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Revisión bibliográfica sobre la fisiopatología de la Peste Porcina Africana (PPA)

Literature review on the pathophysiology of African Swine Fever (ASF)

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Palabras claves:
PPA, enfermedad, fisiopatología, clínicos, mortalidad, animales, continente.

Resumen

Introducción. La Peste Porcina Africana que afecta a suidos salvajes y domésticos, es una enfermedad muy contagiosa y letal que causa grandes pérdidas económicas. Alcanza un 100% de mortalidad y morbilidad porque se sacrifican de forma obligatoria los animales infectados y sospechosos. Se debe declarar obligatoriamente ante la Organización Mundial de Sanidad Animal (OMSA – OIE). **Objetivo.** Esta revisión pretende contribuir con datos relevantes sobre avances científicos que describen la fisiopatología de la Peste Porcina Africana (PPA). **Metodología.** Se realizó una investigación documental, crítica y descriptiva la misma que permitió obtener información de la fisiopatología de la Peste Porcina Africana (PPA) utilizando bases de datos científicos. Se utilizaron 39 artículos escritos en español e inglés desde el año 2018 hasta el año 2023. **Discusión.** El virus se originó en África, se encuentra en dicho continente y en otros como Europa, Asia, Oceanía y América, afectando a 50 países en todo el mundo. Morfológica y bioquímicamente el virus es complejo y puede resistir a distintas condiciones ambientales, se inactiva mediante calor, pero resiste temperaturas bajas. Los cuadros clínicos que se presentan en la PPA son hiperagudo, agudo, subagudo, crónico y asintomático. Los cuadros hiperagudos producen la muerte súbita, mientras que, en los moderados o asintomáticos, los animales superan la infección y el virus perdura convirtiéndolos en portadores aparentemente sanos, pero persistentemente infectados. **Conclusión.** La peste porcina africana (PPA) es una amenaza mundial con gran impacto sobre la industria porcina. Su compleja interacción con el sistema inmune, la rapidez con la que se propaga, la falta de un tratamiento eficaz y la inexistencia de una vacuna comercial convierten a esta enfermedad en un desafío enorme para el sector porcino mundial. **Área de estudio general:** Medicina Veterinaria. **Área de estudio específica:** Fisiopatología. **Tipo de estudio:** Artículo de Revisión Bibliográfica.

Keywords:
ASF, disease, physiopathology,

Abstract

Introduction. African Swine Fever that affects wild and domestic swine. It is a very contagious disease, and lethal that

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Clinical, Mortality, Animals, Continent.

causes great economic loss. It reaches 100% mortality and death, because they are obliged to sacrifice infected animals and animals suspected of having the disease. It is mandatory to report to the World Organization for Animal Health (WOAH). objective. This review wishes to contribute with relevant facts about scientific advances that describe the Physiopathology of African Swine Fever (ASF). Methodology. A documentary investigation was carried out, both critical and descriptive, which permitted obtaining information about the physiopathology of the African Swine Fever. (ASF). Utilizing as its basis scientific facts. 39 written articles were used in Spanish and English from the year 2018 to 2023. Discussion. The virus originated in Africa and was found on that continent and others such as Europe, Asia, Ocean, and America affecting 50 counties around the world. Morphologically and biochemically the virus is complex and can resist distinct environmental conditions that inactivate with heat but resist low temperatures. The clinical conditions that are displayed in (ASF) are Hyper-severe, severe, sub-severe chronic and asymptomatic. The Hyper-severe types produce sudden death, and when moderate or asymptomatic, the animal survives the infection, and the virus continues converting them into healthy carriers. Conclusion. African Swine Fever (ASF) is a worldwide threat to the swine industry. Its complex interaction with the immune system, the quickness with which it expands, the lack of an effective treatment, and the inexistence of a commercial vaccine makes this disease a huge challenge for the worldwide swine sector. General area of study: Veterinarian medicine. Specific Area of Study: Physiopathology. Type of Study: Article of Bibliographic Revision.

Introduction

African Swine Fever (ASF) or African Swine Fever (ASFV) is classified as an infectious-contagious disease, caused by a DNA-type arbovirus, which attacks wild and domestic swine. The virusPPA is not zoonotic and does not represent a danger to public health.In addition, you cantransmitted and replicated in ticks of the species (Ornithodoros spp.)(1–3).

The “East African Swine Fever” virus was discovered in Kenya by R. Eustace Montgomery in 1921 thanks to an outbreak transferred from wild suids to domestic suids.(4)This virus, later identified as the ASF virus, originated in southeastern Africa, and is found on that continent and in other continents such as Europe, Asia, Oceania and America. Currently, 50 countries have reported its presence in the World Animal Health Information System (WAHIS) of the World Organization for Animal Health (WHO - OIE), as it is mandatory to report it to this organization. In this sense, China considers the virus as a major threat and classifies it as the cause of a Class I epidemic disease of animal origin.(5–7).

The ASF virus has a long chain and its genome is variable, it can survive in environments with the presence of organic matter or in products of animal origin, this being an important factor in its dissemination. Morphologically and biochemically this virus is complex and can resist different environmental conditions, it is inactivated by heat, but it resists low temperatures.(8).The clinical pictures that occur in ASF are hyperacute, acute, subacute, chronic and asymptomatic. The clinical signs of hyperacute symptoms are lethargy and sudden death, but if the swine manage to survive a few more days, they may present diarrhea, hematemesis and skin lacerations. On the contrary, in moderate or asymptomatic cases, the animals overcome the infection and the virus persists, turning them into apparently healthy carriers, but persistently infected.(9).

Within the swine industry, ASF causes great economic losses, since it is a lethal disease that reaches 100% mortality and morbidity, causing the mandatory slaughter of infected and suspected animals.(3).Since 2019, it has spread rapidly from Vietnam to different countries and continents, directly affecting producers and consumers, putting food security at risk, since 37% of the world's population eats pork; this source of animal protein is consumed more frequently due to its easy reproduction and rapid growth, in addition to its high feed conversion rate (10,11).

The lack of complete and up-to-date information on the characteristics of the virus and on the pathological mechanisms, effects on organ systems and the host immune response to the disease has contributed to its rapid spread and entry into farms around the world. This virus is a threat to pig farmers in developing countries, mainly in small and backyard farms which have limited biosecurity measures.(10)This problem is not foreign to our continent, as the virus was already detected in 2021 in pig farms in the Dominican Republic and Haiti. The ASF virus is very complex, so there is currently no treatment. There are only experimental and non-commercial vaccines to immunize pigs.(11–13).

This bibliographic review aims to contribute with relevant data on scientific advances in the pathophysiology of African Swine Fever (ASF). It is hoped that this document will allow health authorities, veterinarians, pig producers and citizens to Obtain a knowledge

base that will help implement biosecurity measures and actions that are effective against the spread of ASF and minimize economic losses.

Methodology

In the present work, a documentary, critical and descriptive research was carried out which allowed obtaining information on the pathophysiology of African Swine Fever (ASF). The sources of scientific information used for the research were: Google Scholar, Redalyc, PubMed, Dialnet and SciELO.

The number of articles identified was 350, written in Spanish and English. In addition, only those articles published in the last five years, from 2018 to 2023, were considered. The keywords used to select the documents in Spanish and English were: "African Swine Fever", "ASF", "physiopathology", "virus", "porcines", "immunity", "pathogeny", "African swine fever", "ASF", "pigs", "pathophysiology", "immunity", "pathogenesis".

Of these 350 articles, 300 were discarded after reading the abstract, because they did not directly match the description of the pathophysiology of the disease or because they included very little information related to this topic. 50 articles were read completely, after this reading, 11 more articles were discarded because they presented information similar to that of other documents, but used very old bibliographic references. The final number of articles selected was thirty-nine, with which the present bibliographic review was prepared.

Discussion

Etiology

African swine fever is caused by an arbovirus of the alphavirus genus, belonging to the asfaviridae family, which infects only pigs.(14). This is large in size, as its viral particle measures approximately 200 nm. The virus is a DNA type with a double-stranded or two-strand linear genome, with a size of 170 - 193 kbp, containing between 151 and 167 ORFs that vary according to the viral strain. It groups more than 150 to 200 proteins (structural, replication, transcription, enzymes and virus-host interaction)(1, 2).

The VPPA presents an icosahedral morphology, with five centered layers; the external lipid membrane (envelope or fifth layer) has proteins identified as p12 - CD2v and is acquired in the budding process thanks to the plasma membrane of the cell. The fourth layer or external icosahedral protein capsid, is formed by the proteins pE120R - p49 - p72. The internal lipid membrane is the third layer constituted by the following proteins pE183L - pE199L - pH108R - pE248R - p12 - p22 - p17. The internal icosahedral protein capsid is the second layer and is formed by pS273R - p5- p8 - p14 - p15 - p34 - p35 - p37 - p150. The core shell is the first layer which covers the nucleoid which is formed by the

proteins pA104R - p10 and contains the double-stranded DNA. For this reason, in veterinary medicine the virus is classified biologically and structurally as highly complex.(1, 2, 7, 15).

The ASF virus has a unique serotype, thanks to a partial genomic sequence of the P72 primary capsid protein, however, its genotypes can be different. In this sense, 24 different genotypes were discovered in wild boar refuges in the southern and eastern regions of Africa, in addition to numerous strains with heterogeneous virulence. This variety can occur due to cloning, alteration or loss of one of the genomic sequences.(13,16).

Idiotype I belonging to (Italy) and idiotype II belonging to (Asia and Europe) are the only genotypes found outside the African continent. Genotype II of ASF has the ability to produce subacute and acute forms of the disease with high virulence and mortality, while genotype I has low virulence causing chronic and asymptomatic forms, which makes it difficult to control and prevent the disease.(12, 17, 18).

Incubation Period

Contact between an infected pig with ASF and a healthy one induces viral infection in the pig, which sheds the virus during the incubation phase even if no clinical signs are observed. At the time clinical signs appear, the viral level increases in all types of fluids.(9). After infection, domestic pigs are able to excrete the virus 24 to 48 hours prior to the appearance of clinical signs.(3).The incubation phase varies between 4 to 19 days, this will depend on the transmission routes and virulence, it is also noted that short incubation periods last between four to eight days, while long periods last 15 to 19 days.(4)The incubation period in experimental infections is short, from two to five days, and depends on the route of inoculation and dose of the virus.(19).On the other hand, if the pig was infected by a tick bite, the incubation period is less than five days.(20).Swine can be infected with as few as five viral particles, which is important since so few units of the virus cause clinical disease.(21).

Reports mention that the same viral strain can cause different effects in herds of different ages or species, meaning that the decrease in virulence is probably not related to the host's genetics or viral changes, but could be the result of combinations of elements that are still unknown. In this sense, in recent studies in which different amounts of virus were administered and by different routes, an alteration in pathogenesis has been found.(3).

Reports mention that the same viral strain can cause different effects in herds of different ages or species.

Infected Species

ASF is a disease that threatens the Suidae family. There is a wide variety of African wild pigs such as the African warthog (Phacochoerus africanus), the giant wild boar (Hylochoerus meinertzhageni), the red potamochoerus (Potamochoerus larvatus) and the river potamochoerus (Potamochoerus porcus), which are natural hosts of the virus and are distinguished by not presenting symptoms when infected, they are resistant to the disease. The European wild suids, the European wild boar (Sus scrofa), and the domestic suids (Sus scrofa domesticus) are also hosts and are distinguished by presenting clinical signs when infected with the virus. American pigs (Tayassuidae) are not sensitive to the virus, in this group are the white-lipped peccary (Tayassu albirostris) and the collared peccary (Tayassu tajacu)(11, 16, 17). Soft-shelled, visionless ticks (Ornithodoros spp) are also infected by the ASF virus.(22).

Transmission routes

Secretions and Excretions

The ASF virus is transmitted through direct contact with secretions and excretions of infected domestic or wild pigs, from one to seven days post infection, and is considered a source of high pathogenic risk. The above varies depending on the route of infection, strain, age, immunological and health status of the infected pig.(3, 8, 12).

The viral level in oronasal fluids and blood is high in the clinical stage.In addition, the virus is associated with erythrocytes and travels throughout the body, for this reason, blood is considered one of the main sources of infection, a recently infected pig has a high concentration of virus in its blood 105.3 to 109.3 HAD50 / ml (heme adsorption dose test in blood at 50% per milliliter). The disease can be spread through blood from necropsies, bloody diarrhea and wounds from fights between pigs. On the other hand, theThe lowest viral level is found in excretions from the conjunctiva, genitals and feces, all associated with intermittent excretions of the virus.(23–25)Aerosol transmission affects pigs that are within a short distance, however, if there is separation with walls or partitions, it will prevent the ASF virus from spreading.(3, 13).

Transmission of the virus from infected animals can begin from the second day after infection through saliva, blood, nasal and ocular secretions. After a few days, the virus is eliminated through semen, urine, feces.(25)The virus has the ability to remain alive in different temperature ranges and environmental conditions. In whole blood it remains for 15 weeks and in decomposed blood for 18 months at a temperature of 4 o C, in feces for 11 days at room temperature, in urine for 45 days under appropriate conditions, in nasal sprays for 5 minutes, provided the humidity exceeds 30%.(26)There is still no reliable evidence that the ASF virus can be transmitted through sexual contact or during gestation, that is, from mother to fetus. However, a boar was experimentally infected and it was found that its semen contained ASFV.(23).

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Fomites

The virus remains stable in environmental conditions where the pH ranges between 4 and 10, thus, it is active and can be transmitted indirectly to animals that come into contact with different fomites (non-living objects) that can become contaminated with VPPA such as clothing, boots, different foods, water, feed, transport vehicles, work materials, waste, gloves, needles, surgical equipment, among others.(8, 20, 27).Carcasses of dead pigs are also a source of transmission of the virus in the habitat of wild suids, since these animals can consume infected carcasses.(17).

The virus is latent in meat products derived from infected pigs for long periods of time, i.e. weeks, months, even years. The virus is inactivated by subjecting the meat to a temperature of 70 o C for 30 min.(26)In raw, frozen meat, the virus can survive for up to 1,000 days, while in refrigeration it can survive for 100 days. In the fat and skin of the pig, the virus can survive for 300 days, and in the offal for 105 days. In the meaty bone, the virus can persist for 150 days at a temperature of 4 o C. In cured meat products, the inactivation time varies, depending on the type of curing to which they are subjected, ranging from 30 days (pepperoni and salami) to 400 days (Iberian ham).(11, 25, 28).

Another characteristic that facilitates the transmission of the virus is its resistance to various temperatures, which has been essential to extend the distribution of the disease in different areas with different climates. In addition, pig waste and its products have been transported by airplanes or ships on international routes, spreading the virus to countries previously free of ASF.(11).

Bites

The ASF virus is the only one that is transmitted by the bite of arthropods (Ornithodoros spp), these mites have been recognized as a reservoir and biological vector of the disease.(17). In this case, the infected soft tick (Ornithodoros spp.) has the ability to transmit the virus to pigs through saliva emitted from the hypostome (sucking organ), when they feed on the pig's blood. Soft ticks (Ornithodoros moubata and Ornithodoros porcinus) can infect each other sexually, transovarially and transstadially. Ticks (Ornithodoros erraticus) became infected with the virus transstadially, when in Portugal and Spain, among other countries, the virus became enzootic in the European wild boar population.(22).

Mites (Ornithodoros spp) are commonly found in burrows in various African areas. In the Iberian Peninsula they are found in certain places, while on the island of Sardinia they are absent. Wild suids tend to hide in burrows and surfaces with a lot of humidity, so when an African wild boar is born it can be infected with the ASF virus through the bite of ticks (Ornithodoros moubata) that live in such places.(3, 23).

Ticks of the species (Ornithodoros spp), native to Africa and the Iberian Peninsula, have an average life expectancy of five years and it has been shown that the ASF virus remains latent for months or years inside them, after consuming infected blood. The ASF virus in ticks has the ability to replicate in tissues, and they transmit the virus vertically to their offspring.(17, 23).

On the other hand, black flies or mosquitoes are capable of mechanically transmitting the virus. Flies (Stomoxys calcitrans) present in stables transmit the African swine fever virus 24 to 48 hours after having consumed blood from an infected pig, and the ability to transmit high levels of the virus is maintained for two days.(20, 29).

Pathophysiology

Entry and replication

The ASF virus can enter the pig's body through different routes, enteral and parenteral, among the vehicles of the virus are excretions (urine, feces), secretions (nasal, oral, conjunctiva, semen, blood) and fomites (clothing, boots, various foods, water, feed, transport vehicles, work materials, waste, gloves, parenteral injections, surgical equipment). In addition, it is found in wild boar carcasses, raw meat, other carcass derivatives,European and African soft ticks (Ornithodoros spp) and flies (Stomoxys calcitrans)(17, 29)

After 30 minutes post infection (mpi), ASFV binds to the host cell via membrane receptors that have not yet been identified, possibly CD163, MHC-II and CD45. It enters the cell by macropinocytosis or endocytosis. The difference between these viral entry processes depends on the mediator proteins, which differ until the formation of early endosomes.(17, 29).

In endocytosis, the virus binds to cell receptors and forms a membrane-lined pit. This process depends on the presence of clathrin, a protein that assembles and agglomerates in the invagination of the cell membrane, thanks to AP-2 (adapter protein) that captures and binds clathrin. Another protein, Dab2 (deactivated protein two) is only responsible for binding clathrin and the AP180 and Eps15 proteins that accelerate the process. In addition, the amphiphysin protein is responsible for capturing dynamin simultaneously, which will be necessary later. The cell membrane-lined pit is invaginated by actin, dynamin, and endophilin, after which the clathrin-containing cell membrane captures the viral particles and cell receptors. The dynamins fuse to the neck and cut the corpuscles, thus separating the cell membrane. The detached tiny vesicles lose clathrin and early endosomes are formed. (7, 30).

In macropinocytosis, the virus incites and activates Ras – RKT which will then activate molecules such as Rab 5 - Rac 1 – PI3K 1 – Arf6, activating actin, which reorganizes and,

accompanied by microfilaments, forms blisters on the surface of the membrane. The Arf6 – PAK 1 proteins regulate changes in the cytoskeleton and cause the cell membrane to flex to envelop viral particles, then myosin accompanied by Rababkyrin - Rab 5 – CtBP1 and PAK1 close the blisters, causing the vesicles to form the early, middle and late endosomes in the cell. The formation of macropinosomes or early endosomes shows that the virus has successfully completed the internalization phase.(1.7).

The virion enters early endosomes 1 to 30 minutes post infection (mpi) and is directed to late endosomes 30 to 90 mpi, where the SFV is cleaved and the genome is released. Early gene expression occurs 4 to 6 hours post infection (hpi) and encodes proteins that are associated with virus replication, thus causing multiplication of the ASF viral genome in the nucleus and then in the cytoplasm (6 to 8 hpi). Later, intermediate and late gene expression encodes proteins associated with the structure of viral particles, this begins to manifest between 8 to 16 hpi; viral particles are assembled with all proteins between 16 to 24 hpi. Around 24 hpi, viral particles that were assembled in a viral factory leave the cytoplasmic membrane through the budding process towards the interstitial fluid.(1,7,11).

Spread of the disease

The ASF virus can enter the pig's body via any route, however, the most common route of transmission is the oronasal route; by this means, the ASFV enters the dorsal pharyngeal mucosa or the tonsils and from there to the submandibular and retropharyngeal lymph nodes, where primary replication occurs in the main host cells, which are monocytes and macrophages. After this replication, the virus is distributed via the bloodstream (viremia), bound to erythrocytes and leukocytes, reaching all the tissues of the pig (systemic distribution).(11, 31, 32).

The virus appears two days after inoculation in the bone marrow, liver, kidney, lung and spleen. It multiplies rapidly in these organs, which is why they are classified as secondary centers of viral replication, causing hemorrhagic lesions, necrosis and damage to the vascular endothelium.(11, 32)However, it is not clear why the ASF virus induces generalized hemorrhage and subsequent hypovolemic shock in hyperacute, acute, and subacute clinical pictures of the disease.(33).

Infected macrophages and monocytes produce an elevated amount of cytokines (inflammatory mediators), which in excess cause the death of T and B lymphocytes. The above may explain why lymphopenia occurs during PPA infection. Cytokines also damage the vascular endothelium and change vessel permeability, causing hemorrhages and edema, as well as vasodilation and obstructive intravascular coagulation (thrombi).

Necrosis, degeneration of lymphatic vessels and edema are observed in lymphatic tissue. (9,32).

Immune response

The immune response of animals has three defense mechanisms against any type of infection, whether viral or bacterial. The first line is the physical barrier, which is classified as the most effective. It includes healthy skin, as well as mucous membranes (conjunctiva, respiratory, gastrointestinal and genital) that are self-cleaning through sneezing, coughing, vomiting, mucus, diarrhea, urination, among others. The epithelium of the mucous membranes is covered by mucus, which allows it to trap microorganisms, thus defending the host's body. In addition, the fluids that cover the mucous membranes can be media that facilitate the transmission of viral particles.(13).

The second line is the innate response where dendritic cells, macrophages and mast cells induce an inflammatory response by distinguishing pathogen-associated molecular patterns (PAMPs) through cellular pattern recognition receptors (PRRs). The inflammatory response causes different cells, primarily macrophages and neutrophils, to be activated and migrate from the blood to the site of invasion. PRRs recognize the ASF virus through the cGAS-STING pathway, the same ones that distinguish viral DNA and infected cells in the cytoplasm. Plasmacytoid dendritic cells (pDC) produce type I IFN, which acts on infected cells by inhibiting the development of viruses.(34).

The third line is the adaptive immune response in which macrophages and dendritic cells process and capture antigens efficiently, protecting the individual in the long term. Dendritic cells are antigen presenting cells that activate naïve T cells. When processing antigens, they become small polypeptides that bind to the major histocompatibility complex (MHC) molecules of the expert antigen receptor.(35).Dendritic cells use the exogenous antigen pathway to process the ASF virus. This begins by phagocytosing the antigen to produce endosomes, which are acidified during intracellular movement and maturation, and then bind to lysosomes that have proteases. The ingested peptides are degraded into pieces of different sizes thanks to proteases. In addition, some of these peptides bind to MHC-II molecules, thus being displayed to CD4+. After acidification of the endosome, the virus separates from its capsid, binds its internal capsule to the endosomal membrane and releases its central capsid and DNA.(13,36).

Exogenous molecules (antigens) are those that induce acquired immunity, through multiple signals the B cells are activated. The binding of the antigen to the BCR is necessary to cause the response of these cells, but sometimes it is not sufficient to form antibodies. For their activation, B cells require the stimulation of CD4+ and the presence of cytokines.(13).

Antigens are divided into small polypeptides and placed in the major histocompatibility complement binding groove (MCH-I). The antigen is then located on the cell surface, binds to the antigen receptor on T cells, and completes presentation. CD8+ lymphocytes then become cytotoxic and become fully activated, leaving the lymphatic organs and searching for infected cells on their own.(37).

Once activated, CD8+ cells can recognize the MHC-antigen complex expressed on other cells and cause the death of target cells contaminated with the ASF virus. Natural killer (NK) cells fight the infection by killing other infected cells, inciting the antibody-mediated cellular cytotoxicity (ADCC) mechanism, or releasing proinflammatory cytokines such as tumor necrosis factor alpha (TNF-α) and interferon (IFN).(30, 37).

In order to survive and replicate, the ASF virus has the ability to encode proteins associated with the host cell and thus evade immune responses. Eight days after infection, antibodies increase, indicating that the host has humoral immunity. However, this level of antibodies is not sufficient to neutralize all viral particles and it is estimated that between four and 13% of viruses escape neutralization. This is attributed to the nature of the virus and not to the lack of immune response from the hosts, since helper T lymphocytes (CD4+) and cytotoxic T lymphocytes (CD8+) are capable of protecting the pig against the virus, but not completely.(7, 13).

If the pig resisted the disease for a long time, it causes a high immune reaction with the presence of circulating molecules. These are important to protect the pigs in the event of a new infection with the same virus. It should be noted that these antibodies will not provide protection against other viral strains.(3). The lack of knowledge about the relationship between the ASF virus and the immune system of swine has caused a delay in the development of vaccines.(13).

Clinical signs and lesions

ASF presents clinically in very varied forms. Five clinical pictures have been described: peracute, acute, subacute, chronic and asymptomatic. The above depends on the level of virulence, route of transmission, the particularities of the host, the amount of virus introduced and some other factors that are still unknown.. Mortality in pigs depends greatly on the virulence of each strain. Three levels have been described: high, moderate and low, related to different presentations of the disease. If the virulence is high, almost 100% lethality occurs, while if it is low, death is < 30%.(3, 16, 18).

In the hyperacute form, swine may die between the first and fourth day post-infection, without showing symptoms or visible lesions in internal organs, only mild cutaneous hyperemia. In the acute form, swine show fever, cyanosis in the snout, ears, limbs, ventral thorax, abdomen and tail, skin with diffuse or focal erythema, prostration, anorexia,

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dyspnea, ataxia, conjunctivitis and sometimes hemorrhagic or watery diarrhea. Pigs die between four and 21 days dpi, with a mortality rate of 90 to 100%. It should be noted that this clinical symptomatology is the most common in pigs infected with the European viral strain (genotype II); In the subacute form, pigs die within a few weeks, from day 20 post infection with clinical symptoms similar to those of the acute disease but less severe. The ASF lesions in pigs that do not resist the disease are more marked (edema and vascular changes), and it also produces mortality that varies between 60 and 90%.(9, 19).

In places where ASFV is endemic, chronic and asymptomatic forms are frequently observed, which are caused by isolates of moderate or low virulence. The two forms mentioned above are characterized by a low mortality rate, nonspecific clinical signs, which make it difficult to recognize ASFV disease, and other lesions in pigs that are attributed to secondary infections.(19)There are cases where 2 to 10% of pigs acutely infected with ASF have recovered, these animals develop a persistent infection in all tissues, for this reason, for greater safety of the farm, the surviving animal is sacrificed and the facilities are disinfected.(31).

Table 1. Clinical forms of PPA

	Hyperacute	Acute	Subacute	Chronicle	Asymptomatic
Virulence	High	High	Average	Low	Low
Death	100 %	90 – 100%	60 – 90 %	20 – 60 %	2 - 20 %
Post incubation infection	Imperceptible	7 dpi	6 to 15 dpi	Weeks or months	Imperceptible
Pyrexia	> 42 °C	> 40 °C, < 42 °C	Less than or equal to 40 °C	Very slight or imperceptible	Imperceptible or non-existent
Thrombocytopenia	-	Absent or mild	Transient	-	-
Signs	Imperceptible	Diarrhea, vomiting, seizures and bleeding	Diarrhea, vomiting, seizures and bleeding	Vomiting, cough, diarrhea and conjunctivitis	Diarrhea, conjunctivitis, cough and arthritis
Fur	Erythema	Erythema, petechiae	Erythema, petechiae and cyanosis	Necrotic areas, hyperpigmentation, bruising	Necrotic areas, hyperpigmentation, bruising.
Reproductive disorder	-	-	Abortion	Abortion	Abortion
Behavior	Immediate death < 4 to 6 dpi*	Loss of appetite, apathy, drowsiness, overcrowding, death from 6 to 9 dpi*	Loss of appetite, confusion, lethargy and death 6 to 9 dpi*	Normal or lethargic, death months after contracting the infection.	Normal or death months after contracting the infection.
Pathological findings	Imperceptible	Hyperemic splenomegaly, petechial hemorrhages	Focal infarction or hyperemic splenomegaly	Pneumonia, pleurisy, fibrinous pericarditis,	Pulmonary consolidation, pleuritis, fibrinous pericarditis.

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	in the gallbladder and renal cortex, marbled infarcted lymph nodes (renal and gastrohepatic) , lung with alveolar edema, hemorrhage in the endocardium and epicardium.	, blood clot-like nodes, petechial hemorrhages in the kidney (cortex, pelvis, medulla) and perirenal edema, hemorrhage in the endocardium and epicardium, edematous gallbladder wall.	sometimes hemorrhages, swollen lymph nodes, enlarged spleen with normal coloration, tonsils with necrotic foci.

Fountain: Carrillo 2020 (3) and Li et al. 2022 (31) *dpi = days post infection

Differential diagnosis

African swine fever sometimes does not present with all the lesions and signs observed in Table 1, therefore, it must be differentiated from other hemorrhagic pathologies. It should be noted that other bacterial or viral diseases may also present with similar lesions and clinical signs. The main disease that can be differentially diagnosed is classical swine fever (CSF). In addition, there are other diseases such as porcine reproductive and respiratory syndrome (PRRS), porcine dermatitis and nephropathy syndrome, acute salmonellosis, porcine erysipelas and Aujeszky's disease that constitute other differential diagnoses. No lesion or clinical sign present in pigs will be the reason for establishing a definitive diagnosis of ASF; only through laboratory analysis can the definitive diagnosis be determined.(11, 25).

Laboratory tests

When ASF is suspected on the farm, it is necessary to perform clinical examinations and define the result with laboratory analysis. To perform ASF tests on dead animals, it is recommended to collect samples from the lymph nodes, spleen, kidney and lung, while if the pig is alive, serum and blood are collected. In addition, in wild boar, the disease has been diagnosed through bone marrow, since it is difficult to find preserved samples that contain ASF in other tissues.(38)Currently, there are various methods for diagnosing ASF disease, including molecular diagnosis, which is responsible for detecting viral DNA, virological diagnosis, which is responsible for detecting viral proteins or the virus, and serological diagnosis, which is responsible for detecting antibodies.(25).

The methods used to detect the virus are serological, which includes the heme adsorption test, polymerase chain reaction (PCR), direct immunofluorescence (DIF) and direct

enzyme-linked immunosorbent assay (ELISA). Virological methods include indirect immunofluorescence (IFI), immunoblotting and indirect ELISA. It should be noted that the most commonly used tests to detect ASF are PCR and indirect ELISA. These have different presentations, whether private or commercial. It is also advisable to perform several diagnostic techniques simultaneously, i.e. antibody and virus detection.(9).

As ASF is an exotic disease, diagnosis begins by detecting the viral agent by PCR (whole blood). In addition, ELISA (serum) tests will be used in the sampling that is part of the monitoring of animals that were in contact with other suspected or confirmed pigs, being an option to support disease surveillance.(8)It is important that the diagnosis of the disease is fast and effective, this helps to limit its spread, in addition to applying mitigation measures in the shortest possible time, the two aforementioned factors are essential to prevent the ASF disease from evolving, since there is still no treatment, much less a commercial vaccine to combat the virus in swine.(12, 25).

Mitigation and control

PPA is a disease that must be declared to national and international control bodies.(20). All countries must have a contingency plan to control the disease. It must be clear and contain information to act appropriately from the moment the disease is suspected until the end of the outbreak. Each country has its own contingency plan against ASF based on standards and recommendations provided by the WHO, which states that it must be adapted to the health, epidemiological, productive and infrastructure situations of each country.(25).

Various stages are used in countries to control and prevent the disease in accordance with the WHO recommendation. Each stage is made up of different activities, including the rapid activation of the early warning system through active and passive surveillance. To prevent the disease, pigs are collected, controlled and safely disposed of, hunting and feeding wild suids is prohibited in the surroundings where infected populations of this species have been identified, tourist movements and the handling of wild suids are controlled, which is accompanied by talks and information campaigns on ASF in the affected areas. In addition, surveillance is carried out through tests performed on sick or deceased animals, this is the most effective method to detect ASF early, mainly in cases where there is low mortality, this occurs when the virus enters a farm for the first time. (12).

Within the farm, good husbandry practices focused on maintaining biosecurity are the most effective strategies to prevent the entry of the ASF virus into pig facilities. Research indicates that the lack of biosecurity and the inability to detect the virus early are the most important problems that allow the introduction and rapid spread of the virus. In countries with an ASF outbreak, sanitary control measures are implemented such as early

identification of the virus and culling of infected and suspected pigs, asepsis of the farm, control of affected areas and preventing the movement of pigs. It should be noted that there is still no vaccine, for this reason it is of vital importance to carry out sanitary inspection and maintain optimal surveillance.(9, 12).

In countries that have had outbreaks of the disease, a sanitary cordon of approximately ten kilometers is established from the point where the outbreak occurred. There is strict control over vehicles transporting dead or live animals, as well as animal feed or by-products. Within 10 km, wild boars and suids are captured, blood samples are taken for viral identification and early detection in order to prevent the spread through the identification and elimination of affected animals. In some cases, it is recommended to build fences, in this way the separation of sites free from those at risk of ASF is ensured.(3).

Mitigation is achieved by preventing contact between pigs from different herds; prohibiting the entry and exit of pigs from the farm and if the movement of animals is strictly necessary, pigs arriving at a new farm must undergo a mandatory quarantine; do not reintroduce pigs that were transported to fairs into the herds; in the event that the total sale does not take place, a prior quarantine is necessary for re-entry; carcasses are eliminated in compliance with biosecurity protocols; laboratory tests must be performed before incinerating or burying them; these allow decisions to be made about the herd; natural mating with external boars is considered a dangerous practice since the boars are moved to different herds; do not feed pigs with waste and fodder collected in risk areas; implement facilities that guarantee biosecurity; hunt wild suids to control and eliminate ASF since these animals are a source of infection and can be healthy carriers.(23, 39).

Mitigation measures applied in Ecuador

In Ecuador, African swine fever (ASF) is an exotic disease. In this sense, the Phytosanitary and Zoosanitary Regulation and Control Agency (AGROCALIDAD), based on the functions granted to it by the Organic Law on Agricultural Health, develops and issues resolution 0187 of August 8, 2019, which establishes a contingency plan for the disease. This plan aims to offer tools to confront the virus and prevent its spread, and measures were also established to prevent the virus from entering the country. Mitigation measures include strengthening border inspections, limited authorization for the entry of products that are considered risky or dangerous for the national swine industry, streamlining risk management tables, and using good practices and biosecurity on farms.(8).

Ecuador borders Colombia, Peru and the Pacific Ocean. Colombia is free of ASF, however, two surveillance and quarantine points have been set up, one in Rumichaca and another in San Miguel in the provinces of Carchi and Sucumbíos respectively. Peru also

has no cases of ASF, here four quarantine points have been set up: one at the Huaquillas international bridge, another in Chacras located in the province of Oro, another Chacras located in Macará province of Loja, and Zapotillo located in the province of Loja. In our country, ASF can also enter through livestock merchandise or people who are contaminated, these entry routes can be land, air or sea. Ecuador maintains 16 airports, which are divided into international and national airports. The international airports are the highest risk and are located in the provinces of Pichincha, Cotopaxi, Esmeraldas, Guayas, and Manabí. Each of these has equipment and personnel in charge of zoosanitary control.(8).

Conclusions

- African swine fever (ASF) is classified worldwide as a threat that significantly affects the pig industry. The virus has an indisputable spread; currently, the existence of ASF has been reported in 50 countries belonging to the five continents. Its complex interaction with the immune system due to its particular viral structure, the speed with which it spreads, the lack of an effective treatment and the nonexistence of a commercial vaccine make the disease become an immense challenge for the sector. Thoroughly understanding the transmission mechanisms, the pathophysiology and the preventive measures of the disease is essential to efficiently fight the ASF virus. The biosecurity and surveillance measures that are implemented must be rigorous, in addition, the help of national and international entities is key to minimize the risks and preserve the health of domestic and wild swine worldwide.

Conflict of interest

The authors declare that there is no conflict of interest.

Authors' contribution statement

Erika Evangelina Altamirano Martínez. Information search and article writing.

MVZ M. Sc. Ana Rafaela Burgos Mayorga. Project conception and design, document writing and revision.

Bibliographic References

1. Andrés G, Charro D, Matamoros T, Dillard RS, Abrescia NGA.The cryo-EM structure of African swine fever virus unravels a unique architecture comprising two icosahedral protein capsids and two lipoprotein membranes. Journal of Biological Chemistry [Internet]. January 3, 2020 [cited October 1, 2023]; 295(1):1-12. Available at:

https://www.sciencedirect.com/science/article/pii/S0021925817495457?via%3Dihub

2. García E. Study of macropinocytosis as an endocytic mechanism of entry of the African swine fever virus [Internet]. [Madrid]: Autonomous University of Madrid; 2013 [cited 11 October 2023]. Available at: https://repositorio.uam.es/bitstream/handle/10486/660467/garcia_sanchez_elena.pdf?sequence=1&isAllowed=y

3. Carrillo C. African swine fever epidemic: current status. Veterinaria México OA [Internet]. 2020 [cited July 11, 2023];01-21. Available at: https://www.medigraphic.com/pdfs/vetmex/vm-2020/vm203d.pdf

4. Eustace Montgomery R. On A Form of Swine Fever Occurring in British East Africa (Kenya Colony). Journal of Comparative Pathology and Therapeutics [Internet]. January 1921 [cited 4 Oct 2023]; 34:159-91. Available at: https://www.sciencedirect.com/science/article/pii/S0368174221800314

5. World Health Organization [WHO]. African swine fever (ASF) situation reports. WHO [Internet]. 2023 [cited 11 October 2023]; Available at: https://www.woah.org/en/disease/african-swine-fever/#ui-id-2

6. World Health Organization [WHO]. African swine fever. 2023 [cited 11 October 2023]. Available at: https://www.woah.org/en/disease/african-swine-fever/#ui-id-1

7. Wang Y, Kang W, Yang W, Zhang J, Li D, Zheng H. Structure of African swine fever virus and associated molecular mechanisms underlying infection and immunosuppression: a review. Front Immunol [Internet]. September 6, 2021 [cited July 12, 2023]; 12:1-17. Available at: https://www.frontiersin.org/articles/10.3389/fimmu.2021.715582/full

8. Agrocalidad. Contingency plan for African swine fever in Ecuador [Internet]. 2019 [cited July 11, 2023]. p. 4-79. Available at: https://www.agrocalidad.gob.ec/wp-content/uploads/2020/05/pla5.pdf

9. Martínez J, Accensi F. African swine fever, an epidemic that is sweeping through Europe. Albéitar 222 [Internet]. 2019 [cited July 7, 2023]; Available at: https://core.ac.uk/download/pdf/189882584.pdf

10. Izzati UZ, Inanaga M, Hoa NT, Nueangphuet P, Myint O, Truong QL, et al.Pathological investigation and viral antigen distribution of emerging African swine fever in Vietnam. Transbound Emerg Dis [Internet]. July 1, 2021 [cited

July 26, 2023]; 68(4): 2039-50. Available at: https://onlinelibrary.wiley.com/doi/10.1111/tbed.13851

11. Elarre I, Rodríguez E. African Swine Fever, a real threat that is increasingly closer. Navara Agraria Journal [Internet]. 2023 [cited July 7, 2023];15-9. Available at: file:///C:/Users/USUARIO/Downloads/254-Peste-Porcina.pdf

12. Plavsic B, Rozstalnyy A, Park JY, Guberti V, Depner K, Torres G. Strategic challenges for global control of African swine fever. OIE [Internet]. 2019 [cited 26 July 2023];1-14. Available at: https://web.oie.int/downld/SG/2019/E_87SG_10.pdf

13. Wang Z, Ai Q, Huang S, Ou Y, Gao Y, Tong T, et al. Immune escape mechanism and vaccine research progress of African swine fever virus. vaccines [Internet].March 1, 2022 [cited July 11, 2023];10(3). Available at: file:///C:/Users/USUARIO/Downloads/vaccines-10-00344-v2%20(1).pdf

14. Dixon LK, Chapman DAG, Netherton CL, Upton C. African swine fever virus replication and genomics [Internet].Vol. 173, Virus Research. 2013 [cited 4 October 2023]. p. 3–14. Available at: https://www.sciencedirect.com/science/article/pii/S0168170212004091?via%3Dihub

15. Blome S, Franzke K, Beer M. African swine fever – A review of current knowledge [Internet].Vol. 287, Virus Research. Elsevier BV; 2020 [cited 11 July 2023]. Available at: https://www.sciencedirect.com/science/article/pii/S0168170220304019?fr=RR-2&ref=pdf_download&rr=7ed65015ca98953d

16. Dixon LK, Stahl K, Jori F, Vial L, Pfeiffer DU. African swine fever epidemiology and control.Annu Rev Anim Biosci [Internet]. 2020; 8:221-67. Available at: https://doi.org/10.1146/annurev-animal-021419-

17. Calcina J, Rivera H. Main epidemiological aspects of African swine fever [Internet]. [Lima]: Universidad Nacional Mayor de San Marcos; 2022 [cited July 11, 2023]. Available at: https://cybertesis.unmsm.edu.pe/bitstream/handle/20.500.12672/18734/Calcina_ij.pdf?sequence=1&isAllowed=y

18. Urbano A, Ferreira F. African swine fever control and prevention: an update on vaccine development. Vol. 11, Emerging Microbes, and Infections. Taylor and Francis Ltd.; 2022. p. 2021-33.

19. Arias M, Gallardo C, Delicado V, Torre A. African swine fever: an overview of the current challenge. ResearchGate [Internet]. 2017; 30-5. Available at: https://www.researchgate.net/publication/291898770

20. Iowa State University. African Swine Fever. 2010 [cited 11 July 2023]. Available at: https://www.cfsph.iastate.edu/Factsheets/en/peste_porcina_africana.pdf

21. Wozniakowski G. Small doses of viruses can cause African swine fever. 3tres3 LATAM [Internet]. 2020 [cited 5 November 2023]; Available at: https://www.3tres3.com/latam/articulos/dosis-pequenas-de-virus-pueden-causar-peste-porcina-africana_12466/

22. Boshoff CI, Bastos ADS, Gerber LJ, Vosloo W. Genetic characterization of African swine fever viruses from outbreaks in southern Africa (1973-1999).Vet Microbiol [Internet]. 2007 Mar 31 [cited 2023 Jul 26];121(1-2):45-55. Available from: https://www.sciencedirect.com/science/article/abs/pii/S0378113506004627

23. Bellini S, Rutili D, Guberti V. Preventive measures aimed at minimizing the risk of African swine fever virus spread in pig farming systems.Acta Veterinaria Scandinavica [Internet]. 29 November 2016 [cited 25 October 2023];58(1). Available at: https://actavetscand.biomedcentral.com/articles/10.1186/s13028-016-0264-x

24. International Regional Organization for Agricultural Health. Risk analysis on the probability of entry, establishment and dissemination of the African swine fever virus in pig farming in the countries of the OIRSA region. OIRSA [Internet]. June 2020 [cited October 29, 2023]; Available at: https://www.oirsa.org/contenido/2020/AR_PPA_Edici%C3%B3n%20revisada%2001_07_20.pdf

25. Manuel Sánchez-Vizcaíno J. Early detection and contingency plans for African swine fever. OIE [Internet]. 2010 [cited 29 October 2023];129-37. Available at: https://www.woah.org/app/uploads/2021/03/2010-129-137-sanchezvizcaino-e.pdf

26. European Food Safety Authority EFSA. Scientific Opinion on African Swine Fever. EFSA journal [Internet]. 1 March 2010 [cited 29 October 2023];8(3). Available at: https://efsa.onlinelibrary.wiley.com/doi/abs/10.2903/j.efsa.2010.1556

27. Cwynar P, Stojkov J, Wlazlak K. African swine fever status in Europe.Viruses [Internet]. April 1, 2019 [cited July 11, 2023];11(4). Available at: file:///C:/Users/USUARIO/Downloads/viruses-11-00310%20(1).pdf

28. Mazur-Panasiuk N, Żmudzki J, Woźniakowski G. African swine fever virus - persistence in different environmental conditions and the possibility of its indirect transmission.Journal of Veterinary Research (Poland) [Internet]. 1 September 2019 [cited 4 November 2023];63(3):303-10. Available at: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6749736/pdf/jvetres-63-303.pdf

29. Olesen AS, Hansen MF, Rasmussen TB, Belsham GJ, Bødker R, Bøtner A. Survival and localization of African swine fever virus in stable flies (Stomoxys calcitrans) after feeding on viremic blood using a membrane feeder. Vet Microbiol [Internet]. August 1, 2018 [cited October 29, 2023]; 222:25-9. Available at: https://www.sciencedirect.com/science/article/abs/pii/S0378113518305029?via%3Dihub

30. Gaudreault NN, Madden DW, Wilson WC, Trujillo JD, Richt JA. African Swine Fever Virus: An Emerging DNA Arbovirus. Front Vet Sci [Internet]. May 13, 2020 [cited Nov 10, 2023]; 7:1-17. Available at: https://www.frontiersin.org/articles/10.3389/fvets.2020.00215/full

31. Li Z, Chen W, Qiu Z, Li Y, Fan J, Wu K, et al.African swine fever virus: a review. Life [Internet]. August 1, 2022 [cited July 11, 2023];12(8):2-41. Available at: file:///C:/Users/USUARIO/Downloads/life-12-01255.pdf

32. Blome S, Gabriel C, Cerveza M. Pathogenesis of African swine fever in domestic pigs and European wild boar.Virus Res [Internet]. April 2013 [cited 23 October 2023];173(1):122-30. Available at: file:///C:/Users/USUARIO/Downloads/Pathogenesis%20of%20African%20swine%20fever%20in%20domestic%20pigs%20and%20european%20wild%20boar%20-%20Patog%C3%A9nesis%20de%20la%20peste%20porcina%20africana%20en%20cerdos%20dom%C3%A9sticos%20y%20jabal%C3%ADes%20europeos.en.es.pdf

33. Jover A, Fernández J, Blanco A, Carrasco A, Méndez A, Moyano E, et al. Pathogenesis of acute African swine fever. Journal of the National University of Córdoba [Internet]. January 1990 [cited July 27, 2023]; 01-10. Available at: https://www.researchgate.net/publication/350374645

34. Yang B, Shen C, Zhang D, Zhang T, Shi X, Yang J, et al.Mechanism of interaction between virus and host is inferred from the changes of gene expression in macrophages infected with African swine fever virus CN/GS/2018 strain. Virol J [Internet]. December 1, 2021 [cited December 20, 2023];18(1). Available at: file:///C:/Users/USUARIO/Downloads/s12985-021-01637-6.pdf

35. Hurtado C, Bustos MJ, Granja AG, de León P, Sabina P, López-Viñas E, et al.The African swine fever virus lectin EP153R modulates the surface membrane expression of MHC class I antigens. Arch Virol [Internet]. Feb 2011 [cited 2023 Dec 20];156(2):219-34. Available at: https://link.springer.com/article/10.1007/s00705-010-0846-2

36. Sánchez EG, Pérez-Núñez D, Revilla Y. Mechanisms of entry and endosomal pathway of African swine fever virus [Internet].Vol. 5, Vaccines. MDPI AG; 2017 [cited 20 December 2023]. Available at: https://pubmed.ncbi.nlm.nih.gov/29117102/

37. Leita A, Cartaxeiro C, Coelho R, Cruz B, Parkhouse RM E, Portugal FC, et al.Printed in great Britain the non-haemadsorbing African swine fever virus isolate ASFV/NH/P68 provides a model for defining the protective anti-virus immune response [Internet]. Vol. 82, Journal of General Virology. 2001 [cited December 20, 2023]. Available at: https://www.microbiologyresearch.org/docserver/fulltext/jgv/82/3/0820513a.pdf?expires=1703186220&id=id&accname=guest&checksum=B43D8BBBA1FDA8562500EBBDE00F88D5

38. Zimmerman J, Karriker L, Ramírez A, Schwartz K, Stevenson G, Zhang J. Diseases of Swine [Internet].11th ed. USA: Wiley Blackwell; 2019 [cited 29 November 2023]. 5-1132 p. Available at: https://edisciplinas.usp.br/pluginfile.php/7954215/mod_folder/content/0/Diseases%20of%20Swine%2C%2011th%20Edition%20%28VetBooks%20%281%29.ir%29.pdf

39. Food and Agriculture Organization of the United Nations/ World Organization for Animal Health/ World Bank. Good practices for biosecurity in the pig sector. FAO [Internet].2010 [cited 4 December 2023];01-89. Available at: https://www.fao.org/3/i1435e/i1435e.pdf

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