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Granulomatous lymphadenitis in swine: validation of national data based on identification by the service of federal inspection (SFI)

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Introduction

The granulomatous lymphadenitis (GL) in pigs is mainly caused by *Mycobacterium avium hominissuis* (MAH), who belongs to the *Mycobacterium avium* Complex (MAC), considered non-tuberculous mycobacteria (NTM). Although GL does not affect swine zootechnical performance, economic losses occur during the slaughter line by condemning viscera and carcasses. The lesion is characterized by one or more foci of granuloma, which most frequently affect the organs of the digestive tract and peripheral lymph nodes. The main differential diagnosis encompasses *Mycobacterium bovis* (*M. bovis*) and *Mycobacterium tuberculosis* (*M. tuberculosis*), who belongs to the *Mycobacterium tuberculosis* Complex (MTbC), of relevant zoonotic potential. Nevertheless macroscopic examinations and histopathology are insufficient to determine the etiologic agent involved. Federal Meat Inspection had registered the frequency of 0,81% of lymphadenitis in Brazil from 2012 to 2014. The aim of this study was to investigate the etiology of the granulomatous lesions for validation of the national data of inspection and build a database to further risk analysis.

Material and Methods

In 2017 the federal veterinary inspectors collected mesenteric lymph nodes with granulomatous lesions of 399 swine production farms located in eight states (Rio Grande do Sul, Santa Catarina, Paraná, São Paulo, Minas Gerais, Mato Grosso, Mato Grosso do Sul e Goiás), representing 158 brazilian municipalities. If available, mesenteric lymph nodes with GL of three swine on each farm were sampled, totaling 257 lymph nodes from finisher pigs and 142 from sows/boars. The tissues were submitted to histological examination and bacterial analysis. Mycobacterial isolation and identification were performed according to OIE (2018). Briefly, the isolates from Lowenstein Jensen and/or Stonebrink media, positive to acid-fast bacteria in Ziehl-Neelsen (ZN), were typified by Polymerase Chain Reaction (PCR) protocols (Table 1) for genus and species, supporting the distinction between MAH, *Mycobacterium avium avium/silvaticum*, *M. bovis* and *M. tuberculosis*.

Results

Comparing histological findings with macroscopic examination, the Service of Federal Inspection (SFI) correctly identified 85% of granulomatous lesions in individual basis. The rate isolation of mycobacteria was 32,08% (128/399), of which 76,56% (98/128) were positive for MAH, 1,56% (02/128) for *M. bovis*, and 21,87% (28/128) only for *Mycobacterium* spp. The identification of *Mycobacterium* species by state is shown in table 2.

Discussion and Conclusion

Overall, the results had shown a good assurance between the evaluation performed in the slaughter line by the veterinarians inspectors and the histopathologic exam. The positive predictive value is higher than 80% when we compare the macroscopic examination

Table 1: Description of the primers used in the Polymerase Chain Reaction (PCR)

Primers	Sequence	Lenght of PCR product	Reference
DNAJ	5´- GGG TGA CGC GAC ATG GCC CA -3´ 3´- CGG GTT TCG TCG TAC TCC TT -5´	236bp	(TAKEWAKI et al., 1993)
IS1245	5´- GCC GCC GAA ACG ATC TAC- 3´ 3´- AGG TGG CGT CGA GGA AGAC -5´	427bp	(GUERRERO et al., 1995)
IS901	M IS901F 5´- GGATTGCTAACCCACGTGGTG -3´ M IS901R 3´- GCCAGTAGCTTGATGAGCG -5´	577bp	(MORAVKOVA et al., 2008)
INS	INS1 5´- CGTGAGGGCATCGAGGTGGC - 3´ INS2 3´- GCG TAGGCGTCGGTGACAAA -5´	245bp	(VAN EMBDEN et al., 1993)
RD4	5´ AACGCGACGACCTCATATTC 3´ 3´ AAGGCGAACAGATTCAGCAT 5´	400bp	(SALES et al., 2014)

Table 2: Identification of *Mycobacterium* species by swine production farms as a function of the state of origin

State of origin	MAH	<i>Mycobacterium</i> spp.	<i>M. bovis</i>	Negative	Number of samples
Goiás	01(10.00)	0 (0.00)	0 (0.00)	09 (90.00)	10
Minas Gerais	13 (18.06)	11 (15.28)	0 (0.00)	48 (66.67)	72
Mato Grosso do Sul	0 (0.00)	0 (0.00)	0 (0.00)	03 (100.00)	03
Mato Grosso	0 (0.00)	0 (0.00)	0 (0.00)	4 (100.00)	04
Paraná	17 (31.48)	06 (11.11)	0 (0.00)	31 (57.41)	54
Rio Grande do Sul	15 (28.30)	06 (11.32)	02 (3.77)	30 (56.60)	53
Santa Catarina	42 (24.85)	05 (2.96)	0 (0.00)	122 (72.19)	169
São Paulo	09 (27.27)	0 (0.00)	0 (0.00)	24 (72.73)	33

and the lymph nodes that showed histopathological lesions of granulomatous lymphadenitis. The present study shows a high prevalence of MAH causing GL in Brazilian farms, confirming this subspecies as the most prevalent in the swine population as it has been described in other countries. The hypothesis of fecal-oral transmission between animals may justify the permanence and prevalence of MAH subspecies in pig farms. The two samples that were positive for *M. bovis* were collected at the same slaughterhouse, but they were from different farms, located in different towns. Both of farms raise pigs and dairy cattle. Anyway the source of the infection was not defined. Nevertheless, due to the disease prevalence in pigs and differences in zoonotic potential between the etiological agents, lesions of porcine granulomatous lymphadenitis should be considered in the definitions of the exams performed by the SFI.

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