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Investigation of characteristics and classification for swine vertebral osteomyelitis in South Korea

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Abstract

Background Vertebral osteomyelitis (VO) is a major cause of condemnation in swine slaughterhouses, leading to economic losses for farmers. This study aimed to investigate the characteristics and classification of VO cases in South Korean slaughterhouses, focusing on their relationship with pyemia and their potential to reduce unnecessary total condemnation.

Results Our findings confirmed that swine VOs are often associated with tail-biting injuries, particularly in the posterior vertebrae, underscoring tail biting as a prominent risk factor. *Trueperella pyogenes* were the most prevalent among the bacterial pathogens, while additional less common bacteria were also identified, warranting further research on their potential pathogenic roles. According to the VO classification scheme used in this study, 75% of the 20 VO cases examined were classified as acute VO, whereas the remaining cases were chronic.

It was revealed that only 10% (2/20) of the VO cases were in a state of pyemia at the time of slaughter (true pyemia) and these true pyemia cases were found only in the acute VOs.

Conclusions The VO classification scheme tested in this study demonstrated high sensitivity (100%), indicating its robustness in avoiding false negatives and ensuring food safety. Of the carcasses that could have undergone unnecessary condemnation, 22.2% were excluded. The results indicate that the VO classification scheme is recommended as a measure to reduce unnecessary total condemnation induced by VO.

Keywords Swine, Slaughterhouse, Vertebral osteomyelitis, Pyemia, Total condemnation

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Background

Vertebral osteomyelitis (VO) is an abnormal condition characterized by vertebral infection and inflammation that results in bone destruction and abscess formation [1, 2]. VO is primarily caused by secondary infections with pyogenic bacteria, such as *Staphylococcus aureus*, *Trueperella pyogenes*, *Actinobacillus pleuropneumoniae*, and hemolytic *Streptococcus*, which reach the vertebrae via hematogenous spread [3]. Contributing factors such as tail biting, skin wounds, or other portals of entry for bacterial pathogens can result in systemic infections that may subsequently localize in the vertebral bones [2, 4].

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VO and pyemia are closely related in etiology. The bone marrow is richly vascularized, making it highly susceptible to bacterial infections via the bloodstream [2]. Pyemia is a type of septicemia characterized by the formation of pus-filled abscesses in various organs or tissues owing to the spread of pyogenic bacteria. These bacteria circulate in the bloodstream, leading to secondary infections at multiple sites, including the bones, joints, lungs, and soft tissues. Thus, pyemia provides favorable conditions for VO development.

VO can also occur in various animals including pigs. VO in pigs is a significant public health concern, and is a major cause of total condemnation in slaughterhouses. Therefore, a meat inspection system that filters out contaminated meat is important and efforts to improve this system continue to enhance food safety [5, 6]. In many countries, post-mortem inspection is implemented in slaughterhouses to identify hazards during the slaughtering operation process, including various types of diseases (pneumonia, multiple abscesses, arthritis, osteomyelitis, etc.), leading to the total consumption of pig carcasses under meat inspection regulations for food safety. The regulations on post-mortem inspection specify that meat from animals affected by systemic diseases, which are likely to cause septicemia, pyemia, toxemia, and viremia, must be declared unsuitable for human consumption [1, 2, 7]. Among the risk factors, suppurative VO is one of the major causes of total carcass condemnation because it is a significant potential outcome of pyemic infection [4]. Previous studies have identified swine VO as a major cause of total carcass condemnation in slaughterhouses, accounting for 61.03 and 38.5% of total condemned carcasses in Portugal and Spain, respectively [1, 8].

Pathologically, VOs can be classified into two stages based on the degree of inflammation. Acute VO is characterized by rapid onset, typically occurring within days to weeks. Pathologically, the condition is marked by shiny and moist lesions, evident bone destruction, and the presence of purulent fluid exudate. These features distinguish acute vertebral osteomyelitis from its chronic counterpart, reflecting the intense and active nature of the infection in its early stages [9, 10]. Thus, acute VO cases have a much higher likelihood of developing pyemia than chronic VO cases [2]. Chronic VO is characterized by gradual onset, typically developing over months to years. These lesions exhibit sclerotic changes in the bone and are often circumscribed by adjacent remodeling tissues. Unlike acute lesions, chronic lesions are less likely to have purulent exudate. These features reflect the long-term nature of chronic lesions and the adaptive responses to prolonged inflammation or infection. Additionally, chronic VOs present a lower risk of pyemia than acute cases [2, 9].

In the post-mortem inspection implemented in slaughterhouses, VO is strong evidence of total carcass condemnation owing to its close relationship with pyemia. Nevertheless, it remains controversial whether all pig carcasses affected by VO should be discarded. Acute VO is more likely to be an indicator of pyemia at the time of slaughter. However, chronic VO may be less associated with pyemia. Since pyemia ends weeks or months before slaughter, pyogenic bacteria may have already been eliminated from the body. Therefore, in chronic VO, extensive contamination of the entire carcass is unlikely [1, 7]. In fact, a previous study reported that 33 of 39 VO cases (33%) were determined to be in the current state of pyemia, specifically 60% (12/20) of acute VO cases and 5.26% (1/19) of chronic VO cases were determined to be in the current state of pyemia [2]. If VO cases unrelated to the current pyemia status can be distinguished and excluded from total condemnation, it would be helpful to improve the income of swine farms. Therefore, it is necessary to develop appropriate criteria for classifying VOs [2].

In South Korea, VO is currently observed in slaughterhouses, resulting in economic damage. However, research on VO in Korean slaughterhouses is lacking. Accordingly, to provide timely and accurate information on VOs in slaughterhouses, we conducted a comprehensive investigation of the current status of VOs. In addition, chronic VOs are known to be less associated with pyemia. Therefore, it seems that total condemnation for all VO cases, regardless of whether acute or chronic, appears to be economically wasteful, suggesting that a more targeted approach could be more appropriate and cost-effective. Accordingly, we investigated the relationship between acute/chronic VO and pyemia to validate a VO classification scheme for lowering the total condemnation rate due to VOs while maintaining food safety.

Methods

Retrospective data

Data on postmortem inspections over the past three years were obtained from a slaughterhouse located in Gimje City, South Korea. This slaughterhouse processes approximately 540,000 pigs annually, which accounts for approximately 2.8% of South Korea's total slaughter volume (19 million), and finishing LYD (Landrace × Yorkshire × Duroc) pigs (110–115 kg, 180–200 days) are subjected to slaughter. The data consisted of the date of slaughter, farm ID, and meat inspection codes of carcasses. Based on these data, the prevalence of VO in the carcasses was analyzed.

Observation and classification of VO cases

Observation and sampling of VO cases were carried out at the slaughterhouse mentioned above between

Table 1 Observation of vertebral osteomyelitis (VO) lesions

Item	Description
location of VO lesions	cervical (C), thoracic (T), lumbar (L), sacral (S), and coccygeal (Co) vertebrates
Presence of pyemia-related lesions (PRL)	skin and muscular abscesses, tail, and joint injury
Number of purulent lesions	Total count of VO lesions + PRL
Chronicity of VO lesions	Acute
	Shiny and moist lesions with sometimes congested areas / Evident bone destruction not circumscribed by adjacent remodeling tissue / Fluid purulent exudate
	Chronic
	Moderate bone destruction entirely circumscribed by remodeling tissue / Thickened exudate / No evidence of congestion

February 2023 and April 2024. Gross lesions in VO cases were assessed by directly observing the internal or external surfaces of the carcasses during post-mortem inspection. There was a time constraint to conduct more thorough examinations of VO-affected carcasses because all VO cases were declared unfit for human consumption and were rapidly removed from the slaughter line. Therefore, it was difficult to observe the potential presence of deeper lesions in VO-affected carcasses. Investigators examined bisected carcasses for VO lesions on the carcass splitting section line, where the carcasses were suspended upside down. Additionally, sampling was conducted simultaneously with observation. While observing the lesions, information on VO cases was recorded according to the criteria shown in Table 1.

The VO cases were classified according to the scheme proposed by Vieira-Pinto et al. (2020). The classification scheme was established based on the degree of chronicity (acute/chronic) and the number of purulent lesions (VO + PRL), as shown in Fig. 1. In Vieira-Pinto et al.'s study [2], of a total of 39 VO cases, 33 cases (33%) were determined to be pyemia. Specifically, 60% (12/20) of AVO cases and 20% (1/5) of CVO3 cases were determined to be pyemia. No pyemia was identified in CVO1 (0/12) and 2 (0/2). Therefore, in this scheme, AVO (acute VO case) and CVO3 (chronic VO case with over 3 purulent lesions) cases are designated for total carcass condemnation to strictly prevent the release of contaminated carcasses.

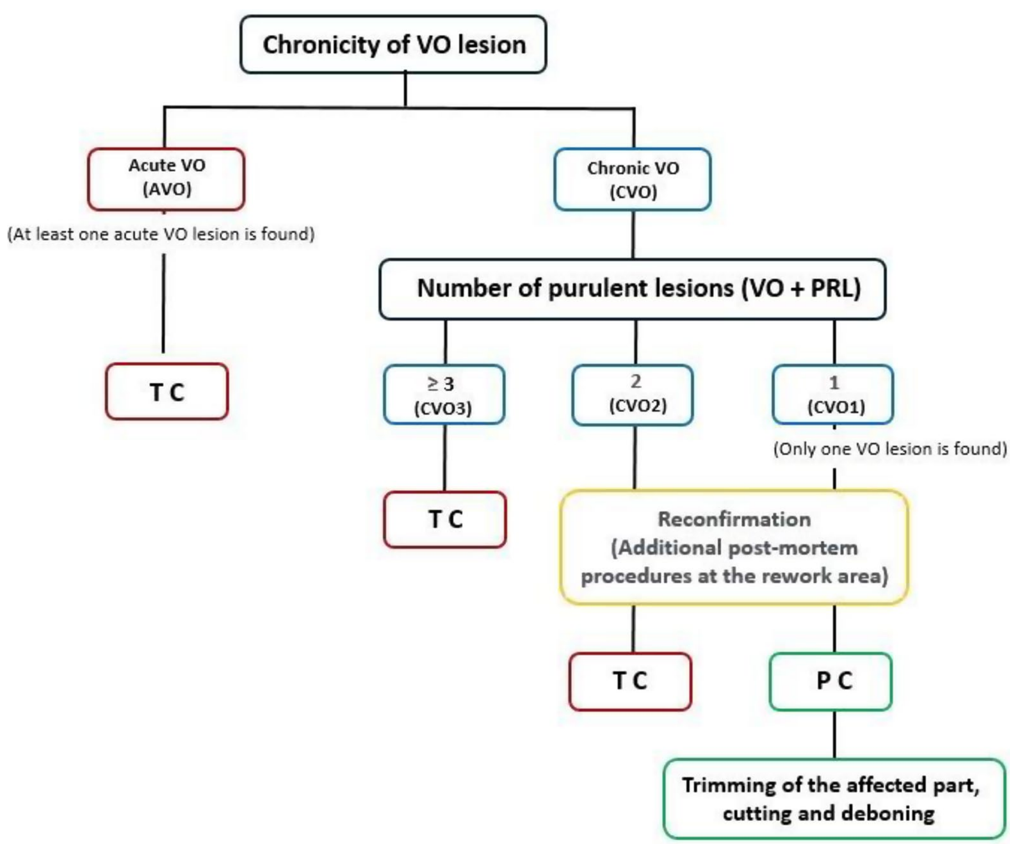

Fig. 1 The VO classification scheme proposed by Vieira-Pinto et al. (2020). The scheme was established based on the degree of chronicity (acute/chronic) and the number of purulent lesions (VO + PRL). VO: vertebral osteomyelitis, PRL: pyemia-related lesions, TC: total condemnation, PC: partial condemnation

Sample collection and identification of true pyemia

To confirm whether the pig with VO was actually in a pyemic state on the day of slaughter, the method used in previous studies [2, 4] was applied with slight modifications. Swab samples from the VO lesions were collected using bacterial culture transport swabs (BD Corporation, US) along with additional muscular tissue samples. All the collected samples were transported to the laboratory, maintained at 4 °C, and subjected to further microbial examination within a day.

VO lesions are likely caused by the hematogenous spread of pyogenic bacteria. This hematogenous spread is considered indicative of pyemia, which serves as the basis for total carcass condemnation. If the pig was indeed in a state of pyemia, there was a high probability that various muscle areas of the carcass would also be contaminated with pyogenic bacteria. Therefore, additional muscular samples were collected for microbial examination. Small pieces of muscular tissue were obtained from three areas of all carcasses affected by VO: the diaphragmatic muscle and the central areas of the thoracic and abdominal walls. If the same bacteria were identified in both VO and muscular tissue samples, it was determined to be true pyemia (indicating the current state of pyemia). Conversely, VO lesions were considered traces of prior pyemia. As controls for VO cases, tissue samples were collected from a normal carcass adjacent to the carcass affected by VO using the same method mentioned above.

Identification of bacteria

Abscess swab samples were directly applied to 5% sheep blood agar (Kisanbio, Seoul, Korea) and streaked using a sterile loop. The surfaces of the muscle samples were decontaminated with 70% ethanol. Subsequently, a piece (approximately 1 g) was cut from each tissue sample. Pieces from the three tissue samples were suspended in 10 mL sterile saline solution and homogenized. After homogenization, the supernatant was added to 5% sheep blood agar and streaked onto a sterile loop. Two streaked agar plates were prepared for each sample and incubated under aerobic and anaerobic conditions (5% CO_2). All plates were incubated at 37 ℃ for 24–48 h if no growth was visible. After the incubation, species identification of the bacterial colonies was performed using Matrix Assisted Laser Desorption Ionization - Time of Flight Mass Spectrometry (MALDI-TOF MS) in a VITEK® MS MALDI-TOF machine (bioMerieux, France) per the manufacturer's protocol. One pure colony was selected and added to 10 µL of 70% formic acid; 1 µL of solution was then mixed with 0.5 µL of matrix solution, dried, and evaluated using MALDI-TOF MS on a VITEK MS platform. Spectral data were analyzed by comparison with typical spectra.

Table 2 Occurrence of vertebral osteomyelitis (VO) cases by month in the slaughterhouse during 2021–2023

Month	2021y	2022y	2023y
Jan	14	23	48
Feb	9	11	31
Mar	27	11	14
Apr	7	25	16
May	8	19	15
Jun	11	27	29
Jul	5	14	24
Aug	7	30	26
Sep	8	25	13
Oct	18	22	11
Nov	28	62	58
Dec	28	48	77
Sum of VO cases	170	317	362
Total no. of carcasses	531,907	554,293	539,571

Statistical analysis

Statistical analysis of all data in this study was performed using R studio v 4.3.1. The trend of VO occurrence in the slaughterhouse over the past three years (2021–2023) was analyzed using a linear mixed model (fixed effect: year, random effect: month) with the lme4 package. VO classification and pyemia determination data were analyzed using binominal or chi-square tests. Data on the prevalence of VO-causing pyogenic bacteria were analyzed using Fisher's exact test.

Results

Occurrence of VO cases during 2021–2023

Table 2. presents the number of VO cases per month. At the slaughterhouses investigated in this study, 531,907 (2021y), 554,293 (2022y), and 539,571 (2023y) pigs were slaughtered, and 170 (0.03%), 317 (0.06%), and 362 (0.07%) VO cases were found in 2021, 2022, and 2023, respectively. When analyzing the variance of VO occurrence by month YoY (year-over-year) with linear regression analysis, it demonstrated an increasing trend in most months except March and October over the past 3 years (Fig. 1). The variance in annual VO cases was analyzed using a linear mixed model. As a result, the number of VO cases continuously increased from 2021 to 2023 ($p < 0.01$, Fig. 2).

Classification of VO cases

A total of 20 cases were classified into four grades according to the classification scheme (Fig. 1). First, the VO cases were classified into acute and chronic states according to the criteria mentioned above. The chronicity of VO lesions was evaluated by focusing on the purulent exudate status. If evidence of a fluid (or fluidal) purulent exudate was found in the lesion, it was determined to be an acute VO (AVO) to avoid ambiguity in the decision

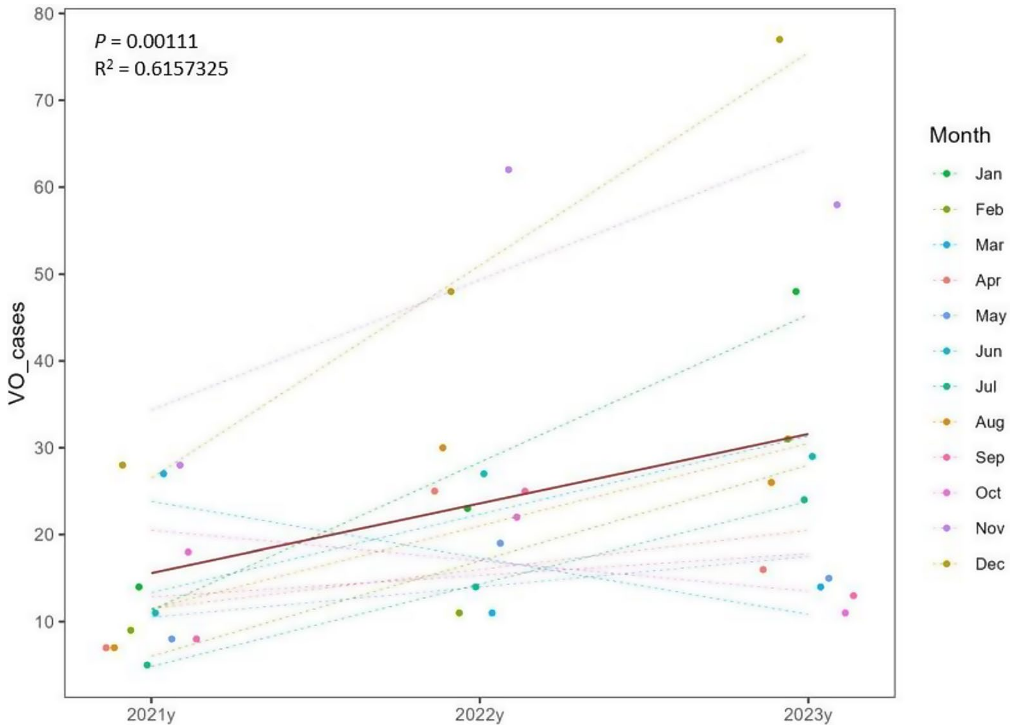

Fig. 2 The scatterplot for the number of vertebral osteomyelitis (VO) cases by month from 2021 to 2023. In the plot, the solid line is the overall linear regression line, and the dashed lines are linear regression lines for each month. In the linear mixed model analysis (fixed effect: year, random effect: month), the number of VO cases tended to increase over the past three years ($p < 0.01$)

regarding chronicity. As a result of the classification of VO cases, acute forms (AVOs) were significantly more prevalent than chronic forms (CVOs, $p < 0.05$). Most VO cases (75%, 15/20) were classified as AVO. The remaining chronic VO (CVO) cases (25%, 5/20) were classified as 1 CVO1, 3 CVO2, and 1 CVO3 cases (Table 3).

A total of 28 VO lesions were found in 20 cases, with 9 (32.1%) in the thoracic vertebrae, 7 (25.0%) in the lumbar vertebrae, 11 (39.3%) in the sacral vertebrae, and 1 (3.6%) in the coccygeal vertebrae. No lesions were observed on the cervical vertebrae. Among the 20 VO cases, more than half of the VO cases (65.0%, 13/20) showed traces of tail biting, but this finding was not statistically significant. Interestingly, among the 17 lesions found in the 13 VO cases with tail injury, most VO lesions (82.4%, 14/17) were found to be significantly higher in the posterior vertebrae (lumbar to coccygeal) ($p < 0.05$), and only 3 VO lesions (17.6%, 3/17) were found in the anterior vertebrae (cervical to thoracic) (Table 3).

Identification of bacteria and determination of true pyemia

Various types of bacteria were detected in the samples. Bacteria identified only in VOs were considered VO-related bacteria, whereas those identified across all samples, including the control carcasses, were regarded as environmental bacteria. In 18 of the 20 VO cases, VO-related bacteria were successfully recovered from the VO lesions. In the 18 VO cases where bacteria were isolated, *Trueperella (T) pyogenes* exhibited the highest prevalence with significance (55.6%, 10/18) ($p < 0.05$), followed by *Streptococcus spp.* (11.1%, 2/18), *Paenibacillus (P) durus* (11.1%, 2/18), *Actinobacillus (A) rossii* (11.1%, 2/18), *Acinetobacter (A) johnsonii* (5.6%, 1/18), *Helcococcus (H) kunzii* (5.6%, 1/18), *Schaalia (S) hyovaginalis* (5.6%, 1/18), and *Staphylococcus (S) hyicus* (5.6%, 1/18) (Table 4).

In the presence of true pyemia in the 20 VO cases, the rate of true pyemia was significantly low, as only two cases (10%, 2/20) were determined to be true pyemia ($p < 0.05$). Specifically, of the AVOs, two were classified as true pyemia, as *T. pyogenes* was isolated from both VO lesions and muscular tissues. The remaining 18 VO cases were classified as false pyemia, and their VO lesions were considered traces of prior pyemia (Table 4).

Association between the results of VO classification scheme and true pyemia

Among the grades from the VO classification scheme, AVO and CVO3 (total condemnation) were assigned as diagnostically positive, while CVO1 and CVO2 (partial condemnation) were assigned as diagnostically negative. The sensitivity (true positive / (true positive + false negative)) and specificity (true negative / (false positive + true negative)) of the VO classification scheme were 100% (2/2) and 22.2% (4/18), respectively (Table 5).

Table 3 Macroscopic characteristics of vertebral osteomyelitis (VO) cases

Sample ID	Grade[1]	Location[2]	A(acute) / C(chronic)[3]	Pyemia-Related Lesions (PRL)	No. of purulent lesions[4]
1	AVO	S3	C	tail injury	1
2	AVO	T2	C	-	1
3	AVO	T7	A	muscular abscess (1), tail injury	2
4	CVO3	L7	C	tail injury	3
		S1	C		
5	AVO	T3	A	muscular abscess (1), tail injury	2
6	AVO	T9	A	skin abscess (1), muscular abscess (1), tail injury	3
7	AVO	L6	A	-	2
		S1	A		
8	AVO	L6	A	tail injury	1
9	AVO	S2	A	skin abscess (1), tail injury	3
		S4	A		
10	AVO	S3	A	skin abscess (2), tail injury	3
11	AVO	S2	A	muscular abscess (1)	2
12	AVO	S3	A	tail injury	3
		Co	A		
13	AVO	L7	A	tail injury	3
		S3	A		
14	AVO	T8	A	-	2
		T11	A		
15	AVO	T3	A	-	3
		L7	A		
		S3	C		
16	AVO	L7	A	-	1
17	CVO2	T4	C	tail injury	2
18	CVO2	L2	C	tail injury	2
19	CVO2	S1	C	tail injury	2
20	CVO1	T1	C	-	1

[1]Grade determined by the classification scheme proposed by Vieira-Pinto et al. (2020). AVO: acute vertebral osteomyelitis; CVO: chronic vertebral osteomyelitis

[2]Vertebrae number of vertebral sections (T: thoracic, L: lumbar, S: sacral, Co: coccygeal) affected by VO

[3]Chronicity of VO lesion

[4]Sum of VO and PRLs

Discussion

VO is one of the major factors leading to a decision of total condemnation in swine slaughterhouses, and it remains a persistent issue in several countries [2, 4, 7, 11]. Recently, as the occurrence of VO has increased in slaughterhouses (Fig. 2), the total number of condemnations caused by VO has also increased, leading to growing complaints from swine farmers. In response to this issue, we investigated the current status of VOs occurring in slaughterhouses and explored potential measures to reduce the total number of condemnations caused by VO by applying the VO classification scheme.

VO can be induced by secondary infections due to various factors, including contaminated injections, tail biting, skin wounds, and abscesses of internal organs. Several studies have highlighted the close relationship between VO and tail biting, which is considered an important source of secondary infection, leading to pyemia and subsequent VO [8, 12, 13]. Especially, it is known that coccygeal and sacral VOs are typically a result of tail-biting [2, 7]. In this study, results supporting the above findings were obtained. Among the 20 VO cases investigated in this study, many (65.0%, 13/20) presented with tail injuries. Furthermore, among the 17 VO lesions found in the 13 VO cases with tail biting, most (82.4%, 14/17) were observed to be significantly higher in the posterior vertebral region (lumbar to coccygeal) than in the anterior vertebral region (cervical to thoracic) ($p < 0.05$), suggesting that pyogenic bacteria invading through tail injuries may have migrated to the nearby or adjacent vertebrae. Cervical osteomyelitis can occur through direct invasion of bacteria from muscular abscesses, which may develop from contaminated injections [2, 7]. However, cervical osteomyelitis was not observed in this study. Based on these results, tail biting is considered a more potent risk factor than contaminated injections in porcine VO.

T. pyogenes, *Streptococcus*, and *Staphylococcus* spp. are well-known pathogens related to VO [2, 4, 7, 11]. Although they are a part of the commensal microbiota, they can cause opportunistic infections with suppurative

Table 4 Bacterial identification and determination of true pyemia

Sample ID	Grade[1]	No. of PRL	Pyemia (T/F) [2]	Samples		
				vertebral osteomyelitis (VO)	**Muscular tissues[3]**	**Control carcass[4]**
1	AVO	1	F	*Aeromonas (A) sobria,* *Aeromonas (A) hydrophila*	*A. hydrophila,* *A. sobria,* *Enterobacter (E) asburiae*	*A. sobria,* *A. hydrophila*
2	AVO	1	F	*A. sobria*	*A. sobria,* *A. hydrophila*	*A. sobria,* *A. hydrophila* *E. asburiae*
3	AVO	2	F	***Paenibacillus (P) durus***[5]	*A. sobria,* *A. hydrophila*	*A. sobria,* *A. hydrophila*
4	CVO3	3	F	*A. sobria,* ***Trueperella (T) pyogenes***	*A. sobria,* *A. hydrophila*	*A. sobria,* *A. hydrophila*
5	AVO	2	F	*A. sobria,* ***P. durus***	*A. sobria,* *A. hydrophila*	*E. asburiae,* *A. seifertii*
6	AVO	3	F	***T. pyogenes***	*Lactococcus (L) garvieae,* *Acinetobacter (A) seifertii*	*E. asburiae,* *A. seifertii*
7	AVO	2	F	***T. pyogenes***	-	-
8	AVO	1	F	***T. pyogenes***	-	-
9	AVO	3	F	***Helcococcus (H) kunzii,*** ***Schaalia (S) hyovaginalis***	*Aeromonas (A) veronii,* *Acinetobacter (A) nosocomialis,* *Acinetobacter (A) haemolyticus*	*A. haemolyticus,* *A. nosocomialis*
10	AVO	3	F	*A. haemolyticus,* *A. nosocomialis,* ***T. pyogenes***	*A. nosocomialis,* *E. asburiae*	*A. haemolyticus,* *A. nosocomialis*
11	AVO	2	**T**	*E. asburiae,* ***T. pyogenes***	***T. pyogenes*** *L. garvieae* *Klebsiella (K) oxytoca*	*Serratia (S) marcescens,* *Escherichia (E) coli,* *A. hydrophila,*
12	AVO	3	F	***Acinetobacter (A) johnsonii***	*Staphylococcus (S) warneri* *Acinetobacter (A). seifertii* *A. hydrophila,*	*A. hydrophila,* *E. asburiae*
13	AVO	3	F	***T. pyogenes***	*A. sobria*	*A. sobria*
14	AVO	2	F	***T. pyogenes***	*A. sobria*	*A. sobria*
15	AVO	3	**T**	***T. pyogenes,*** ***Staphylococcus (S) hyicus***	***T. pyogenes***	-
16	AVO	1	F	***Streptococcus (S) oralis***	*E. asburiae*	*A. hydrophila* *E. asburiae*
17	CVO2	2	F	***Actinobacillus (A) rossii***	*Citrobacter (C) braakii* *A. hydrophila*	*A. hydrophila* *E. asburiae*
18	CVO2	2	F	***Streptococcus (S)dysgalactiae ssp. dysgalactiae***	*Citrobacter (C) freundii,* *Klebsiella (K) pneumoniae* *A. seifertii, E. coli*	*A. seifertii* *E. asburiae*
19	CVO2	2	F	***A. rossii***	*C. freundii* *A. nosocomialis*	*A. hydrophila* *C. freundii,* *E. asburiae*
20	CVO1	1	F	***T. pyogenes***	*A. hydrophila* *A. nosocomialis*	*A. hydrophila* *A. nosocomialis*

[1]Grade determined by the classification scheme proposed by Vieira-Pinto et al. (2020)

[2]T: true pyemia (consistency between bacteria recovered from both VO and muscular samples), F: false pyemia (inconsistency between bacteria recovered from both VO and muscular samples)

[3]Tissues samples from diaphragmatic muscle, thoracic and abdominal walls of a carcass affected by VO

[4]A normal carcass adjacent to the carcass affected by VO

[5]Underlined bold letters indicate putative bacteria causing VO

lesions [14–16]. Particularly, *T. pyogenes* is the most common pathogen causing VO in pigs [2, 7]. Similarly, in this study, *T. pyogenes* exhibited the highest prevalence (55.6%, 10/18) in VO cases where bacteria were isolated ($p < 0.05$), whereas *Streptococcus* (*oralis* and *dysgalactiae*) and *Staphylococcus* (*hyicus*) demonstrated a relatively lower prevalence of 11.1% (2/18) and 5.6% (1/18), respectively.

Table 5 Association between the results of vertebral osteomyelitis (VO) classification scheme and true pyemia

VO Classification Scheme (diagnostic)		True Pyemia Status[1] (Microbiological Analysis)		Total
		Positive	Negative	
Positive	AVO	2	13	16
	CVO3	0	1	
	Subtotal	**2**[a]	**14**[b]	
Negative	CVO1	0	1	4
	CVO2	0	3	
	Subtotal	**0**[c]	**4**[d]	
Total		2	18	20
Sensitivity / Specificity*		100% (2/2) / 22.2% (4/18)		

[1]Sensitivity = a / (a + c), Specificity = b / (b + d)

[a]True positive, [b]False positive, [c]False negative, [d]True negative

Uncommon bacteria have also been isolated from VO patients. These bacteria comprised (11.1%, 2/18) *A. rossii* (11.1%, 2/18), *Schaalia* (previously named *Actinomyces*) *hyovaginalis* (5.6%, 1/18), *A. johnsonii* (5.6%, 1/18) and *H. kunzii* (5.6%, 1/18). Information regarding the association between these bacteria and porcine VO is lacking; therefore, further investigations are necessary. However, the pathogenicity of *A. rossii*, *S. hyovaginalis*, and *H. kunzii* in pigs has been previously reported. *A. rossii* is thought to cause abortion because it is typically isolated from vaginal discharge/exudate or aborted fetuses [17, 18]. *S. hyovaginalis* and *H. kunzii* have been isolated from pyemic lung [19] and purulent urocystitis [20] lesions in pigs, respectively, suggesting their possible pathogenic roles in pigs. However, swine infections caused by the other two bacteria have not been reported. The genus *Paenibacillus* (including *P. durus*) has been isolated from various environments, animals, and plants. Some species are pathogens of honeybees or other invertebrates, whereas others cause opportunistic infections in humans [21]. *A. johnsonii* is found in aquatic sources, human skin, and animals and can cause severe human infections, such as bacteremia [22].

It is nearly impossible to maintain completely aseptic conditions on the operation line during slaughter; therefore, bacterial contamination of the carcasses is inevitable. In this study, various environmental bacteria were identified in these samples. These bacteria, including *Aeromonas spp.* [23], *Acinetobacter spp.* [24], *Citrobacter spp.* [25], *Klebsiella spp.* [26], *Enterobacter asburiae* [27], *Lactococcus garvieae* [28], *Serratia marcescens* [29], and *Staphylococcus warneri* [30], are usually found in the surrounding environment (water, soil, medical instruments, etc.) or are commensal bacteria of animals, but they can also induce nosocomial or opportunistic infections in humans. Similar to the VO-related bacteria, these bacteria can cause opportunistic infections in slaughterhouses. Thus, as a potential occupational risk for workers, management and caution regarding these bacteria are required.

In this study, the rate of true pyemia in the 20 VO cases investigated was significantly low, at 10% (2/20) ($p < 0.05$).

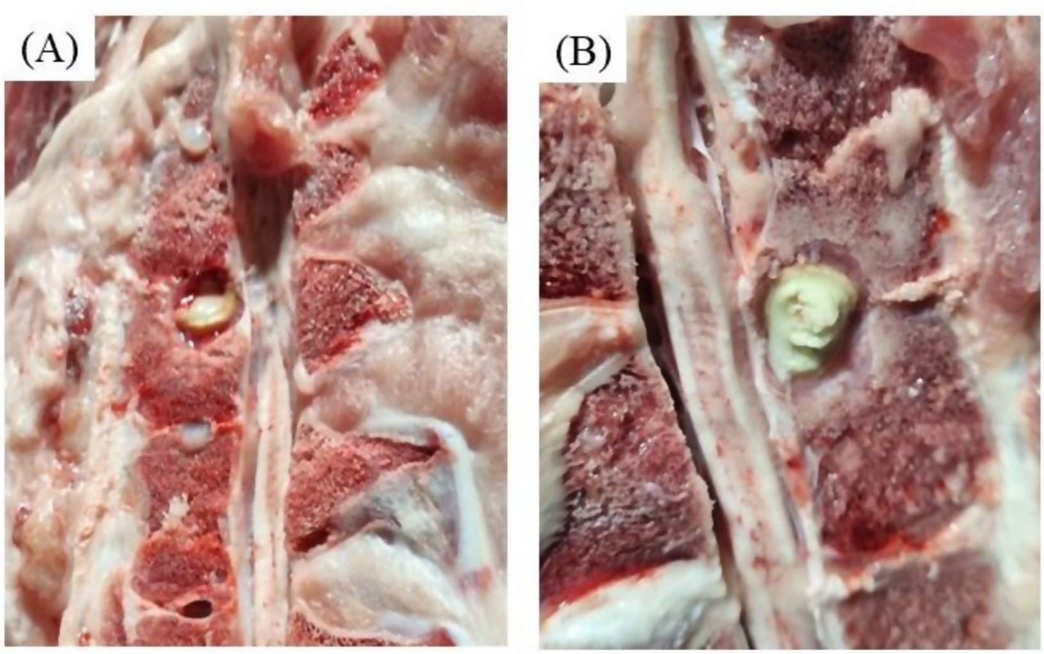

Fig. 3 Chronicity of vertebral osteomyelitis (VO) lesions. VO lesions were classified into acute or chronic status. (**A**) Acute VO presented a shiny and moist lesion with congested area, bone destruction not circumscribed by remodeling tissue and fluidal purulent exudate. (**B**) Chronic VO presented bone destruction well circumscribed by remodeling tissue and thickened and dried exudate

This result suggests that the probability of actual pyemia being present at the time of slaughter in pigs affected by VO is very low, and the current practice of condemning all VO cases at slaughterhouses results in significant economic loss. Therefore, the VO classification scheme to address this issue was validated in this study. In this scheme (Fig. 1), determining chronicity is a critical criterion for determining total condemnation. Acute lesions are related to systemic (generalized) disease before slaughter; therefore, it is important to correctly classify them during post-mortem inspection to avoid false negatives (classify chronic rather than acute) [2, 7]. For instance, AVO cases with true pyemia can be misclassified as CVO1 or CVO2, leading to partial instead of total condemnation. To avoid generating false negatives, strict criteria were applied in the evaluation of the chronicity of VO lesions in this study. When evidence of fluid (or fluidal) purulent exudate was found in the lesion, it was classified as an AVO. Of course, such strict evaluation increases the possibility of generating false positives (classifying meat as acute instead of chronic). However, from a food safety perspective, it is important not to release contaminated meat into the supply chain, so this method was adopted. When the VO classification scheme was applied, the sensitivity was 100% (Table 5), indicating that the scheme is safe because it does not generate false negatives. The specificity was 22.2% (4/18), indicating that 4 carcasses could be saved from the 18 carcasses that were unnecessary for total condemnation (designated for total condemnation under the current system). Considering our results, if CVO3 is assigned as a diagnostic negative (leading to partial condemnation), one more carcass can be spared from total condemnation. However, there is a risk of generating false negatives, and because this study did not secure sufficient samples for each classification grade (AVO and CVO1-3), further investigation with more VO samples is required to improve the scheme. If this scheme is implemented in the field, CVO1 and CVO2 will be released to the market after the partial condemnation process. Purulent materials derived from VO can potentially contaminate the carcass, either directly or through the cutting saw. Thus, according to the standard operating procedure of slaughterhouses, the cutting saw should be more thoroughly disinfected (by washing with hot water at 82 °C) after each cutting operation, and the bisected carcasses should be washed more meticulously.

This study had a limited sample size ($n = 20$). Nevertheless, it was successfully revealed that only 10% (2/20) of the VO cases investigated were in a state of pyemia at the time of slaughter. This finding suggests that determining all VO cases as total carcass condemnation is economically wasted. Furthermore, when applying the VO classification scheme, we were able to exclude 20% (4/20) of the VO cases from the total carcass condemnation while ensuring that contaminated carcasses were not released, resulting in economic benefits. These results provide a basis for further research on this topic. Post-mortem inspection at slaughterhouses is a crucial procedure for public health; therefore, caution is necessary when considering the application of the scheme used in this study to field conditions. Therefore, more extensive (large sample size, different regions) and continuous (longitudinal) research is needed to validate this scheme more thoroughly.

Conclusion

VO remains a major cause of condemnation in swine slaughterhouses. Our findings confirmed that swine VOs are often associated with tail-biting injuries, particularly in the posterior vertebrae, underscoring tail biting as a prominent risk factor. *Trueperella pyogenes* were the most prevalent among the bacterial pathogens, while additional less common bacteria were also identified, warranting further research on their potential pathogenic roles. The presence of environmental bacteria, although less likely to cause VO, highlights occupational risks for slaughterhouse workers and suggests the need for enhanced hygiene protocols. The VO classification scheme tested in this study demonstrated high sensitivity (100%), indicating its robustness in avoiding false negatives and ensuring food safety. When applying this scheme, 22.2% of the carcasses affected by VO were excluded from unnecessary total condemnation. With limited sample data, this study recommends further research to refine this classification scheme, potentially saving more carcasses from total condemnation without compromising food safety. Nonetheless, this VO classification scheme is recommended as a measure to reduce unnecessary total condemnation induced by VO.

Abbreviations

VO	Vertebral osteomyelitis
C	Cervical vertebrates
T	Thoracic vertebrates
L	Lumbar vertebrates
S	Sacral vertebrates
Co	Coccygeal vertebrates
PRL	Pyemia-related lesions
AVO	Acute vertebral osteomyelitis
CVO	Chronic vertebral osteomyelitis

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Author contributions
Conceptualization of the study was carried out by DL, CI, SK, and JH. The supervision of the project was overseen by DL, CI, and JH. The investigation was conducted by SH, SK, and YK. Formal analysis was performed by SH, SK, and YK, while data curation was handled by SH and SK. The original draft of the manuscript was written by SH, SK, and YK. The writing and review of the manuscript, including editing, were done by SH, YK, DL, CI, SK, and JH.

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Data availability
No datasets were generated or analysed during the current study.

Declarations

Ethics approval
This study was not submitted for ethics committee review because it is a retrospective analysis of Dodram Pig Farmers Cooperative Veterinary Clinic, Daejeon, Korea. Ethics approval is unnecessary in owners submitted to the slaughterhouse office according to the Animal Protection Act (No. 13023; Ministry of Agriculture, Food and Rural Affairs, South Korea).

Consent for publication
Not applicable.

Competing interests
The authors declare no competing interests.

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