ABANICO VETERINARIO E-ISSN 2448-6132 abanicoveterinario@gmail.com
Creative Commons (CC BY-NC 4
https://abanicoacademico.mx/revistasabanico-version-nueva/index.php/abanico-veterinario

Abanico Veterinario. January-December 2022; 12:1-13. http://dx.doi.org/10.21929/abavet2022.231
Research Note. Received:14/07/2022. Accepted:26/10/2022. Published:02/12/2022. Code: e2022-45.
https://www.youtube.com/watch?v=KNjfhacdsKQ

Partial resistance of *Pasteurella multocida* isolated from rabbits to lysis induced by the complement system

Supervivencia de *Pasteurella multocida* aislada de conejo a la lisis inducida por el sistema complemento

Alcaraz-Sosa Elena*[1, 2] ID , Carmona-Gasca Carlos[3] ID, Salgado-Moreno Socorro[3] ID, Sahagún-Ruíz Alfredo[1] ID**

[1]Laboratorio de Inmunología Molecular, Departamento de Microbiología e Inmunología, Facultad de Medicina Veterinaria y Zootecnia, Universidad Nacional Autónoma de México, Av. Universidad 3000, Ciudad Universitaria, Coyoacán, C.P. 04510, Ciudad de México, México. [2]Departamento de Producción Agrícola y Animal, Universidad Autónoma Metropolitana, Unidad Xochimilco, Medicina Veterinaria y Zootecnia, Calzada del Hueso 1100, Colonia Villa Quietud, Coyoacán, 09460, Ciudad de México, México. [3]Unidad Académica de Medicina Veterinaria y Zootecnia, Universidad Autónoma de Nayarit, Km 3.5 Carretera Compostela-Chapalilla, Compostela, Nayarit, México CP 63700 *Responsible autor: Elena Alcaraz Sosa. **Author for correspondence: Alfredo Sahagún Ruiz. E-mail: ealcaraz@correo.xoc.uam.mx, carmonagasca@uan.edu.mx, socorro.salgado@uan.edu.mx, sahagun@unam.mx

ABSTRACT

Pasteurellosis is one of the most serious bacterial diseases of rabbits, causes considerable economic damage in production systems worldwide and is a widely proven zoonosis with cases in humans infected by companion animals. The activated complement system causes lysis of various pathogenic microorganisms by the formation of the membrane attack complex. The objective of the present work was to evaluate the survivability of a virulent isolate of *Pasteurella multocida* type A to human serum complement activity. A female rabbit was referred as a clinical case of pneumonic pasteurellosis, a nasal swab was performed for isolation and identification. *P. multocida* was isolated in pure culture, characterized by standard biochemical identification tests, and then challenged for 30 min at 37 °C with normal serum and heat-inactivated serum. Lysis by complement action had a significant reduction in the viability of the microorganism with respect to the culture challenged with inactivated complement ($p< 0.05$), however, a significant percentage 51.07 ± 1.70 % managed to evade complement lytic activity. In the context of health, it is concluded that the complement system offers partial protection against infections by *P. multocida* serotype A causing pneumonic pasteurellosis in rabbits.

Keywords: Pasteurellosis, developmental inhibition, bacteria, membrane attack complex.

RESUMEN

La pasteurelosis es una enfermedad bacteriana que provoca daños económicos en sistemas de producción cunícola de todo el mundo y es una zoonosis comprobada en humanos infectados por los animales de compañía. El sistema complemento causa la lisis de diversos microorganismos patógenos formando el complejo de ataque a la membrana. El objetivo del presente trabajo fue evaluar la actividad del complemento sérico humano sobre la capacidad de supervivencia de un aislado virulento de *Pasteurella multocida* tipo A. La bacteria se obtuvo de un caso clínico de una coneja con neumonía, por medio de un hisopado nasal para aislamiento en cultivo puro e identificación por pruebas bioquímicas. *P. multocida* fue

enfrentada por 30 min a 37°C, a suero humano normal y a suero humano inactivado por calor. La lisis por la acción del complemento tuvo una reducción significativa en la viabilidad del microorganismo comparado con el cultivo enfrentado al complemento inactivado (P< 0.05), sin embargo, el 51.07 ± 1.70 % logró evadir la actividad lítica del complemento. En la presente investigación se concluye que el sistema complemento ofrece protección parcial contra infecciones por *P. multocida* serotipo A causante de pasteurelosis neumónica en conejos.
Palabras clave: Pasteurelosis, inhibición del desarrollo, bacteria, complejo de ataque a la membrana.

INTRODUCTION

Pasteurella multocida is a Gram-negative bacterium that affects a wide range of domestic species. It is a pathogen that is part of the normal microbiota of oral cavity, oropharynx and upper respiratory tract, acts as a primary and opportunistic agent (Aktories *et al*., 2012; Register & Brockmeier, 2019). In leporidae it causes acute and chronic bronchopneumonia, purulent atrophic rhinitis, sinusitis, atrophy of the nasal turbinates, distortion of the maxilla, it is also associated with otitis media, genital infection, pyoderma, septicemia and affects in their growth (Massacci *et al*., 2018; D'Amico *et al*., 2022).

Pasteurellosis is one of the most serious bacterial diseases of rabbits causing economic damage to production systems worldwide (El-Sheikh *et al*., 2021). This disease is usually endemic in rabbit production and its prevalence has been estimated between 7 and almost 100 % (Zhu *et al*., 2020). Disease occurrence is inevitable in any animal production unit and leads to economic losses (Quesada *et al*., 2013). Rabbits can become infected with *P. multocida* after birth and the incidence of infection increases with age up to about 5 months, with this, most adult rabbits have been infected and are carriers of *P. multocida* (Palócz *et al*., 2014).

Anthropozoonotic transmission occurs through animal bites, scratches, and licking of skin abrasions or contact with nasal secretions, not to mention that *P. multocida* is the most frequent isolate observed in human infections (Souza, 2009; Wilson & Ho, 2013; Abreu *et al*., 2018; D'Amico *et al*., 2022). The prevalence of antisera against *P. multocida* was 2-fold higher in healthy individuals with occupational exposure (Wilson & Ho, 2013), indicating that exposure to animals increases the likelihood of infection and in almost all cases death appears to be the result of a complication of infection acquired through contact with animals (Hey *et al*., 2012). The use of rabbits as companion animals exposes owners to *P. multocida* infection; the above has been documented but the issue has not been determined as a public health problem (Lin *et al*., 2006; Per *et al*., 2010; Ferreira *et al*., 2016; D'Amico *et al*., 2022).

The complement system is constituted by more than 50 circulating proteins that are produced in the liver, extrahepatic biosynthesis occurs in fibroblasts, T and B cells, adipocytes and endothelial cells. Complement generates an enzymatic chain reaction that can be activated by three pathways: the classical pathway, the alternate pathway and the lectin pathway (Bajic *et al*., 2015; Mathern & Heeger, 2015), participates in the immune response and it is activated by the recognition of molecular patterns associated with pathogens, damaged cells and immune complexes (Bajic *et al*., 2015). To eliminate such pathogens, the complement cascade triggers their opsonization that facilitates their phagocytosis, their lysis by the polymerization of the membrane attack complex, as well as a pro-inflammatory response leading to the recruitment and activation of immune cells of both the innate immune response, as well as the adaptive immune response (Ricklin *et al*., 2010; Bajic *et al*., 2015; Mathern & Heeger, 2015).

The current trend for the treatment of pasteurellosis is based on the application of antibiotics, but in recent years there was an increased interest in having alternative therapies for its treatment (Kubatzky, 2012; Moreno-Torres *et al*., 2019). The complement system effect and its modulation to control the development of various infectious microorganisms have been demonstrated, but serum complement activity against *P. multocida* isolated from rabbit has not been evaluated. Therefore, the present study aims to evaluate the activity of human serum complement on the survivability of a virulent *P. multocida* type A isolate.

MATERIAL AND METHODS

A six-month-old female "cabeza de león" rabbit (*Oryctolagus cuniculus*) presenting semiology compatible with productive pneumonia, with moderate whitish secretion from the nasal cavities, was treated with enrofloxacin orally for more than three months. The secretion was swabbed for isolation of the causative agent in blood agar (AS) and incubated for 24 h at 37 ºC. After isolation in pure culture, characterization was performed by standard phenotypic identification techniques and for the identification of the capsular type, the hyaluronidase and acriflavine test was performed following the methodologies described by Koneman & Allen (2008). The procedures contained in the present work were endorsed by the Internal Committee for the Care and Use of Animals (CICUA) trade DC-2016/2-1.

For survival trial, the bacteria were subcultured on blood agar (BA) for 24 h, subsequently a colony was inoculated in brain heart infusion broth (BHIB) for 16 h at 37 °C with 185 rpm shaking until the culture reached an optical density of 0.43 at 600 nm (approximately 3.42 × 10^8 bacteria/mL). It was then centrifuged at 4,400 *Xg* at 21 °C for 15 min, the bacterial pellet was washed once with phosphate buffered saline (PBS) and adjusted to 1x10^3 bacteria/tube. Normal human serum (NS) with active complement was used and for control complement was inactivated by heating human serum at 56 °C for 30 min (ICS),

the final serum concentration was 40 %. Bacteria were incubated with NS or ICS at 37 °C for 30 min under 190 rpm agitation. Then the culture was centrifuged at 5,500 Xg decanting the supernatant, the pellet was suspended in 100 µL of PBS, tenfold dilutions up to 10-4 were performed, to seed by extension in BA in triplicate incubating at 37 °C for 18 h to determine the number of colony forming units (CFU), the survival percentage was calculated considering the number of colonies obtained after incubation with ICS as 100 % survival.

The results of the survival test were analyzed using mean, range and standard deviation, they were also compared using the one-way ANOVA test where a P value < 0.05 was interpreted as significant. The above was performed using GraphPad Prism software version 6.0.0 for Windows (GraphPad Prism, 2012).

RESULTS

The bacterium was isolated in pure culture and abundantly in BA from the nasal swab of the "cabeza de león" rabbit (*Oryctolagus cuniculus*) developing within 24 h of incubation. Gram staining was then performed, Gram-negative bacilli were observed and with Giemsa staining bacilli with bipolar appearance were observed. Biochemical tests performed are described in Table 1 and the images in Figure 1. The identification of the causal agent was *P. multocida* and by the hyaluronidase test it was subclassified in capsular type A.

Table 1. Standard biochemical tests and antibiogram applied to the *P. multocida* isolate obtained from the diseased rabbit

IDENTIFICATION TEST	Result	SEROTYPE IDENTIFICATION	Result
Oxidase	+		
TSI Agar	6	Acriflavin	-
Motility	-	Hyaluronidase	+
Glucose	+		
Development on blood agar	+	**ANTIBIOGRAM**	
Hemolysis	-	Cefotaxime	S
Development on MacConkey	-	Ciprofloxacin	S
Nitrate reduction	+	Neomycin	R
Indole	+	Norfloxacin	S
Urea	-	Sulfamethoxazole-trimethoprim	S
Simmons Citrate	-	Imipenem	S

Positive (+), Negative (-), Sensitive (S), Resistant (R)

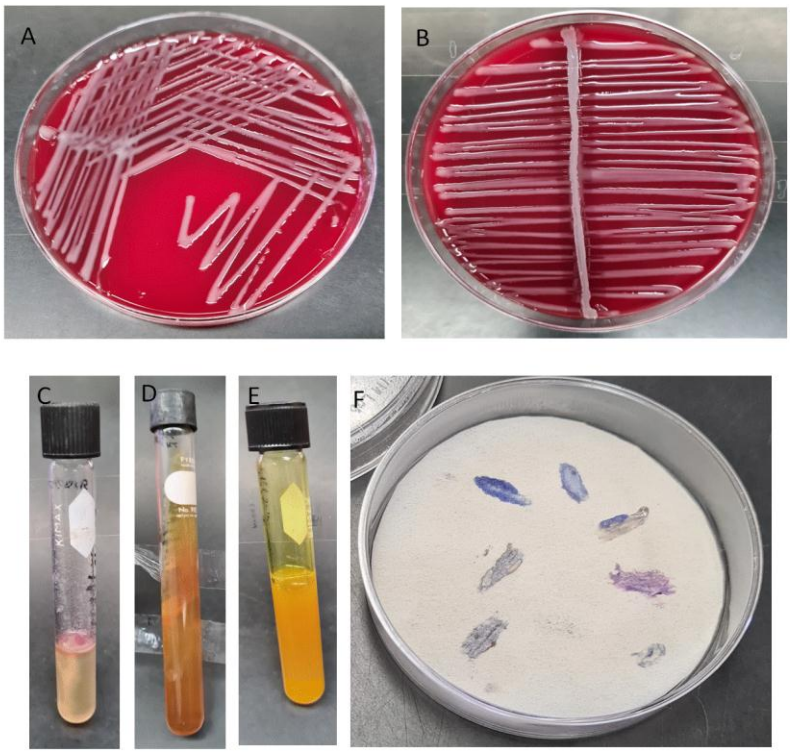

Figure 1. Biochemical tests applied for the identification of *P. multocida* isolated from the nasal swab of "cabeza de león" rabbit. A) Pure culture on blood agar plate without hemolysis. B) Positive hyaluronidase test. C) SIM test; indole positive, hydrogen sulfide production negative and motility negative. D) TSI test; negative gas production, negative hydrogen sulfide production and yellow background and surface. E) Acriflavine test negative. F) Positive oxidase test

The effect of the complement system on *P. multocida* was evaluated after incubation of the bacterial culture with NS or with 40 % ICS for 30 min. In ten independent experiments the viable bacteria were quantified, the percentage of surviving cells was calculated by comparing the number of viable bacteria incubated in the two treatments. Results indicate that the serum complement system has bactericidal action on *P. multocida*; the trial showed that the culture treated with the NS allowed *in vitro* survival of 51.07 ± 1.70 %, with respect to the ICS ($P<0.05$) (Figure 2).

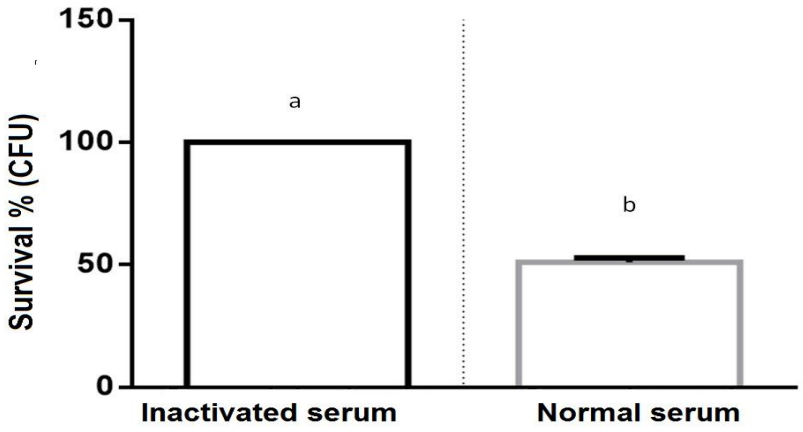

Figure 2. Survival of *P. multocida* to the action of the complement system present in the serum. The number of *P. multocida* CFUs that developed after incubation with ICS is shown on the left and on the right when incubated with NS. ICS= serum inactivated at 56 ºC for 30 min. NS= normal serum (n=10). [a-b] Means with different letters are statistically different (P<0.05)

DISCUSSION

The presence of *P. multocida* capsular type A was confirmed in a "cabeza de león" rabbit (*Oryctolagus cuniculus*) that only presented whitish discharge from the nostrils. Commonly observed signs and lesions are rhinitis, sinusitis, conjunctivitis, dacryocystitis, nasal and ocular discharge. Other non-respiratory findings are meningitis, dermatitis and pyometra which the patient did not present (Rosell & De La Fuente, 2016; Massacci *et al*., 2018). *P. multocida* is the bacterium responsible for pasteurellosis in rabbits (Aktories *et al*., 2012; Wilson & Ho, 2013), despite the diversity of signs and lesions that these animals may present, this disease should be suspected even with little respiratory signology.

The tendency to use rabbits as companion animals has increased; their coexistence with humans is closer compared to rabbits destined for production; therefore, their health status should be monitored to avoid zoonotic diseases (D'Amico *et al*., 2022). Pasteurellosis causes high morbidity and mortality in rabbits regardless of their zootechnical purpose, whether in production, as laboratory animals or as companion animals (Aktories *et al*., 2012; D'Amico *et al*., 2022), therefore, surveillance for pasteurellosis in rabbits that live closely with humans should be monitored.

Capsular type A is mostly isolated in cases of pasteurellosis in rabbits, and has also been reported to cause the disease in other mammalian animal species such as goats, sheep, pigs and cattle (Soriano-Vargas *et al*., 2012). Capsular type A is also closely related to fowl cholera worldwide in various avian species such as turkeys, chickens (Harper *et al*., 2012; Guan *et al*., 2020), ducks (Soriano-Vargas *et al*., 2012) and other wild birds (Wilson & Ho, 2013).

Internationally there are trends to identify *P. multocida* by molecular tests such as PCR, but identification by standard phenotypic techniques is reliable to provide a definitive characterization (Dziva *et al*, 2008). This consists of isolation of the bacterium on 5 % ram BA or brain and heart infusion agar (BHIA), no development on MacConkey agar, microscopic observation of pleomorphic, non-flagellated, Gram-negative, bipolar staining short bacilli with Giemsa stain, which develop as facultative aerobes at 37 °C (Dziva *et al*., 2008; WOAH, 2012; Panna *et al*., 2015; D'Amico *et al*., 2022). Biochemical tests result in positive catalase, oxidase and indole, sucrose, glucose and maltose fermentation (Dziva *et al*., 2008; WOAH, 2012; Wilson & Ho, 2013; Panna *et al*., 2015). The above associated with the presentation of the disease in rabbits, is sufficient to definitively characterize the bacterium (Dziva *et al*., 2008), as observed in the isolate, where in addition the positive hyaluronidase test was observed to identify it as *P. multocida* capsular type A.

The antibiogram performed after identification shows that the *P. multocida* isolate is susceptible to cefotaxime, ciprofloxacin, norfloxacin, sulfamethoxazole-trimethoprim and imipenem and resistant to neomycin; the clinical history refers that the patient was treated with enrofloxacin for more than 3 months without any improvement. Palócz *et al*. (2014) reports that enrofloxacin is a good antibiotic against *P. multocida* infections, but that the oral route of administration does not show good effect. Bourély *et al*. (2019) and Jamali *et al*. (2014) also report that strains isolated from pigs and rabbits are sensitive, but those isolated from cattle and ducks, are resistant. Antibiotic sensitivity and resistance to antibiotics against *P. multocida* is highly variable, this depends largely on the concentrations of the bacterium, antibiotic dosages and animal species.

P. multocida capsular type A isolates of all species are sensitive to a wide range of antibiotics such as florfenicol, trimethoprim-sulfamethoxazole and penicillins (Gunathilake *et al*., 2015). Contrary to the above, it is reported that bovine isolates, show marked resistance to penicillin G, streptomycin, oxytetracycline, ampicillin and thiamphenicol (Jamali *et al*., 2014), and they are susceptible to amoxicillin, amikacin, cefazolin, ceftiofur, cefquinome, chloramphenicol, enrofloxacin, florphenicol and kanamycin (Jamali *et al*., 2014; Bourély *et al*., 2019). For greater efficacy against *P. multocida*, the antibiotics of choice should be administered in combination. The best combinations are with amoxicillin and clavulanic acid, doxycycline with metronidazole, clindamycin with ciprofloxacin or trimethoprim-sulfamethoxazole or ceftriaxone combination (Wilson & Ho, 2013). The choice of antibiotics in rabbits is limited, as many oral antibiotics affect the intestinal microbiota, leading to infections such as clostridiasis (Elazab *et al*., 2018). Given the variability of susceptibility and resistance shown by *P. multocida* it is very important that before administering antibiotic therapy, antibiotic susceptibility testing should always be performed.

Blood plasma has elements capable of protecting the organism from the presence of pathogens, among them the complement system, in this study the inhibitory effect of serum complement against *P. multocida* was observed. The inhibition of the development of *P. multocida* in the NS by the action of complement was demonstrated in contrast to ICS, where the proteins of the complement system were denatured. The complement system is the main humoral response of the innate immune system and has great relevance in protection against bacteria and other invading pathogens due to its opsonic and cytolytic activity on the pathogen, in addition to its proinflammatory activity (Carroll & Isenman, 2012). Even though the complement system is shown to have bactericidal action against *P. multocida*, there are other components in the blood that have inhibitory action on bacterial growth (Nicholson, 2016). The ability of the individual to control the infection also depends on other components of the immune system such as the production of IgA antibodies that inhibit the infection and the development of the bacterium in the respiratory mucosa, while, in its invasion to tissues, it depends on the production of IgG antibodies together with the activation of the Complement system (Aktories *et al*., 2012).

Encapsulated strains of *P. multocida* serogroup A of avian origin are highly complement resistant and actively grow in serum (Harper *et al*., 2012; Guan *et al*., 2020). However, the *P. multocida* isolate obtained from the rabbit was partially sensitive to complement action as is the case with spontaneous mutants with reduced capsule, or strains that have been treated with hyaluronidase, which are generally more susceptible to complement- and neutrophil-mediated serum agglutination, destruction and phagocytosis of serum (Guan *et al*., 2020; Li *et al*., 2021). *Rodentibacter pneumotropicus* (formerly *Pasteurella pneumotropica*) survives by 50 % when incubated with NS, so *R. pneumotropicus* has evolved mechanisms to evade the human complement system that may increase efficiency to access and colonize internal tissues where it can cause serious infections (Sahagun-Ruiz *et al*., 2014).

P. multocida infections in humans arise as a result of wounds where rabbit bites or scratches are involved evidencing anthropozoonosis (Lin *et al*., 2006; Per *et al*., 2010; D'Amico *et al*., 2022). When these infections do not result from bite wounds, they are usually related to contact of skin, naso-oropharyngeal or upper respiratory mucosal lesions, especially in young children, the elderly, pregnant or immunocompromised individuals (Baud *et al*., 2012; Wilson & Ho, 2013, D'Amico *et al*, 2022). Rabbits used for domestic use that are carriers of *P. multocida* are a potential source of infection for humans exposed to infected rabbits, such as: veterinary doctors, owners, breeders, slaughterhouse personnel who are in contact with them.

CONCLUSIONS

There is a reduction in the number of *P. multocida* CFUs when exposed to active complement present in normal human serum compared to those exposed to inactivated serum. Human complement is a component of the immune system that offers partial resistance to the development of *P. multocida* capsular type A.

ACKNOWLEDGMENTS

This research was funded by project PAPIIT IN221021 DGAPA-UNAM awarded to Alfredo Sahagún Ruiz. In the same vein, the Mexican National Council of Science and Technology (CONACyT) granted a doctoral scholarship to Luz Elena Alcaraz Sosa (grant 168668).

CITED LITERATURE

ABREU F, Rodríguez-Lucas C, Rodicio MR, Vela AI, Fernández-Garayzábal JF, Leiva, PS, Fernández J. 2018. Human *Pasteurella multocida* infection with likely zoonotic transmission from a pet dog, Spain. *Emerging Infectious Diseases*. 24(6):1145. ISSN: 1080-6059. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6004854/

AKTORIES K, Orth JH, Adler B. 2012. *Pasteurella multocida*. *Current Topics of Microbiology and Immunology.* 361:1. ISSN: 0070-217X. http://www.ncbi.nlm.nih.gov/pubmed/23230602

BAJIC G, Degn SE, Thiel S, Andersen GR. 2015. Complement activation, regulation, and molecular basis for complement-related diseases. *Journal of the European Molecular Biology Organization.* 34(22):2735-2757. ISSN: 0261-4189. https://doi.org/10.15252/embj.201591881

BAUD D, Bizzini A, Jaton K, Achtari C, Prod'hom G, Greub G. 2012. *Pasteurella multocida* zoonotic ascending infection: an unusual cause of tubo-ovarian abscess. *Vector-Borne and Zoonotic Diseases*. 12(1):84-85. ISSN: 1530-3667. https://doi.org/10.1089/vbz.2011.0610

BOURÉLY C, Cazeau G, Jouy E, Haenni M, Madec JY, Jarrige N, Gay E. 2019. Antimicrobial resistance of *Pasteurella multocida* isolated from diseased food-producing animals and pets. *Veterinary Microbiology*. 235:280-284. ISSN: 0378-1135. https://doi.org/10.1016/j.vetmic.2019.07.017

CARROLL MC, Isenman DE. 2012. Regulation of humoral immunity by complement. *Immunity*. 37(2):199-207. ISSN: 1074-7613. https://doi.org/10.1016/j.immuni.2012.08.002

D'AMICO F, Casalino G, Bozzo G, Camarda A, Lombardi R, Dimuccio MM, Circella E. 2022. Spreading of *Pasteurella multocida* Infection in a Pet Rabbit Breeding and Possible Implications on Healed Bunnies. *Veterinary Sciences*. 9(6):301. ISSN: 2306-7381. https://doi.org/10.3390/vetsci9060301

DZIVA F, Muhairwa AP, Bisgaard M, Christensen H. 2008. Diagnostic and typing options for investigating diseases associated with *Pasteurella multocida*. *Veterinary Microbiology.* 128(1-2):1-22. ISSN: 0378-1135. https://doi.org/10.1016/j.vetmic.2007.10.018

EL-SHEIKH SM, Youssef FM, Mohamed HI, El-Saber Batiha G, Albrakati A, Galal AA. 2021. Efficacy of grape seed hydro-alcoholic extract in the treatment of experimentally *Pasteurella multocida* infected rabbits. *Veterinary Medicine and Science*. 7(3):923-934. ISSN: 2053-1095. https://doi.org/10.1002/vms3.446

ELAZAB ST, Schrunk DE, Griffith RW, Ensley SM, Dell'Anna G, Mullin K, Hsu WH. 2018. Pharmacokinetics of cefquinome in healthy and Pasteurella multocida-infected rabbits. *Journal of veterinary pharmacology and therapeutics*. 41(3):374-377. ISSN: 1365-2885. https://doi.org/10.1111/jvp.12489

FERREIRA TSP, Moreno LZ, Felizardo MR, de Gobbi DDS, Filsner PHdLN, de Moura Gomes VT, Moreno AM. 2016. Pheno-and genotypic characterization of *Pasteurella multocida* isolated from cats, dogs and rabbits from Brazil. *Comparative Immunology, Microbiology and Infectious Diseases.* 45:48-52. ISSN: 2356-6140. https://doi.org/10.1016/j.cimid.2016.02.004

GRAPHPAD Prism. 2012. GraphPad Software®. version 6.0.0 for Windows, San Diego, California USA. www.graphpad.com

GUAN L, Zhang L, Xue Y, Yang J, Zhao Z. 2020. Molecular pathogenesis of the hyaluronic acid capsule of *Pasteurella multocida*. *Microbial pathogenesis*. 149:104380. ISSN: 0882-4010. https://doi.org/10.1016/j.micpath.2020.104380

GUNATHILAKE R, Verma A, Caffery M, Sowden S. 2015. *Pasteurella multocida* peritonitis after cat scratch in a patient with cirrhotic ascites. *Infectious Disease Reports.* 7(2):5937. ISSN: 2036-7449. https://doi.org/10.4081/idr.2015.5937

HARPER M, Boyce JD, Adler B. 2012. The Key Surface Components of *Pasteurella multocida*: Capsule and Lipopolysaccharide. In: Aktories K, Orth J, Adler B. *Pasteurella multocida*. Current Topics in Microbiology and Immunology. 361. Springer, Berlin, Heidelberg. ISBN: 9783642310164. https://doi.org/10.1007/82_2012_202

HEY P, Gow P, Torresi J, Testro A. 2012. Cirrhosis, cellulitis and cats: a 'purrfect'combination for life-threatening spontaneous bacterial peritonitis from *Pasteurella multocida*. Case Reports. *British Medical Journal*. 2012:007397. ISSN: 1757-790X. http://dx.doi.org/10.1136/bcr-2012-007397

JAMALI H, Rezagholipour M, Fallah S, Dadrasnia A, Chelliah S, Velappan RD, Ismail S. 2014. Prevalence, characterization and antibiotic resistance of *Pasteurella multocida* isolated from bovine respiratory infection. *The Veterinary Journal*. 202(2):381-383. ISSN: 1090-0233. https://doi.org/10.1016/j.tvjl.2014.07.024

KONEMAN EW, Allen S. 2008. Koneman. Diagnostico Microbiologico/Microbiological diagnosis: Texto Y Atlas En Color/Text and Color Atlas: Ed. médica panamericana. ISBN: 9500608952.
https://books.google.com.ec/books?id=jyVQueKro88C&printsec=frontcover&source=gbs_vpt_reviews#v=onepage&q&f=false

KUBATZKY KF. 2012. *Pasteurella multocida* and immune cells. *Pasteurella multocida*. 53-72. ISBN: 9783642310171. https://link.springer.com/book/10.1007/978-3-642-31017-1

LI N, Feng T, Wang Y, Li P, Yin Y, Zhao Z, He F. 2021. A single point mutation in the hyaC gene affects *Pasteurella multocida* serovar A capsule production and virulence. *Microbial pathogenesis*. 159:105145. ISSN: 0882-4010.
https://doi.org/10.1016/j.micpath.2021.105145

LIN PH, Bush RL, Zhou W, Peden EK, Lumsden AB. 2006. Endovascular treatment of traumatic thoracic aortic injury—should this be the new standard of treatment? *Journal of vascular surgery*. 43(2):22-29. ISSN: 1097-6809.
https://doi.org/10.1016/j.jvs.2005.10.068

MASSACCI FR, Magistrali CF, Cucco L, Curcio L, Bano L, Mangili P, Christensen H. 2018. Characterization of *Pasteurella multocida* involved in rabbit infections. *Veterinary Microbiology*. 213:66-72. ISSN: 1873-2542. https://doi.org/10.1016/j.vetmic.2017.11.023

MATHERN DR, Heeger PS. 2015. Molecules great and small: the complement system. *Clinical Journal of the American Society of Nephrology*. 10(9):1636-1650. ISSN: 1555-905X. https://doi.org/10.2215/cjn.06230614

MORENO-TORRES A, Malvido-Jiménez IR, de la Peña-Moctezuma A, Castillo-Sánchez LO, Fraga TR, Barbosa AS, Sahagún-Ruiz A. 2019. Culture-attenuated pathogenic *Leptospira* lose the ability to survive to complement-mediated-killing due to lower expression of factor H binding proteins. *Microbes and Infection*. 21(8-9):377-385. ISSN: 1769-714X. https://doi.org/10.1016/j.micinf.2019.03.001

NICHOLSON LB. 2016. The immune system. *Essays in biochemistry*. 60(3):275-301. ISSN: 1744-1358. https://doi.org/10.1042/ebc20160017

PALÓCZ O, Gál J, Clayton P, Dinya Z, Somogyi Z, Juhász C, Csikó G. 2014. Alternative treatment of serious and mild *Pasteurella multocida* infection in New Zealand White rabbits. *BMC Veterinary Research.* 10(1):1-7. ISSN: 1746-6148. https://doi.org/10.1186/s12917-014-0276-6

PANNA SN, Nazir KNH, Rahman MB, Ahamed S, Saroare MG, Chakma S, Majumder UH. 2015. Isolation and molecular detection of Pasteurella multocida Type A from naturally infected chickens, and their histopathological evaluation in artificially infected chickens in Bangladesh. *Journal of Advanced Veterinary and Animal Research*. 2(3):338-345. ISSN: 2311-7710. http://dx.doi.org/10.5455/javar.2015.b104

PER H, Kumandaş S, Gümüş H, Öztürk MK, Çoşkun A. 2010. Meningitis and subgaleal, subdural, epidural empyema due to *Pasteurella multocida*. *The Journal of emergency medicine*. 39(1):35-38. ISSN:0736-4679. https://doi.org/10.1016/j.jemermed.2008.04.008

QUESADA SP, Paschoal JAR, Reyes FGR. 2013. Considerations on the aquaculture development and on the use of veterinary drugs: special issue for fluoroquinolones-a review. *Journal of Food Science*. 78(9):R1321-R1333. ISSN: 1750-3841. https://doi.org/10.1111/1750-3841.12222

REGISTER KB, Brockmeier SL. 2019. Pasteurellosis. In: Zimmerman JJ, Karriker LA, Ramirez A, Schwartz KJ, Stevenson GW, Zhang J. Diseases of swine 884-897. John Wiley and Sons. Hoboken. ISBN: 9781119350927. https://doi.org/10.1002/9781119350927.ch57

RICKLIN D, Hajishengallis G, Yang K, Lambris JD. 2010. Complement: a key system for immune surveillance and homeostasis. *Nature immunology*. 11(9):785-797. ISSN: 1529-2916. https://doi.org/10.1038/ni.1923

ROSELL J, de La Fuente L. 2016. Causes of mortality in breeding rabbits. *Preventive Veterinary Medicine*. 127:56-63. ISSN:1873-1716. https://doi.org/10.1016/j.prevetmed.2016.03.014

SAHAGUN-RUIZ A, Granados Martínez AP, Breda LC, Fraga TR, Castiblanco Valencia MM, Barbosa AS, Isaac L. 2014. *Pasteurella pneumotropica* evades the human complement system by acquisition of the complement regulators factor H and C4BP. *PloS One*. 9(10):e111194. ISSN: 1932-6203. https://doi.org/10.1371/journal.pone.0111194

SORIANO-VARGAS E, Vega-Sánchez V, Zamora-Espinosa JL, Acosta-Dibarrat J, Aguilar-Romero F, Negrete-Abascal E. 2012. Identification of *Pasteurella multocida* capsular types isolated from rabbits and other domestic animals in Mexico with respiratory diseases. *Tropical Animal Health Production*. 44(5):935-937. ISSN:1573-7438. https://doi.org/10.1007/s11250-011-9995-x

SOUZA MJ. 2009. Bacterial and Parasitic Zoonoses of Exotic Pets. *Veterinary Clinics of North America: Exotic Animal Practice.* 12(3):401-415. ISSN: 1558-4232. https://doi.org/10.1016/j.cvex.2009.06.003

World Organization for Animal Healt (WOAH). 2012. Manual of diagnostic tests and vaccines for terrestrial animals. *Infection Bursal Disease*. 12:549-565. ISBN: 9290446226. https://doi.org/10.1017/S0031182005007699

WILSON BA, Ho M. 2013. *Pasteurella multocida*: from zoonosis to cellular microbiology. *Clinical microbiology reviews*. 26(3):631-655. ISSN: 1098-6618. https://doi.org/10.1128/cmr.00024-13

ZHU W, Fan Z, Qiu R, Chen L, Wei H, Hu B, Wang F. 2020. Characterization of *Pasteurella multocida* isolates from rabbits in China. *Veterinary Microbiology*. 244:108649. ISSN: 1873-2542. https://doi.org/10.1016/j.vetmic.2020.108649

Errata Erratum

https://abanicoacademico.mx/revistasabanico-version-nueva/index.php/abanico-veterinario/errata