sensitive marker for inflammation measured in equine serum (*Nunokawa* et al. 1993). It is produced primarily by the liver (*Husby* et al. 1994) in response to an inflammatory or infectious stimulus, induced by inflammatory cytokines such as Interleukin-1, Interleukin-6 and tumour necrosis factor alpha (*Uhlar* et al. 1999). However, it has been discovered that certain extrahepatic tissues seem to be able to synthesize the protein as well (*Uhlar* and *Whitehead* 1999, *McDonald* et al. 2001, *Jacobsen* et al. 2006, *Christoffersen* et al. 2010, *Berg* et al. 2011). The SAA serum concentrations rise rapidly following tissue injury or infection and may increase up to a thousandfold (*Pepys* et al. 1983, *Pepys* et al. 1989, *Sletten* et al. 1989, *Uhlar* and *Whitehead* 1999, *Jacobsen* et al. 2007) – the highest levels being measured in response to bacterial infections (*Pepys* et al. 1989, *Chavatte* et al. 1992, *Stoneham* et al. 2001, *Jacobsen* et al. 2006, *Ludwig* et al. 2016). The SAA's half-life in plasma is very short (30 min–2 h), resulting in a decrease in concentration shortly after the inflammatory stimulus subsides (*Hoffman* et al. 1983, *Uhlar* and *Whitehead* 1999). The SAA can be used to either diagnose or monitor the progression of an inflammatory disease in equine patients due to its distinct increase during inflammation and the strong correlation between its serum levels and the status of the inflammation (*Pepys* et al. 1989, *Uhlar* and *Whitehead* 1999, *Petersen* et al. 2004, *Jacobsen* and *Andersen* 2007).

Studies on the occurrence of SAA in correlation to equine eye diseases are limited. *Labelle* et al. (2011) compared SAA levels in the serum of horses suffering from systemic disease to those affected with ocular disease (uveitis, keratitis) and healthy horses. The SAA levels were significantly increased in the systemic disease group, whereas there was no significant difference in the SAA levels of the healthy controls and the

group affected by eye disease. *Wang* et al. (2008) found significantly elevated levels of SAA mRNA in the trabecular meshwork of human eyes suffering from glaucoma in comparison to healthy eyes. The level of the SAA protein itself found in the trabecular meshwork also showed a significant increase compared to controls. However, the SAA levels in serum did not differ significantly between patients suffering from glaucoma and controls.

Eight per cent of the German horse population suffer from equine recurrent uveitis (ERU) (*Szemes* et al. 2000), a painful disease that is still the number one cause of blindness in horses (*Spiess* 1997, *Gerhards* et al. 2001, *Gilger* et al. 2011). In addition, the eye with uveitis commonly develops secondary glaucoma, even after the ERU attacks have been treated and stopped by vitrectomy (*Schinagl* 2017).

Equine intraocular fluids have not been tested for SAA to date. However, deposits of amyloid A, a degradation product of SAA, have been demonstrated in equine eyes suffering from chronic recurrent uveitis or glaucoma (*Cielewicz* 2014, *Ostevik* et al. 2014). Therefore, it is speculated that SAA is being released intraocularly during a uveitis-related acute-phase response and then metabolized and deposited as amyloid a, adding to an increased intraocular pressure by obstructing the outflow pathway. The main goal of the present study was to determine the occurrence of SAA in equine ocular diseases and evaluate its use as a diagnostic tool. Consequently, SAA concentrations were assayed in serum, aqueous humour (AH) and vitreous body (VB) of healthy horses and horses suffering from various types of ocular disease. Serum concentrations were compared to the SAA levels in corresponding ocular fluids.

Material and Methods

A total of 220 intraocular samples from 151 horses, consisting of 90 samples of AH, 130 samples of VB and 86 serum samples were examined (Table 1). The intraocular samples used in this study were obtained by paracentesis, vitrectomy or after euthanasia from horses admitted to the Equine Clinic, University of Munich, from June 2013 to June 2016. The serum samples were collected preoperatively during routine clinical procedures.

Paracentesis was performed under general anesthesia. After applying eyedrops containing gentamycin (Gentamicin-POS® Ursapharm) as well as a local anesthetic (Proparakain-POS® 0,5% Ursapharm), a 27 gauge needle was inserted into the anterior chamber via the limbus and 1 ml of aqueous was aspirated. 4 ml of undiluted vitreous were obtained through tubes connected to the vitrectomy cutter, which was inserted through the pars plana into the vitreus chamber. In case of euthanasia a sterile catheter was used instead of the vitrectomy cutter. All samples were stored in plastic tubes (1,5 ml Mikrorohre PCR-PT, Fa. SARSTEDT) directly after the extraction.

Upon presentation to the university clinic, the eyes were examined by the author and then classified according to ophthalmologic findings:

- Equine recurrent uveitis ("ERU") was diagnosed when the eye had suffered from recurrent attacks of uveitis and exhibited findings of acute or chronic inflammation, such as lacrimation, blepharospasm, photophobia, hazy or oedematous cornea, circumlimbal vascularization of the cornea, inflammatory products in the anterior chamber, miosis, posterior synechia or iris residues on the anterior lens capsule, partial or mature cataract, inflammatory products on the posterior lens capsule or in the VB, partial or complete retinal detachment or chorioretinal scars, or diffuse opacity of the VB caused by inflammatory products. All the samples in this group tested positive for leptospira.
- A diagnosis of "Glaucoma" was made when the eye exhibited corneal band opacities or corneal oedema, an enlarged globe, (sub-)luxation of the lens, an increased intraocular pressure (>30 mmHg) or a combination of these findings. Intraocular pressure was measured using the Tono-Pen AVIA Applanation Tonometer (Fa. Reichert).
- Eyes which had suffered from ERU and, thus, received vitrectomy, then subsequently developed glaucoma were grouped as "ERU+Glaucoma".
- "Leopard-piebald (LP)" horses, such as Appaloosas and Knabstruppers, were presented with intraocular changes that coincide with a chronic and insidous form of uveitis, which, in most cases, is not painful for the horse and, therefore, is only recognized once the animal shows signs of impaired vision. Typical findings in these chronically inflamed eyes were anterior and posterior synechia, cataract

Table 1 Overview of the samples used in this study (AH = aqueous humour, VB = vitreous body)
Übersicht über die in der Studie verwendeten Proben (AH = aqueous humour, VB = vitreous body)

Group	Horses	Eyes	AH	VB	Samples
	[n]	[n]	[n]	[n]	total [n]
Controls	14	19	17	7	24
ERU	75	84	31	78	109
Glaucoma	9	12	11	4	15
ERU + Glaucoma	3	3	3	3	6
LP Uveitis	8	10	9	3	12
Miscellaneous	10	11	10	4	14
ERU Partner Eye	2	6	6	1	7
Suspected ERU	5	5	3	3	6
ERU (retrospectively)	26	27	/	27	27
Total [n]	152	177	90	130	220

formation, partial detachment of the retina or phthisis bulbi. All the samples from this group tested negative for leptospira.
- Eyes suffering from keratoconjunctivitis, keratitis, recurrent corneal defects, tumours (squamous cell carcinoma), mature cataract or chronic intraocular findings probably caused by trauma were summarized in the "miscellaneous" group.
- In some cases, when performing vitrectomy on an ERU eye, samples of the healthy "ERU partner eyes" were taken to test them for leptospira as well. Those samples were put into an extra group to prevent a corruption of the SAA levels through the ERU eye already inflamed.
- "Suspected ERU" were eyes that had either exhibited ERU-like symptoms or chronic findings coinciding with ERU but tested negative for leptospira and did not fit into the LP group.
- Additionally, 27 vitreous samples taken during vitrectomy were compared retrospectively in an extra group. Twelve of these samples were taken from eyes that had developed glaucoma one to five years after vitrectomy. Fifteen samples stem from eyes that evidentially had not developed glaucoma up to ten years after vitrectomy.

The control group consisted of clinically healthy eyes.

The status of inflammation at the time the sample was taken was classified according to the clinical findings. "Acute/subacute" describe eyes that showed signs of an acute inflammatory attack on presentation or were still subacutely inflamed at the time the sample was taken, despite anti-inflammatory treatment. Findings in these eyes included haziness of the cornea, fibrin residues in the anterior chamber, incompletely dilated pupil despite being treated with atropine, yellowish-sluggish fundus, cloudiness of the vitreous or a noticeably increased intraocular pressure combined with an oedematous cornea. "No signs of inflammation/chronic" described eyes that did not show any signs of irritation or findings of acute inflammation on presentation. The ophthalmological changes were of a chronic nature, such as synechia, cataract formation, inflammatory residues in the vitreous, partially detached retina or an elongated increase in intraocular pressure.

The samples were tested for leptospira via real time PCR (IDEXX VetMed Laboratory, Ludwigsburg) and MAT/ELISA (Bavarian State Office for Health and Food Safety, Oberschleissheim). Serum and intraocular samples for SAA analysis were stored and frozen at -17 °C until evaluation, using the TRIDELTA multispecies SAA ELISA Kit (LABOKLIN Laboratory, Bad Kissingen). The SAA reference range for this test is $<7\,\mu$g/ml in serum; SAA amounts smaller than $0.1\,\mu$g/ml cannot be detected. A range for intraocular samples has not yet been determined.

Statistical analysis was performed using SPSS 24 (SPSS Inc., Chicago). Correlation between two variables was tested calculating the Spearman's rank correlation coefficient. The SAA concentrations were tested for statistical normal distribution using the Kolmogorov-Smirnov test. Significant deviations were tested using the Mann-Whitney U test or Kruskall-Wallis test. All data were expressed as mean, standard deviation, minimum, maximum and the quartiles. The significance level was determined as 5% for all statistical tests.

Results

In intraocular samples from healthy control eyes median SAA levels were below the assay detection limit (AH: $x = 0.05\,\mu$g/ml, VB: $x = 0.05\,\mu$g/ml). Compared to the controls, significantly increased SAA concentrations were found in diseased eyes (AH: $x = 0.75\,\mu$g/ml, $p = 0.011$) (VB: $x = 4.98\,\mu$g/ml, $p = 0.003$). Acutely inflamed eyes contained significantly more SAA (AH: $x = 3.40\,\mu$g/ml, VB: $x = 16.89\,\mu$g/ml) than eyes with no signs of inflammation (AH and VB $x = 0.05\,\mu$g/ml; $p < 0.001$ each).

The concentrations of SAA in eyes exhibiting non-uveitic related changes (e.g. keratitis, tumours, ERU partner-eyes) were also less than the detection limit (AH: $x = 0.05\,\mu$g/ml, VB: $x = 0.05\,\mu$g/ml). Contrary to those results, the SAA levels in eyes suffering from uveitis, such as ERU and LP uveitis, were significantly increased both in AH (ERU: $x = 3.00\,\mu$g/ml, $p = 0.0008$; LP: $x = 5.21\,\mu$g/ml, $p = 0.0025$) and VB (ERU: $x = 6.56\,\mu$g/ml, $p = 0.0014$; LP: $x = 0.38\,\mu$g/ml, $p = 0.0107$) (Figure 1).

The space of time that has passed between the last uveitis attack and the sampling of the eye (in days) does not correlate with the SAA levels in AH ($R = -0.016$; $p = 0.921$). There is a poor, negative, but significant correlation in VB ($R = 0.274$; $p = 0.023$).

The highest SAA levels determined in this study were found in AH ($285.64\,\mu$g/ml) and VB ($308.03\,\mu$g/ml) of eyes suffering from ERU. The SAA concentrations in eyes with an established leptospiral infection were significantly higher than in those where no

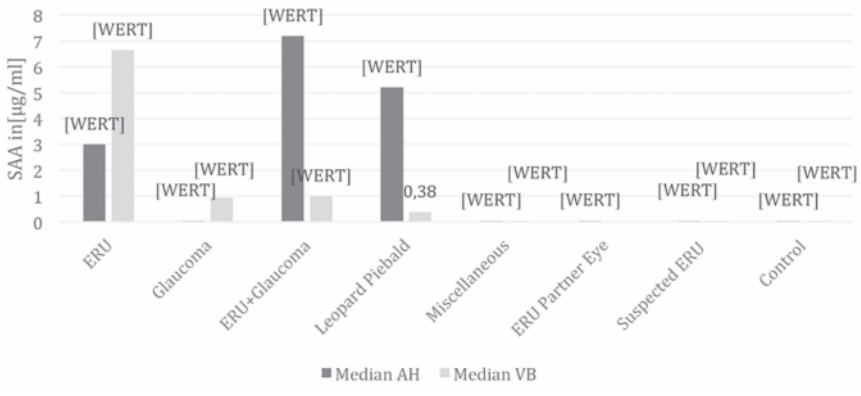

Fig. 1 Median SAA levels in aqueous humour (AH) and vitreous body (VB) of the various diseases.
Mediane SAA Gehalte in Kammerwasser (AH) und Glaskörper (VB) bei verschiedenen Erkrankungen

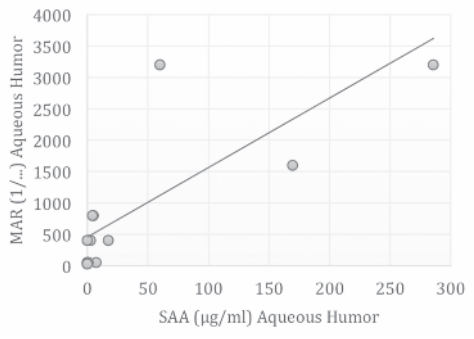

Fig. 2 Spearman's correlation for SAA levels in AH in relation to the intraocular titre against leptospira.
Spearman-Korrelation für die SAA Gehalte im Kammerwasser in Abhängigkeit vom intraokularen Leptospirentiter

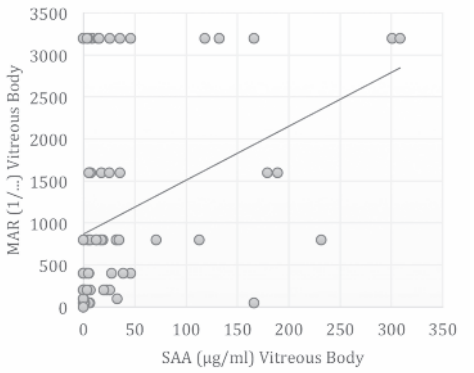

Fig. 3 Spearman's correlation for SAA levels in VB in relation to the intraocular titre against leptospira.
Spearman-Korrelation für die SAA Gehalte im Glaskörper in Abhängigkeit vom intraokularen Leptospirentiter

leptospira could be detected (AH: $p = 0.005$; VB: $p = 0.001$). The amount of intraocular SAA correlated greatly and significantly positive with the level of the intraocular MAT titre for leptospira (AH: $R = 0.833$; VB: $R = 0.469$)(Figures 2 and 3).

Concentrations of SAA in serum and AH correlated weakly and non-significantly positive ($R = 0.237$; $p = 0.121$); the SAA concentrations in serum and VB did not correlate at all ($R = -0.044$; $p = 0.742$). One horse had to be euthanized due to severe and therapy-resisting attacks of leptospiral uveitis in both eyes, resulting in complete blindness. The SAA levels in both eyes were massively elevated (AH right eye: 106.83 µg/ml; VB left eye: 166.09 µg/ml), whereas the SAA level in serum was below the detection limit (< 0.1 µg/ml).

Eyes suffering from glaucoma contained SAA concentrations below the detection limit in AH ($x < 0.1$ µg/ml), however, the SAA in VB was slightly elevated ($x = 0.94$ µg/ml). The levels of intraocular SAA by the time of vitrectomy did not differ significantly in eyes that did and did not develop glaucoma secondarily to ERU ($p = 0.446$).

Discussion

As expected, there was no SAA to be detected in the intraocular fluids of healthy eyes. Diseased eyes generally exhibited significantly more SAA than controls. Acutely inflamed eyes contained considerably greater SAA levels than chronically affected eyes. An increase in SAA by 10- to 1000-fold during inflammation is reported in serum (*Pepys* and *Baltz* 1983, *Sletten* et al. 1989, *Uhlar* and *Whitehead* 1999, *Jacobsen* and *Andersen* 2007). An increase in SAA in acutely inflamed eyes by 68- (AH) and 300-fold (VB), respectively, becomes apparent when comparing the mean SAA levels with eyes having no signs of acute inflammation. The highest SAA concentrations assayed in this study were 285.64 µg/ml in AH and 308.30 µg/ml in VB – representing an increase in SAA by 5000- (AH) and 6000-fold (VB) compared to levels in uninflamed eyes. Since all the inflamed eyes received local and systemical anti-inflammatory treatment before being probed, it has to be assumed that the SAA levels measured in this study might have been lower than they would be in untreated eyes.

Serum amyloid A is characterized by a very short half-life in serum, therefore, a rapid decrease in its serum concentration can be witnessed after the inflammatory stimulus subsides and the protein is degraded by the liver (*Hoffman* and *Benditt* 1983, *Uhlar* and *Whitehead* 1999). In contrast to this observation, there is no (AH) or only a very weak correlation (VB) between the SAA levels measured in intraocular samples and the time that has passed since the last ocular inflammation. A sample was taken in one eye 100 days after the last acute inflammation had occurred, and the AH still exhibited a considerably elevated SAA concentration of 10.94 µg/ml. Intraocular SAA levels probably remain elevated longer than in serum due to the protein's aggregation with various exudates, inflammatory cells and collagen fibrils that are produced during an uveitic episode (*Sebag* 1989, *Niedermaier* 2002, *Wollanke* 2002, Wollanke et al. 2004, Roth 2013). Hidden inside this conglomerate, it seems that SAA is protected from being evacuated out of the eye, therefore, increased levels can still be measured a long time after the last uveitis attack. Additionally, *Roth* (2013) stated that a clinically quiescent state of the eye does not necessarily represent an immunological quiescent state. This leads to the assumption that the intraocular persistence of leptospira might trigger a continuous production of SAA, thereby causing elevated protein levels even in a clinically sound eye.

Similar to observations in other studies (*Wang* et al. 2008, *Labelle* et al. 2011), SAA levels in serum and corresponding intraocular fluids do not correlate significantly. The serum samples used in this study were mostly taken after ample local and systemical anti-inflammatory treatment in preparation for surgery. Therefore, by the time the blood was taken, a systemic participation of acute-phase proteins to an eye disease might have already subsided and circulating SAA would have already been catabolized by the liver (*Hoffman* and *Benditt* 1983, *Uhlar* and *Whitehead* 1999). However, intraocular SAA levels seem to remain elevated for a longer period of time even after the inflammation has ceased, probably due to a less efficient intraocular clearance and a persisting intraocular infection by leptospires (*Waldner* 2017). These issues could explain the discrepancy between serum and intraocular SAA levels at the same time. Yet, it seems more likely that there is no systemic acute-phase response in succession to ocular disease at all: One reason being the relatively small tissue surface affected by ocular diseases such as keratitis or uveitis compared to an inflamed bowel or lung (*Labelle* et al. 2011).

Another reason being the immune privilege of the eye, since it can downregulate a systemic immune response to intraocular inflammation via anterior chamber associated immune deviation (ACAID) to prevent further damage to the eye. The absence of a systemic acute-phase reaction would, consequently, leave SAA serum levels unaltered during intraocular inflammation (*Grisanti* 1998, *Streilein* 1999, *Zhou* et al. 2010). If this were the case, SAA assayed in the intraocular samples examined in this study was probably produced locally instead of having entered the eye by bloodstream once the blood–retina barrier had collapsed. The theory of an intraocular production of SAA is backed up by the detection of SAA m-RNA in cells of the trabecular meshwork in human eyes suffering from glaucoma (*Wang* et al. 2008). It could be useful to determine SAA isoforms in intraocular and serum samples of the same animal and then compare these isoforms for accordance or differences to further verify this theory.

Eyes infected with leptospires and suffering from ERU contain significantly more SAA than eyes with no evidence of leptospiral infection. This observation coincides with reports by several authors describing severely increased SAA levels in serum and synovia, especially in connection with bacterial infections (*Pepys* et al. 1989, *Chavatte* et al. 1992, *Stoneham* et al. 2001, *Ludwig* et al. 2016). As demonstrated in this report, intraocular SAA concentrations correlate strongly with the intraocular antibody titre against leptospira (AH: $R = 0.833$; VB: $R = 0.469$). These findings may be explained by the observation that SAA can bind to outer membrane protein A (OmpA), which is found in many Gram-negative bacteria such as leptospira (*Hari-Dass* et al. 2005, *Oliveira* et al. 2011). By binding to OmpA, SAA opsonizes the leptospira for phagocytosis through activated macrophages and neutrophil granulocytes (*Shah* et al. 2006), thus, joining in on the intraocular defence mechanisms against the Gram-negative bacteria.

Uveitic episodes are supposed to be one of the main reasons for the development of secondary glaucoma in the horse (*Cullen* et al. 2000, *Wilkie* et al. 2004, *Utter* et al. 2011, *Curto* et al. 2014). As demonstrated in this study, eyes suffering from uveitic diseases exhibit significantly increased levels of SAA. A-Serum amyloid A (A-SAA) can be degraded to form amyloid fibrils, which, in an aggregated form, may obstruct the trabecular meshwork in glaucomatous eyes (*Smith* et al. 1986, *Cohen* et al. 1987, *Uhlar* and *Whitehead* 1999, *Cielewicz* 2014). Additionally, A-SAA is considered a serum precursor of the Amyloid A protein, which is the principal component of insoluble amyloid plaques found in amyloid A amyloidosis (*Uhlar* and *Whitehead* 1999). It is assumed that the Amyloid A identified as part of the amyloid deposits found in equines eyes suffering from ERU and glaucoma, probably causing an obstruction of AH outflow, is derived from lens protein which is degraded to form amyloid fibrils in an inflammatory environment (*Meehan* et al. 2004, *Cielewicz* 2014). However, according to the findings in this study, it can be hypothesized that the SAA assayed in uveitic and glaucomatous eyes may be contributing to the formation of such amyloid deposits. The development of glaucoma is not predictable in 79.4% of the cases by the time of vitrectomy, therefore, SAA's use as a prognostic indicator for an increased intraocular pressure regarding ERU was evaluated. The protein's levels did not differ significantly between eyes that did and did not develop glaucoma up to ten years after vitrectomy, thus, unfortunately it is not possible to predict the occurrence of glaucoma by determining SAA concentrations in the VB.

It was demonstrated recently that spirochetes, including Leptospira, can form biofilm to increase resistance against antibiotics and biocides. This mechanism, consisting of microcolonies formed by exopolymeric substances and multiplying cells, is considered to be a possible explanation for the long-term survival of leptospires in environmental water (*Hall-Stoodley* et al. 2004, *Trueba* et al. 2004, *Ristow* et al. 2008). Microcolony-like formations of leptospires can also be witnessed in VBs of horses suffering from ERU (*Brandes* et al. 2007), leading to the assumption that the bacteria might be able to form biofilm intraocularly as a defence mechanism against the immune system. Since Amyloid Beta was determined as a main component in the biofilms and senile plaques formed by certain spirochetes (*Miklossy* 2016), it can be speculated that the degradation of intraocular A-SAA to Amyloid mentioned above, might, without intention, support the formation of an intraocular leptospiral biofilm and thereby contribute to a prolonged intraocular leptospiral infection.

In conclusion, the results of the present study suggest that SAA is participating in equine intraocular disease, particularly in uveitic processes such as ERU and LP Uveitis. The SAA serum levels do not correlate significantly with intraocular levels in diseased eyes, thus, it was not possible to establish the usefulness of SAA determination in blood samples to detect or monitor ocular inflammation. This observation should, however, be verified by examining intraocular fluids and corresponding serum samples from horses with ocular inflammation in the acute stages, without subjecting the horse to a possibly falsifying anti-inflammatory treatment. Based on the current results, a local intraocular production of SAA can be suspected, although further research on this topic is needed. Future studies should include the analysis and comparison of SAA isoforms found in serum and intraocular fluids to assess an intraocular SAA synthesis.

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