VOL II

Estudos em Ciências Agrárias e Ambientais

Eduardo Spers
(Organizador)

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2024

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APRESENTAÇÃO

O campo das Ciências Agrárias e Ambientais desempenha um papel fundamental na compreensão e solução dos desafios contemporâneos relacionados à produção de alimentos, à conservação ambiental e ao bem-estar animal. Em um mundo em constante transformação, questões como a sustentabilidade dos agroecossistemas, o manejo eficiente dos recursos naturais e a saúde pública se tornam cada vez mais relevantes. É com este espírito que apresentamos o volume II da coletânea "Estudos em Ciências Agrárias e Ambientais", que reúne pesquisas de autores de diversas partes do mundo, cada um contribuindo com sua perspectiva e expertise únicos.

Os quinze artigos que compõem este volume abordam uma variedade de tópicos, refletindo a riqueza e a diversidade das Ciências Agrárias. Desde práticas conservacionistas que buscam melhorar e manter agroecossistemas, até investigações sobre o uso de fitohormonas e fertilização na produção vegetal, o uso de tecnologias de processamento de madeira e a promoção do bagre armado - cada estudo traz à tona questões cruciais que impactam tanto a produção agrícola quanto a saúde ambiental.

Neste volume, também exploramos a crescente relevância dos produtos agrícolas locais, especialmente em tempos desafiadores como os que vivemos, marcados pela pandemia da COVID-19. A importância de circuitos curtos de proximidade se torna evidente, promovendo não apenas a segurança alimentar, mas também a resiliência das comunidades.

Além disso, as contribuições da veterinária destacam a importância do cuidado animal e da saúde pública, ilustrando a interconexão entre os seres humanos, os animais e o meio ambiente.

Esperamos que esta coletânea não apenas informe, mas também inspire debates e colaborações futuras entre pesquisadores, profissionais e estudantes da área. Juntos, podemos avançar em direção a um futuro mais sustentável e equilibrado, em que conhecimento pesquisa sejam os pilares para soluções efetivas.

Agradecemos a todos os autores e colaboradores que tornaram este trabalho possível. É nossa esperança que os estudos aqui apresentados contribuam para um entendimento mais profundo das questões agrárias e ambientais, e que possam servir de base para novas investigações e práticas inovadoras.

Eduardo Eugênio Spers

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DIAGNOSTICS IN A PUG DOG WITH ALLERGY REACTION ON RABIES VACCINE, CLINICAL PICTURE AND ATOPIC DERMATITIS– CASE REPORT

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ABSTRACT: The presentation of this case is a dog of the age of 7 years. The clinical picture of this dog was monitored periodically for years on an outpatient basis before a definitive diagnosis of Atopic Dermatitis was made. The symptoms included ear reactions with mixed infections and swelling, increased secretion from the eyes and nose with occasional changes in the skin. Diarrhoea was rarely present. After the annual vaccine, the dog showed a rapid allergic reaction after four years of regular rabies supplementary vaccines. The same reaction was after two years on the rabies vaccine again. During the allergy testing, other diagnostics were also performed according to usual procedures. In the haematology findings of the complete blood count, changes were recorded in white blood cell parameters: lymphopenia, neutropenia, eosinopenia and monocytosis. A positive reaction to both types of allergens was confirmed by serological testing using the Elisa method of dog serum for inhalant allergens and food allergens. A positive and highly positive result was obtained for even 20% of inhalant allergens and 37.71% of food allergens. A positive reaction to an external (OUT) allergen group of 15% included pollen while 5% of positive reactions included indoor (IN) allergen group. The total number of tested allergens is 40+21 on a large panel. Screening Elisa method is recommended for easier and faster determination of therapy in patients with Allergic Dermatitis (AD).
KEYWORDS: Atopic Dermatitis. Dog. Food allergens. Inhalant allergens. SAT.

1 INTRODUCTION

Atopic dermatitis (AD) is a genetically predisposed skin disease, related to IgE-mediated hypersensitivity. The serum allergen-specific IgE test (SAT) is used to detect allergen-specific IgE antibodies. This type of test identifies inhaler allergens from the external (OUT) and internal (IN) environment (Tommaso et al 2021), and food allergens. Serum is used as a test sample. According to the conducted clinical studies, this type of allergy testing is indicated in the case of a visible clinical picture or after monitoring the patient at the veterinarian for a long period (Hensel et al 2015, Tomasso et al

2021). Depending on the type of allergen, which can be seasonal, external, internal or food allergen, clinical signs can be manifested at different times of the year. The external (OUT) group of inhalant allergens most often produces clinical signs in pets when pollen is present in the air during the flowering period. Groups of internal (IN) inhalant allergens (dust mites) cause symptoms in pets during the most intensive period of stay in the house (Chermprapai and Thengchaisri 2021) during rainy days, in winter, in a subtropical climate. From other groups of allergens such as those from food - symptoms can appear at any time of the year (Hensel et al 2015). The clinical picture of such pets without adequate therapeutic control and a medicated diet can be very frustrating for the owner until a definitive diagnosis is made. Such examinations are expensive and require patience in monitoring the patient as well as trust with financial support from the owner.

2 MATERIAL AND METHODS

2.1 ANAMNESTIC AND CLINICAL FINDINGS

The presentation of this case in a dog of the age of 7 years includes a clinical picture with applied diagnostics. The clinical picture of a pug dog was monitored periodically for 4 years on an outpatient basis before a definitive diagnosis of Atopic Dermatitis was made. The symptoms included ear reactions (otitis externa) with mixed infections and swelling, increased secretion from the eyes and nose with occasional changes on the skin. Diarrhoea was rarely present. According to the applied regular annual vaccine Nobivac Rabies (MSD) dose of 1 ml, the dog showed a rapid allergic reaction after four years of regular booster vaccines. The same reaction was on the vaccine Rabigen mono (Virbac) dose of 1 ml. Tests and examinations of the complete blood count, and biochemical parameters were performed after the first allergic reaction. In the findings of the complete blood count, changes were recorded in most white blood cell parameters manifested as lymphopenia, neutropenia, and eosinopenia, while monocytes were seven times more elevated compared to reference values. A positive reaction was confirmed by serological testing using the Elisa method of the dog's serum to inhalant allergens and food allergens.

2.2 BLOOD SAMPLING

Venous blood was taken from the jugular vein or other accessible veins by venipuncture. Test tubes with anticoagulant EDTA were used for complete blood count testing, while test tubes with a separator were used for biochemistry sampling. For a complete blood count, 0.5-1 ml of blood was sampled. To obtain serum from test tubes

with a separator for biochemistry 3-4 ml of blood was sampled. To separate the serum, a centrifuge is used at 3000 revolutions in a time interval of 10-15 minutes.

2.3 COMPLETE BLOOD COUNT ANALYSIS

Examined parameters in the complete blood count after sampling: total leukocyte (WBC), neutrophils, lymphocytes, monocytes, eosinophils, basophils, LUC, percentage of neutrophils, percentage of lymphocytes, percentage of monocytes, percentage of basophils, erythrocytes (RBC), mean corpuscular haemoglobin (MCH), hematocrit (Hct), mean corpuscular volume (MCV), mean corpuscular haemoglobin concentration (MCHC), haemoglobin, platelets (PLT), mean platelet volume (MPV) and platelet crit (PCT). The tests were carried out on the Advia 120 device, Siemens in the Vetlab laboratory, in Belgrade.

2.4 COMPLETE BIOCHEMICAL TESTS

Tested parameters of biochemistry: total proteins, globulins, albumins, A/G, total bilirubin, cholesterol, glucose, urea, creatinine, U/C, ALP, alpha-amylase. Tests were performed on the AU480, Beckman Coulter in the Vetlab laboratory, Belgrade.

2.5 DETECTION OF ANTIBODIES IN THE SERUM OF TESTED DOG

Standardised Elisa method testing with manufacture prescription (Invitros Laboratorios, Madrid, Spain) was used to detect a positive test for the investigated allergens in dog sera. The method is standardised in Spain. The large panel consists of 40 sites for testing inhalant allergens and 21 sites for testing allergens of food origin. Wells (eng. wells) possess specific antigenic proteins, which in the case of the presence of specific antibodies of the IgE class lead to a positive test reaction. The test result is semi-quantitative, and depending on the intensity of the reaction, the obtained values are classified as negative, borderline, positive and highly positive. Before reading the reaction, three well solutions are used and washed at different time intervals. After the last washing of the basin, the reaction is read 18 hours after the end of incubation. Absorbance reading is performed on a spectrophotometer (Thunderbolt, Gold Standard Diagnostics, USA) at two wavelengths, 405 nm and 650 nm. The classification of reaction intensity is determined based on the reading value of the calibrator of known concentration within each individual reading. The total number of allergen tests is 40+21 in a large panel. The panel of inhalant allergens includes 40. The panel of food allergens includes a total of 21. The test was performed in the accredited veterinary laboratory for clinical diagnostics Vetlab in Belgrade, Serbia.

Tested inhaler allergens

OUT serum allergen-specific IgE tests: Phleum pretense, Dactylis glomerata, Poa pratendis, Lolium perenne, Cynodon dactylon, Cerealas mixture, Rumex crispus, Taraxacum vulgare, Artemisia vulgaris, Plantago lanceolata Parietaria judaica, Chenopodium álbum, Urtica dioica, Ambrosia elatior, Brassica spp., Platanus hybrida, Olea europaea, Betula alba, Corylus avelina, Alnus glutinosa, Lingustrum vulgare, Pinus spp., Populus alba, Querqus rubor, Ulmus campestre, Salix spp and Cupressus arizonica.

IN serum allergen-specific IgE tests: Dermatophagoides farinae, Dermatophagoides pteronyssinus, Acarus siro, Tyrophagus putrescentiae, Lepidogliphus destructor, Alternaria alternata, Aspergillus niger, Penicillium notatum, Mucor spp, Cladosporum herbarum, Malassezia, Staphylococcus, and fleas.

Tested food allergens

Serum allergen-specific IgE tests for food: beef, turkey, chicken, pork, lamb, duckling, rabbit, venison, mixed fish, egg, cow's milk, wheat, oats, rice, soy, corn, beetroot, carrots, potatoes, peanuts, yeast.

3 RESULTS

3.1 CLINICAL FINDINGS

Pictures of earlier clinical findings in a dog with ear changes (Picture 1) and on the skin coat (Picture 2). The appearance of a dog with an allergic reaction after the administration of a regular vaccine with generalized tissue swelling on the dog's face (Picture 3 and Picture 4).

Picture 1. Otitis externa on the pug dog.

Picture 2. The skin on the pug dog.

Picture 3. Pug dog facefter the booster vaccine.

Picture 4: Generalized tissue a swelling on the dog's face.

3.2 COMPLETE BLOOD COUNT ANALYSIS AND BIOCHEMICAL TESTS

The ratio of reference and obtained values after the analysis of complete blood count (Figure 1). In the haematology findings of the complete blood count, changes were recorded in white blood cell parameters: lymphopenia, neutropenia, eosinopenia and while mocytosis. The ratio of reference and obtained values after the analysis of the complete biochemical test is shown in Figure 2.

Figure 1. Ratio haematology reference values and analyze values in dog.

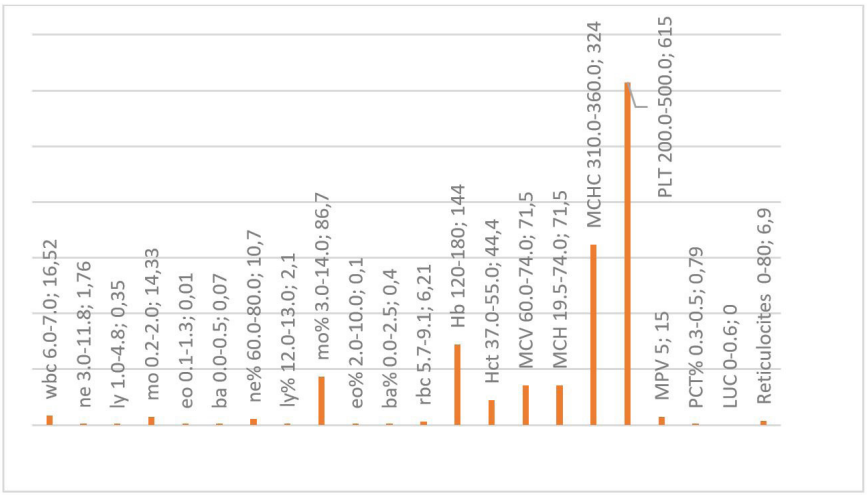


Figure 2. Ratio biochemistry values and analyze values in AD dog.

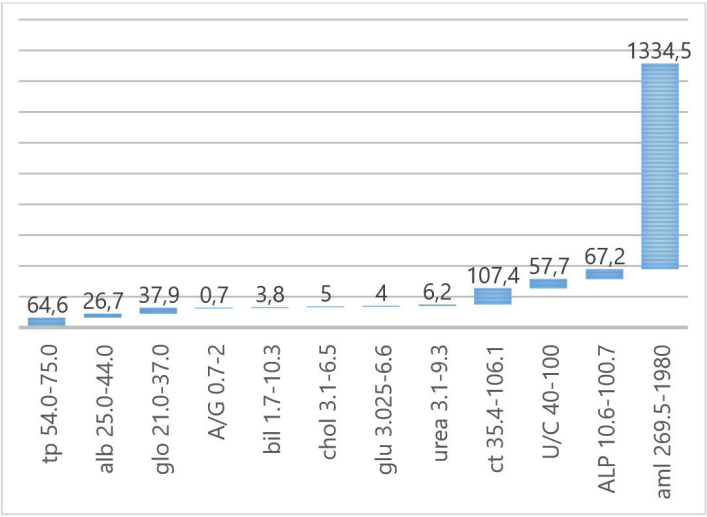


Figure 1, Legend: wbc-total leukocyte, ne-neutrophils, ly-lymphocytes, mo-monocytes, eo-eosinophils, ba-basophils, ne%-percent neutrophils, ly%-percent lymphocytes, mo%-percentage monocytes, ba%-percentage basophils, rbc-erythrocytes, LUC, MCH-mean corpuscular haemoglobin, Hct-hematocrit, MCV-mean corpuscular volume, MCHC-mean corpuscular haemoglobin concentration, Hb-haemoglobin, reticulocytes, PLT-platelets, MPV-mean platelet volume and PCT-platelet crit.

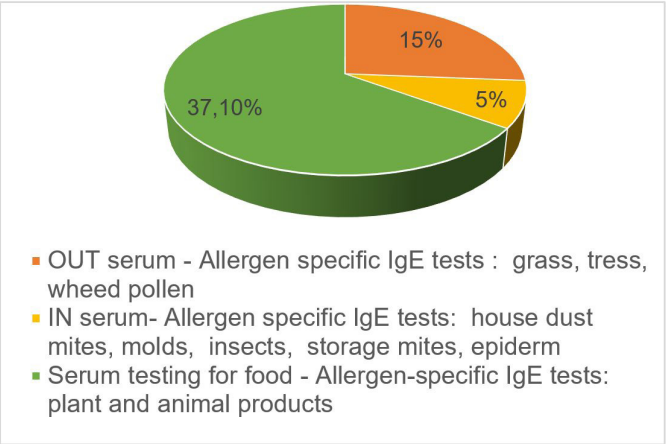
Figure 2, Legend: tp-total proteins, glo-globulins, alb-albumins, glu-glucose, A/G-albumin globulin ratio, bil-total bilirubin, chol-cholesterol, urea, ct-creatinine, U/C- ratio urea creatinine, aml-alpha amylase, ALP.

3.3 DETECTION OF ANTIBODIES IN THE SERUM OF TESTED DOG

The aforementioned tests for the presence of specific antibodies of the IgE class included in the large panel represent inhalant allergens and allergens originating from food

in the said dog (Figure 3). The dog showed a positive reaction to allergens from the pollen group *Quercus robur*, *Phleum pretense, Dactylis glomerata, Poa pratensis, Lolium perenne, Cynodron dactylum, while* within the group of indoor allergens *Dermatophagoides farina, Dermatophagoides pteronyssinus*. The dog showed positivity in the following investigated food allergens: rice, oats, corn, wheat, fish and mixed fish, beef, turkey, and lamb.

Figure 3. Serum allergen-specific IgE tests (SAT) and percentage of positive reactions in pug dog with AD.



4 DISCUSSION

Positive findings on allergens in the tested pug dog were obtained after the serum test was performed during the existence of the clinical picture. Positive findings in dogs in preliminary studies in Italy (Tomasso et al 2021) also gave positive reactions during the examination performed during the existence of the clinical picture. The diagnosis is based on the anamnesis, the clinical picture and according to some authors, also on the presence of itching (Marsella, De Benedetto 2017). In the dog that is being described, itching as a clinical symptom was not significant. Comparisons with other studies are made difficult by the circumstances of pollen distribution in different geographical areas, climate changes, plant characteristics, seasons (Tomasso et al 2021). The positive inhalatory allergens of the tested dog belong to the group of external (OUT) allergens, mostly grass pollen and one tree pollen. When it comes to pollen reactions to grasses, and especially the grass *Cynodron dactylum*, it can be said that a positive reaction in AD dogs was recorded in up to 60%, for example, in Italy (Tomasso et al 2021), while in the examined dog it was measured as highly positive. In some studies, they report that allergic reactions in dogs to inhalant internal allergens are much more common (Chermprapai and Thengchaisri 2020). In a study in Norwegian dogs, it was determined that an allergic

reaction to the mites *D. farinae* and/or *D. pteronyssinus* (Bjelland et al 2014) was present in a higher percentage. A positive reaction was obtained in the results of the tested dog to the mentioned mites. Trials in Europe and the USA have produced similar results in some studies. Practical guidelines for diagnosis exclude other diseases with an applied diagnosis of allergen-specific immunoglobulin E in serology (ASIS). Food reactions can be caused in two ways non-immune mediated (food intolerance) and immune-mediated which includes IgE-mediated hypersensitivity (food allergy) (Hillier, Griffin 2001). Reactions to food components can be clinically presented as Atopic Dermatitis (AD) and common gastrointestinal signs are diarrhoea, vomiting, soft stools, flatulence **(**Picco et al 2008, Favrot C et al 2010, Hensel et al 2015). Vaccination regimens, parasitic diseases, and glucocorticosteroids are some of the factors that can affect the level of IgE in the serum (DeBoer, Hillier 2001). Vaccination in the examined dog was reflected in a strong sensitisation of the organism. Such tests are effective because they do not require sedation, reduce the risk of anaphylactic shock, and do not immediately affect the therapy given to the patient (Chermprapai and Thengchaisri 2020).

5 CONCLUSION

Screening Elisa method is recommended for easier and faster determination of therapy in patients with Allergic Dermatitis (AD).

6 ACKNOWLEDGMENT

There is no conflict of interest.

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SOBRE O ORGANIZADOR

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