

Correlations Between Lung Pneumonic Lesions and Serologic Status to Key Respiratory Pathogens in Slaughtered Pigs in Northern Uganda

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Abstract

Background: A cross-sectional study was conducted in three selected slaughter slabs in Lira district Uganda in 2019 to (i) determine prevalence and severity of pneumonia and (ii) establish associations between lung pneumonic lesions and serologic status to key respiratory pathogens in slaughter pigs. Blood samples were collected from pigs at antemortem from which sera were prepared. Using enzyme-linked immunosorbent (ELISA) assays, sera were screened for antibodies against *Mycoplasma hyopneumoniae* (*M. hyo*), *Actinobacillus pleuropneumoniae* (*App*), porcine reproductive and respiratory syndrome virus (PRRSv) and porcine circovirus type 2 (PCV2). At post-mortem, lungs were scored for possible pneumonic lesions and the type of pneumonia as previously described. Pneumonia types were grossly characterized as catarrhal purulent bronchopneumonia (CPBP), pleuropneumonia (PLP) and pleuritis as previously described. *Metastrongylus spp* infection was determined by examining lungs for gross presence or absence of helminths as previously described. T-tests were used to compute prevalence of pneumonia. Chi-square tests were used to compare the percentage of lungs positive or negative to CPBP, PLP and pleuritis. Ordinal logistic regression model was used to evaluate the odds of multiple pneumonia forms, with serostatus for different pathogens and *Metastrongylus spp.* infection as predictors.

Results: One hundred and sixty seven (n=167) lungs were examined for pneumonic lesions. The prevalences of CPBP, PLP and pleuritis were 29.9% (95% CI 22.9-36.9), 74.2% (95% CI 67.5-80.9) and 17.3% (95% CI 22.4-36.3), respectively. The odds of multiple pneumonia forms increased in pigs with multiple pathogens and concurrent *Metastrongylus spp.* infestation (ORs 2.6, p=0.01 and 2.5, p=0.003, respectively), reaffirming synergistic effects of coinfections in the induction of lesions.

Conclusions: This study revealed a high prevalence and severity of pneumonic lesions in slaughter pigs. It provides baseline information, undeniable evidence on the magnitude of pneumonia and justifies future studies on its potential economic impacts in Ugandan pigs.

Background

Respiratory diseases contribute significant economic losses to swine producers worldwide through increased mortality, retarded growth rates, reduced feed conversion and reproductive performance [1–3]. Other losses arise from additional costs of treatment [4], loss of potential revenue and vaccinations [5, 6]. Various infectious agents are associated with lung lesions at slaughter [7]. Among these agents, *M. hyo*, *App*, PRRSv and swine influenza viruses (SIV H1N1) are the most important agents associated with gross pneumonic lesions in pigs [8, 9]. However, other bacterial or viral agents are known to contribute significant pneumonic lesions in pigs. For example, concurrent infections of *M. hyo* with other agents such as PCV2 or PRRSv were found to increase the severity and duration of mycoplasmal pneumonia [10].

To establish the contribution of respiratory pathogens to lung lesions, it is necessary to score lungs at slaughter. This enables monitoring of herd health [7, 11], and provides baseline information for future epidemiologic studies. One of the cost-effective methods for this purpose is abattoir surveys as they provide a valuable source of data and information useful to support herd health management decisions. Rapid gross visual and detailed lung scores are used to accurately assess extent of pathological lesions associated with

enzootic pneumonia in pigs, due to occurrence of distinct gross lesions [12, 13]. Serologic and clinical evidence provides useful information on the extent and severity of pulmonary lesions. Besides, it is important for monitoring growth, as pneumonic lesions (such as pleurisy) have been associated with growth retardation in pigs [14, 15].

In all types of production systems, pig growth is a key productivity indicator that is affected by respiratory disease in a herd, which in turn affects herd profitability. In Uganda, no information is available on the actual extent of pneumonia, its impact on growth and any associations of lung lesions with serologic or clinical profiles in pigs, as no studies have been previously conducted. Thus, the contribution of pneumonia to overall economic performance of swine herds cannot be estimated, which hampers the design of effective interventions. The aims of this study were to (i) determine the prevalence and the severity of pneumonic lesions in slaughter pigs, and (ii) establish the relationships between lung pneumonic lesions and serologic status to key respiratory pathogens (PCV2, PRRSV, *M. hyo* and *App*) detected in slaughter pigs in Lira district, mid northern Uganda.

Materials And Methods

Study area and design

We conducted a cross-sectional slaughter slab survey in Lira district, mid-northern Uganda from March to September 2019. Lira district is located in latitudes 2° 14' 59.64" North and longitudes 32° 53' 59.46" East. Pigs slaughtered in the selected slabs were sourced from within Lira district (~ 70%) and from neighboring districts of Dokolo, Agago, Alebtong and Kole (~ 30%). Visits were made during early morning hours when slaughters were conducted and on days when slaughters were known to be high.

Sampling of slaughter slabs and pigs

The study was conducted in three purposely selected slaughter slabs in the district based on high daily slaughter capacity (range 8–20 pigs). These slabs represented about 60% of all pigs slaughtered in the district (*DVO, pers. comm*). The three (3) slaughter slabs were: Teso bar, Adekokwok and Amach market. In each slab, pigs brought for slaughter from different sources were randomly sampled (approx. 40%) on a given day. A list of all pigs brought for slaughter was made and each was allocated a number (on a piece of paper) from which a simple random sample was drawn. Other characteristics of pigs (live weights using a measuring tape, body condition scores (BCS) and sex) were recorded at antemortem. The unit of measurement was the individual pig and the outcome variable were serologic status to 4 respiratory pathogens (PCV2, PRRSV, *M. hyo* and *App*).

Determination of sample size

A recent study reported a seroprevalence of 20.9% for *M. hyo* in pigs in Lira district [16]. A review of lung scoring methods by Garcia-Morante et al. showed that 80% of pigs infected with *M. hyo* had lung lesions [17]. Using these estimates, the projected proportion of *M. hyo*-positive pigs with pneumonic lesions in Lira district was estimated to be 0.8 of 20.9% = 16.72%. We assumed no clustering effect within a slab, since pigs were

bought from different farms. Thus, the required sample size for a 5% level of significance was derived from the equation [18]:

$$n = \frac{Z_{\alpha/2}^2 pq}{d^2} \text{----- Eq (1)}$$

Where $Z_{\alpha/2}$ is the standard normal deviate for $\alpha = 1.960$; $p = 0.16.72$, $q = 1 - p = 0.8328$ and d , the effect size estimated to be 6% ($d = 0.06$). Using the above equation, a sample size of 149 pigs was computed.

Blood sample collection

At ante-mortem, blood samples were collected from pigs for sera preparation. Each pig was properly restrained as described in the ILRI Standard Operating Procedures (SOPs) manual, Sect. 2, part (c) & (d) [19]. Blood was collected from the jugular vein, using a 21G, 1.5" needle into plain 5 mL BD® vacutainer tubes. The tubes were labeled and then placed in an ice box containing ice packs at 4°C. After collection, samples were delivered to the district veterinary laboratory, where they were left to stand at room temperature (20°C) overnight. After 24 hours, sera were harvested into 2 mL cryotubes (Sarstedt®, Germany), labelled and stored in a fridge at -20°C until use.

Serologic analysis of sera

Serologic assays were done at the College of Veterinary Medicine, Animal Resources and Biosecurity (CoVAB), Makerere University. Sera were screened using ELISA test kits for each of the four key pathogens using the protocols described by each manufacturer: *M. hyo* and *App-ApxIV* (IDEXX, Westbrook, Maine, USA), while for PRRSv and PCV2 (Krishgen Biosystems, India). Results were computed as a sample to positive ratio (S/P) using the equation:

$$S/P = \frac{(\text{Sample OD} - \text{Average of negative control})}{(\text{Average of positive control} - \text{Average of negative control})} \text{----- Eq (2)}$$

Cut-off sample to positive ratios (S/P%) for *M. hyo* were > 0.40 (positive) and < 0.30 (negative), *APP* were \geq 50% (positive) and < 40% (negative). PCV2 and PRRSv S/P cut-off ratios for positive and negative samples were \geq 0.2 and < 0.2 respectively. Suspect samples were re-tested.

Lung lesion scoring procedures

At post mortem of the animals from which sera were collected, detailed scoring of lung lesions was conducted. To ensure accuracy of data collected, records for each pig were entered into a sheet of paper at antemortem. The first author performed the lesion scoring, while being assisted by a research assistant to record observations on an excel-designed sheet. Lungs were isolated from the thoracic cavity, placed on a flat clean surface, palpated and scored for visible pneumonic and pleuritic lesions. Palpation for hardened areas of hepatization (pneumonia or pneumonia-like) was done and the percent involvement per lung lobe was recorded. Incisions onto the lung parenchyma using a surgical blade were made to identify and characterise any deep-seated lesions. The gross lung lesion scoring procedures for CPBP, PLP and pleuritis were done as previously described [13, 20].

Lesions were classified as catarrhal purulent bronchopneumonia (CPBP), pleuropneumonia (PLP) or pleuritis [21]). CPBP is characterized by cranio-ventral consolidation, reddish to pink areas, with mucous or purulent exudate on the cut surfaces of the lung. PLP includes lesions that are typical of one consolidated focus or more, mainly in the caudal lobes, with red to dark areas with fibrinous pleuritis and hemorrhages and necrosis on the cut surface [13]. Pleuritis lesions were classified into 3 grades as previously described [14, 20]. Grade 0 represents no pleuritis, grade 1 is where up to 5% of the lung surface is affected and grade 2 is where > 5% of lung surface is affected (adhesions between lung lobes or between lobes and the thoracic cavity, mediastinum or pericardium). CPBP and PLP values were binary coded (1 = present and 0 = absent).

The prevalence of pneumonia and the percent of lung tissue affected by pneumonia (proportion of lung surface visibly affected by pneumonia) was determined as previously described [22]. The percent of pneumonia is based on the proportion of lung surface that is abnormally firm and discolored [10, 23, 24]. Each lung lobe was assigned points based on the approximate volume represented by that lobe. In all, ten points were assigned to the right cranial, right middle, left cranial and left middle lobes. For the right and left caudal lobes, each was assigned 27.5 points (15 for dorsal and 12.5 for ventral parts), making a total of 100 points for the entire lung [22]. Score values for macroscopic lung lesions ranged from 0 to 100%. In brief, the following parameters were calculated:

$$i. \text{ Percent of lung surface affected by pneumonia} = \frac{\% \text{ total area affected}}{100} \text{----- Eq (3)}$$

$$ii. \text{ Percent prevalence of pneumonia} = \frac{\text{Number of lungs affected by pneumonia}}{\text{Total number of lungs examined}} \times 100 \text{----- Eq (4)}$$

Scoring for lung helminths infestations

Gross helminths infestations (*Metastrongylus spp*) were detected by examining the diaphragmatic lung lobes for wedge-shaped areas during the lung scoring procedures. Incisions were made on a grossly affected lung lobe with a surgical blade or strips of one centimeter tissue from the edge of diaphragmatic lung lobes were trimmed and squeezed to express adult worms (slender, 30–50 mm in length) from the bronchi [25]. Infestations were scored as present (code = 1) or absent (code = 0).

Data analysis

Data was coded and entered into *Excel 16.0 (Excel Corp, TX)*. The data was then exported to RStudio (R CoreTeam, 2019) for analysis and presentation. Missing data were omitted from the analysis. Descriptive summary statistics was done using t-tests. Chi-square or Fisher exact tests were used to compare the proportion of lungs scored as positive or negative to CPBP, PLP and pleuritis. Ordered logistic regression model was used to evaluate the odds of scoring positive to multiple pneumonia types as a dependent variable, with serostatus to different pathogens as predictors. Adjustment for *Metastrongylus spp* as a potential confounder was made by checking for a change in the model coefficient at 10% cut off when it was excluded from the model. Since the data was non-normally distributed, a Wilcoxon Rank sum test was used to compare median pneumonia lung scores by serologic status at significance level $\alpha = 0.05$.

Results

In total, 167 pigs were sampled and examined from three selected slaughter slabs. Overall, more female pigs (55.7%) were sampled, of which 17.2% (n=16) were pregnant. Live weights varied from 26 to 184 kg, while the age range was from 5 to 50 months. Table 1 below shows summary statistics of pigs sampled.

Table 1: Summary statistics of pigs sampled

Slaughter slab	No. of pigs sampled, % (n)		Age (months) Mean \pm SD	Live weight (kg) Mean \pm SD
	Males	Females		
Teso Bar	21.55 (36)	28.14 (47)	13.9 \pm 9.1	58.7 \pm 31.6
Adekokwok	2.4 (4)	5.4 (9)	13.6 \pm 3.8	70.3 \pm 16.8
Amach market	20.35 (34)	22.15 (37)	14.6 \pm 6.8	68.6 \pm 27.9
Totals	44.3 (74)	55.7 (93)	14.2 \pm 8.0	63.6 \pm 30.0

SD=standard deviation

Prevalence of pneumonia

Overall, the prevalence of pneumonia was generally high in all the slaughter slabs, and varied from 69.20% in Adekokwok to 80.28% at Amach market slaughter slab. Table 2 below shows summary statistics.

Table 2: Percent prevalence of pneumonia in three slaughter slabs

Slaughter slab	% prev (n)	95% CI
Teso Bar	79.51 (71)	77.16-92.60
Adekokwok	69.20 (13)	61.93-76.47
Amach market	80.28 (83)	70.68-89.90
Total	79.04 (132)	72.80-85.27

% prev= percent prevalence, CI = confidence interval

Prevalence of pneumonic lesions (CPBP, PLP and pleuritis) and other lesions observed

Overall, PLP was the most prevalent pneumonic lesion observed. About 30% of sampled pigs also had *Metastrongylus spp* nematodes in the lungs. Table 3 shows summary statistics of forms of pneumonic lesions observed in this study (mean prevalence and 95% confidence interval).

Table 3: Prevalence of CPBP, PLP, pleuritis lesion scores and other lesions observed

Pneumonic & other lesions	% prevalence (n)	95% CI
CPBP	29.9 (50)	22.9-36.9
PLP	74.2 (124)	67.5-80.9
Pleuritis	17.3 (29)	11.6-23.2
<i>Metastrongylus spp</i> infection	29.3 (49)	22.4-36.3
Abscesses	2.39 (4)	0.052-4.73

CI = confidence interval

Associations between total pneumonia scores and respiratory pathogen serologic status

Figure 1 shows that overall, the median lesion scores for pigs that tested seropositive to each of the 4 pathogens were higher than those that tested seronegative. *App*-positive pigs showed significantly higher median lesion scores than *App*-negative pigs. Also, PCV2-positive pigs were found to have marginally higher total median lesion scores than PCV2-negative pigs. Figure 1 below highlights summary statistics of median pneumonia scores by pathogen type.

Fig 1: Boxplot of total lesion scores by pathogen serologic status

Lesion scores for the selected pathogens

The Wilcoxon Rank Sum test showed no significant differences between pigs that tested positive and negative to both PRRSv and *M. hyo*. For PCV2, a marginal difference in median lesion scores was observed. In contrast, there was a significant statistical difference in MLS between pigs that were *APP*-positive and those that were *App*-negative. Table 4 below shows a summary of results.

Pneumonia types by pathogen serologic status

Table 4 below summarises the relationships between serologic status and lung lesion scores. As observed, there was a moderate association in the proportions of pigs scored for the different lung lesions and serologic status. While statistically insignificant, the odds of detecting CPBP, PLP and pleuritis increased in pigs that tested PCV2-, *M.hyo*- and *App*-positive, suggesting possible associations.

Table 4: Results of logistic regression models for pneumonia types and pathogen serologic status

Pneumonia type	Predictors	Estimate	Std Error	OR	95% CI	z-value	Pr(> z)
CPBP	Intercept	-1.262	0.241	0.282	0.172-0.446	-5.233	1.67e-07***
	<i>APP</i>	0.540	0.384	1.717	0.799-3.638	1.405	0.1599
	<i>Met. spp</i>	0.828	0.361	2.290	1.124-4.667	2.291	0.0219*
PLP	Intercept	0.645	0.202	1.906	1.291-2.859	3.194	0.00140**
	<i>Met. spp</i>	1.392	0.515	4.023	1.583-2.398	2.704	0.00685**
	<i>M. hyo</i>	1.739	1.056	5.694	1.074-10.533	1.647	0.09956.
Pleuritis	Intercept	-1.8015	0.2647	0.165	0.095-0.269	-6.805	1.01e-11***
	PCV2	0.4978	0.5286	1.645	0.543-4.452	0.942	0.346
	<i>APP</i>	0.5633	0.4436	1.756	0.714-4.130	1.270	0.204

Met. spp = *Metastrongylus spp*, ORs = Odds ratios, CI = Conf. intervals; *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$

Ordinal logistic regression model of effect of coinfections on pneumonia occurrence

Table 5 below shows that the odds of scoring positive to multiple pneumonia types significantly increased in pigs with concurrent *Metastrongylus spp* infestations and with dual respiratory infections.

Table 5: Ordinal regression model of factors for occurrence of multiple gross pneumonia types

Predictors	Coeff.	Std error	ORs	95% CI	t-value	p-value
Single infection	0.3446	0.365	1.411	0.690-2.898	0.943	0.345
Coinfection (2 pathogens)	0.9876	0.416	2.684	1.192-6.125	2.373	0.0176*
Coinfection (3 pathogens)	0.2313	1.037	1.260	0.155-10.131	0.223	0.823
<i>Metastrongylus spp</i>	0.9526	0.330	2.592	1.364-4.993	2.883	0.0039**
Intercepts						
0 1	-1.2504	0.250	-	-	-4.989	0.0000***
1 2	1.2995	0.248	-	-	5.234	0.0000***
2 3	3.8065	0.444	-	-	8.563	0.0000***

* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, CI = Conf. interval

The figures (2a-2h) below present different gross forms of pneumonia observed in the lungs examined during the study.

Discussion

In general, a high prevalence and severity of pneumonia was observed in this study. The percentage of 79% of pigs with lung lesions in this study was comparable to that found in other studies in comparable production systems, which reported 73.9% in Brazil [26]. In western France, [8] reported that pneumonia (69.3% of lungs) and pleurisy (15% of lungs) were the most frequent lesions seen at slaughter. Large variations in pneumonia and pleuritis (41–76% and 2–35%, respectively) were reported in Brittany, France [14, 27]. A similar observation was made for other studies in Italy, which reported prevalences of 59.6% and 61.4% respectively [11, 28]. A study in Phillipines reported that 48% of slaughter pigs had high lung scores, while 22% had pleurisy [29]. In Brazil, a recent study reported that 68.5% of slaughter pigs presented with macroscopic lung lesions [15]. In Belgium, the prevalence of pleuritis and pneumonia was reported to be 20.76% and 23.85%, respectively [30]. In Makurdi, Benue State, Nigeria, it was reported that 36.4% of sampled pigs had lung lesions [31]. Differences in these studies likely reflect differences in production systems, hygiene and health status overall.

The prevalence of pleuritic lesions found in this study is comparable to other studies elsewhere, in which it was found to vary between 20.5% and 33.1% in Sweden [32]. In south eastern Norway, it was reported that pneumonic or pleuritic lesions were found in 84% of the lungs and that of bronchopneumonia was found in 70% of the lungs examined [33]. A study in France reported that pleuritis and cranio-ventral pulmonary consolidation lesions were recorded in 26.8% and 55.7% of slaughter-aged pigs, respectively [9].

In contrast, the high prevalence of pneumonic lesions found in this study is higher than that reported in Eastern Spain, which reported the mean lesion values for CPBP, PLP and pleuritis as 8.4%, 0.4% and 0.3% of farms, respectively [13]. In Ghana, [34] found that only 5% of slaughtered pigs had pneumonia, though the form of pneumonia was not reported. These studies demonstrate a wide variation in prevalence and severity of lung pneumonic lesions, which may be explained by differences in production systems and management factors, which ultimately influence exposure levels to respiratory pathogens. Also, the different study designs, scoring methods used, environmental conditions and production systems in which pigs are raised account for variations in pneumonia prevalence and severity.

Overall, the odds of detecting CPBP, PLP and pleuritis were higher in pigs that tested seropositive to all the pathogens (ORs 1.2–6.2), except for PRRSv on CPBP. This suggests possible associations between pathogen seropositivity and lesion scores. However, differences in median pneumonic scores between seropositive and seronegative pigs were statistically insignificant. *M. hyo* is reported to be strongly associated with lung lesions [35] and pulmonary consolidation [36]. In contrast, the odds of detecting pigs with CPBP and PLP increased in pigs that tested positive to *APP*. The finding that only *APP*-seropositive pigs resulted in a significantly higher MLS could be due to either infection with pathogenic *APP* serotypes, or that coinfections with *APP* and other pathogens produced considerable lung damage. This conforms to documented reports of the gross morphological features of *APP* as characterised by fibrinonecrotic pleuritis and diffuse haemorrhages [25]. These findings are not unusual, as previous studies have documented associations between lung lesions and serology to respiratory pathogens in pigs [32, 36].

Table 6 showed relationships between pneumonia type and multiple infections. Results of the regression model showed that the odds of detecting multiple pneumonia forms increased in pigs with coinfections and *Metastrongylus spp*. This corroborates previous studies which documented synergistic or potentiating effects of coinfections between PRRSv and other pathogens [10], PCV2 and other pathogens in the induction of respiratory disease [9, 37, 38].

This study did not find association between CPBP scores and PRRSv seropositivity. The observed PLP and pleuritis lesions could be due to the effect of coinfections between PRRSv and other pathogens, notably PCV2 and *M. hyo*. The ability of *M. hyo* infection to potentiate and prolong PRRSv-induced pneumonia clinically and macroscopically has been documented [10]. Our findings that *M. hyo* seropositivity was more associated with PLP than CPBP (OR 6.2 vs 1.3 respectively) instead contrasts with other studies which found that *M. hyo* is associated with CPBP [13]. Notwithstanding differences in the study design by [13], which sampled only heavy pigs (100kg), we sampled pigs of varying ages and live weights from predominantly small scale production systems. The disease progression from acute to chronic as pigs grow older, may explain differences in the lesion scores observed.

The finding that approximately a third of the pigs sampled had *Metastrongylus spp* nematodes, frequently observed in the tips of diaphragmatic lobes, is in agreement with a previous study, which found high prevalences of GIT nematodes in Ugandan pigs [39]. GIT parasites such as *Ascaris suum*, *Metastrongylus spp* are known to induce pulmonary tissue damage through their migratory larvae, increasing the susceptibility of pigs to various respiratory pathogens [25].

Apart from two studies in Nigeria and Ghana, no other published study was found in Africa (with comparable production systems) which documented the magnitude of pneumonia in pigs. It is worthy to mention that in Uganda, no other published study or report was found on the magnitude of pneumonia in pigs. Thus, in the context of different pig production systems documented in Uganda [40], our findings can only be extrapolated to the swine population in northern Uganda, with similar husbandry systems. This study showed that a high proportion of pigs brought for slaughter in the region presented with high prevalence and severity of pneumonic lesions, and the association between lesions and serologic status suggests a significant contribution of the studied pathogens in lung pathology.

Limitations of the study

The scoring methodology used in this study may have underestimated the actual magnitude of pneumonia, since some pigs may have suffered early in life and lesions could have resolved. We acknowledge the method used for estimation of surface area grossly affected by pneumonia may have reduced accuracy in estimation of lesion scores. Besides, due to the need to perform the scoring process quickly to match with the slaughter speed, it is probable that some hidden lesions may have been missed.

Conclusions

This is the first study to document associations of pneumonic lesions with serologic status to key swine respiratory pathogens in slaughter pigs in Uganda. We found that pigs brought for slaughter in the study area

had a high prevalence and severity of pneumonic lesions. The findings of this study establish critical baseline information for future studies on swine respiratory diseases. The high prevalence of pneumonic lesions justifies a need for future studies on potential economic impacts of pneumonia on swine production and productivity in Uganda, as a basis for designing future interventions.

Declarations

Ethics approval and consent to participate

This study was approved by the Institutional Review Committee (IRB), College of Veterinary Medicine, Animal Resources and Biosecurity, Makerere University (IRB # SBLS/REC/18/008), Uganda National Council of Science and Technology (UNCST reg. no. A590); ILRI's Institutional Research Ethics Committee (IREC no. IREC2018-23) and by ILRI's Institutional Animal Care and Use Committee (IACUC2018-22). Prior informed consent was obtained from district local authorities and owners of slaughter slabs before the study commenced.

Consent for publication

Not applicable.

Availability of data and materials

The datasets generated and analysed during the current study are available in this link: <https://data.ilri.org/portal/dataset/multipathogen-survey-and-risk-factors>. We used the STROBE-VET check list (<https://strobevetstatement.files.wordpress.com/2016/09/strobe-vet-checklist.pdf>) in the preparation of this manuscript.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

MMD, FNM, JE and PO designed the study; PO collected data; MMD, BW and PO analyzed and interpreted data, MMD and PO wrote the draft manuscript; all authors have read and approved the final version of the manuscript.

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Prior publication

Data have not been published previously.

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Figures

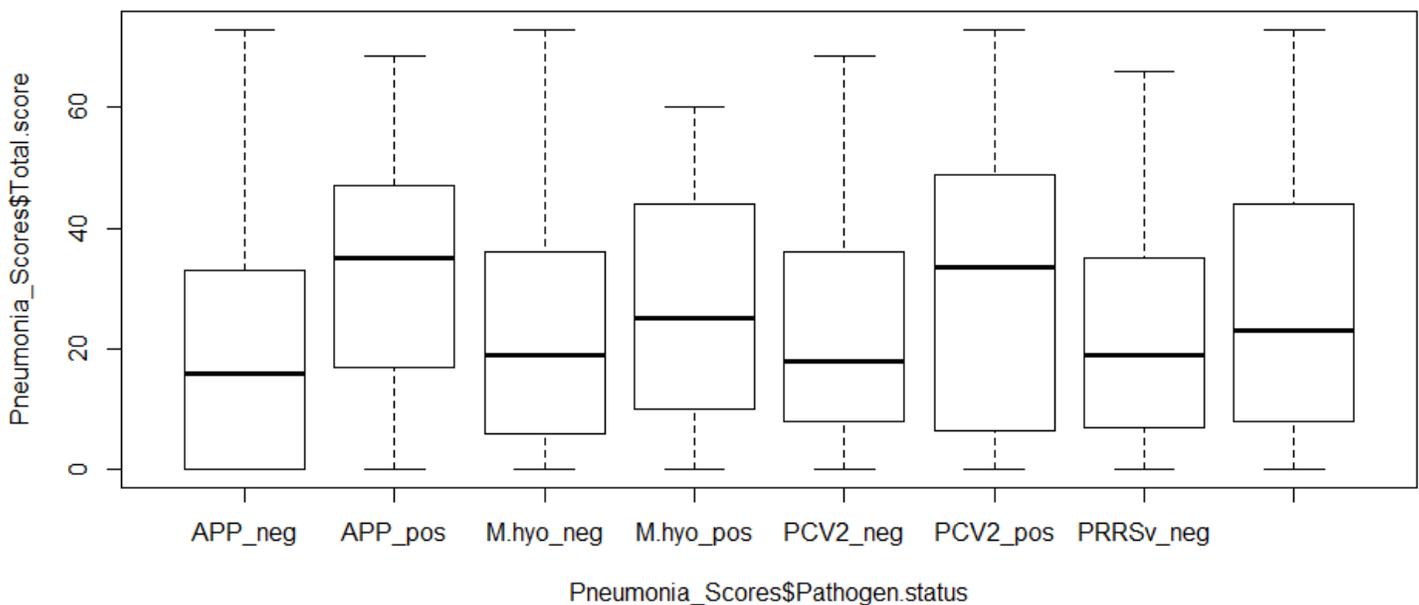


Figure 1

Boxplot of total lesion scores by pathogen serologic status



Figure 2

Pictures of normal lungs (a), lungs with purulent bronchopneumonia showing severe exudation (b); and cranioventral consolidation (c); lungs with diffuse interstitial pneumonia showing a rubbery texture (d); lungs with hemorrhagic bronchopneumonia showing failure to collapse (e) and pleuritis, showing attachments of lung lobes (f); hemorrhagic pleuropneumonia showing hemorrhages on cut surfaces of the lung (g), and a pulmonary abscess with thick caseous yellowish pus (h).

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