



Research article

Classification and correlation of coughing sounds and disease status in fattening pigs

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Abstract

There have been attempts to use technology to distinguish pig coughs from other sounds on farms. Machine learning is being used to classify pig coughs via python. Sound files have been converted to images that are composed of wave plots, spectrograms and log power spectrograms for identification of those sounds. A recorder was used with a total of 45 healthy three-bred weaned piglets, wherein three replications of each were used with 15 weaning pigs per pen during different months. This process was set up within the housing unit at a ratio as 1:1 (recorder per pen). Sounds, blood samples and tonsil swabs were collected every month. Pig cough sounds were then classified from other sounds and a coughing index (CI) was established. Blood samples and tonsil swabs were utilized to determine respiratory diseases via laboratory tests that included ELISA, PCR and bacterial cultures. According to our results, pig coughs sound distinctly different from other sounds as had been classified by python. Moreover, the laboratory results of the seroprofile of *Mycoplasma hyopneumoniae* (*M.hyo*), Porcine Reproductive and Respiratory Syndrome virus (PRRSv), and Porcine Circovirus type 2 (PCV2), as was established by ELISA test, were employed in disease detection during the fattening period. Spearman rank correlations and Kappa analysis were used to establish correlation values between coughing and the results of laboratory tests. CI revealed a high correlation coefficient and agreement with the ELISA results of *M.hyo*, as well as the PCR results of PRRSv and PCV2 ($p < 0.05$), while CI also revealed low correlation coefficient and agreement with the results of the *Streptococcus spp.* and *Pasteurella spp.* cultures ($p > 0.05$). Therefore, the monitoring of coughing can be suited to detect respiratory problems and any potential relationships with *M.hyo*, PRRSv and PCV2 infections.

Keywords: coughing index, correlations, respiratory diseases and fattening pigs

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INTRODUCTION

Respiratory problems on swine farms have resulted in major losses in production, as well as higher mortality rates, reduced feed conversion rates and diminished rates of growth, all of which have led to increased production costs (Ferrari et al., 2008). Zimmerman et al (2019) reported that many pathogens are known to be the cause various respiratory problems including Porcine Reproductive and Respiratory Syndrome virus (PRRSv), Pasteurellosis, Porcine Circovirus type 2 (PCV2), swine influenza, *Mycoplasma hyopneumoniae* (*M.hyo*), Streptococcosis, and others. Pig coughing is an important sign that veterinarians and animal husbandry experts can use to monitor respiratory problems on pig farms (Chedad et al.,2001). The simple observance of coughing is preferred over the laboratory diagnostic process, which involves the collection of bacterial cultures and PCR analysis. This is because the monitoring of coughing can serve as an immediate indicator of respiratory problems and would allow proprietors of farms to more quickly address the problem in a more efficient and affordable manner. That is why coughing can be considered a very important indicator in the monitoring of respiratory problems on pig farms. Even though the importance of coughing diagnosis has been admitted by academic organizations (Mei et al., 2015), there have not yet been any efforts to officially classify pig coughs from other sounds on pig farms. There are a number of limitations associated with the human detection of coughing on pig farms such as the potential for any one person's individual bias; however, there have now been attempts to use technology to help detect pig coughs on farms. The use of machine learning, as a branch of artificial intelligence or AI, can help researchers to more precisely distinguish pig cough sounds from other sounds on pig farms. Moreover, it can also help to accurately identify coughing sounds and can also reduce the chance of any human bias.

Presently, syndromic surveillance can be used to predict any future occurrence of disease outbreaks. This is true in the detection of A/H5N1 Influenza events (Barboza et al., 2013), wherein Web-based expert epidemic intelligence systems were employed to detect health threats through the Global Health Security Initiative (GHSI). This system is responsible for notifying officials about irregularities in drug purchase information and can help identify any sick cases in a given area. There are many systems which use the same principle for early disease detection in animals. These include the surveillance used to identify incidences of diarrhea in small animal practices through the Small Animal Veterinary Surveillance Network (SAVSNET) (Jones et al., 2014). This is also true of the Participatory One Health Disease Detection (PODD) System, which is a community-owned surveillance system that collects data from volunteer reporters, identifies disease outbreaks automatically and notifies the local governments, surrounding villages and relevant authorities (Yano et al., 2018). Moreover, Yamsakul et al. (2023) used infrared temperature sensor for monitoring body temperature of sow herds which it can quickly identify illness sow. Consequently, it would be of great interest to the owners of pig farms if pig cough sounds and animal coughing sounds in general could be detected before serious respiratory problems emerge or a disease outbreak can occur.

The objective of this paper is to classify pig cough sounds from others sounds on pig farms by using AI. This would then allow researchers to identify any potential correlations between pig cough sounds and respiratory disease results obtained from laboratory tests.

MATERIALS AND METHODS

Systemic architecture and hardware

Accordingly, coughing sounds recorded by the Voice Recognition Module V3 system (product by Arduino), which is a compact and easy-to-control speaking recognition board that employs two methods of control: Serial Port (full function) and General Input Pins (partial function). Recorded sounds were then analyzed and coughing sounds were isolated from all other sounds on pig farms, such as the sounds of laborers walking, the opening of doors and the talking of those labors. The computer programming code employed in this study was created via the python language, and sounds were then classified according to three distinct parameters: wave plot, spectrogram and log power spectrogram. Sound files, composed of pig cough sounds and other sounds recorded on the farms, were converted by spectrograms and input into a CNN plus Linear Classifier model. They were then used to produce predictions about the class to which the sound would belong. These datasets would then be presented as images so that comparisons of the different types of sounds can be made. Recorders were set up within the housing unit at a ratio of 1:1 (recorder per pen). The number of pig cough sounds would then be used to establish a coughing index (CI), as will be explained in greater detail below.

Farm and clinical examinations

This study was conducted on farrow to finishing farms located in Lamphun Province, Thailand. On these farms, fattening houses employ open systems where endemic infections of pig populations can occur involving *M.hyo* and porcine reproductive and respiratory syndrome (as has been reported by the Diagnostic Center of the Faculty of Veterinary Medicine, Chiang Mai University). The duration of the study was extended from the weaning period to the fattening period, which lasts about 5 months. A total of 45 healthy three-bred weaned piglets were included in this study (Large White x Landrace x Duroc). Swine Fever Disease vaccine and Foot and Mouth Disease vaccine were administered at 3 and 8 weeks of age, respectively. Three replications were included in this study, while each replication involved 15 weaning pigs per pen. Moreover, each replication took place in a different month as follows: the first replication group was initiated in January of 2021, the second replication group was initiated in April of 2021 and the third replication group was initiated in September of 2021, respectively. The housing unit utilizes an open system, wherein the temperature is dependent upon the existing environmental temperature on the farm. All animals used in the study were cared for according to the standard guidelines of the Institutional Animal Care and Use Committee of the Faculty of Veterinary Medicine, Chiang Mai University (S11/2564). The weight gain of each animal was then recorded during each month.

The coughing index, or CI, was established by [Nathues et al. \(2012\)](#). Accordingly, the coughing sounds of 15 pigs in the experimental pen were

recorded with the use of a recorder between 6:00 - 7:00 am. each month. The number of coughing bouts was counted every 3 minutes (3 counted per time) with a resting time of around 10 minutes between each recording session. The average coughing index for each time period was calculated as follows:

$$\text{Coughing Index (CI)} = \frac{\text{Total number of coughing bouts (CC)}}{[\text{number of examined pigs (n)} * \text{total time of observation (minutes)}]}$$

Sampling and laboratory tests

All pigs included in these experiments were assigned an ID, while the sample collection process involved the implementation of serum and tonsil swabs that were collected every month during the experimental period.

Blood samples and tonsil swabs were randomly collected from five pigs per group every month. Pigs were restrained by snare after placing a mouth gate between the teeth of the upper and lower jaw to keep the mouth wide open. A swab was then carefully wiped on the surface of the tonsil. A blood sample was then collected from the jugular vein of the pig. The first collection time was implemented during the weaning period and the subsequent samples were collected during every month of the experiment until the ultimate selling time. Total time of collection was set at six for each experimental period. Blood was collected from the jugular vein and then stored at 4 °C for least than two hours to guarantee sufficient clotting. After centrifugation at 2,000 g for 10 minutes, serum was separated and stored at -20 C° until being processed at a later time. Tonsil swabs were collected from all pigs via the insertion of rotating swabs in the oral cavity of each pig. The tonsil swabs were employed to obtain a bacterial culture that can specifically be used for the detection of *Streptococcus spp.* and *Pasteurella spp.* All swabs were collected using a sterile collection process. The left nasal swabs were retained in a viral transport medium (VTM) and frozen at -20 C° until further analysis could be implemented.

Enzyme-linked immunosorbent assay examination (ELISA)

A commercial enzyme-linked immunosorbent assay was used to test the sera for antibodies against *M.hyo* (HerdChek *M.hyo*, IDEXX®, USA). In accordance with the manufacturer's instructions, a test result was considered indicative of *M.hyo* when the sample to positive control ratio (S/P) was equal to or greater than a cut-off value of 0.4. Elisa of PRRS antibody detection was carried out using a commercially prepared kit of IDEXX PRRS X3 Ab test (IDEXX®, USA), and each sample was judged according to the sample-to-positive (S/P) ratio (based on the degree of absorbance) with S/P threshold values > 0.4 (Wellenberg et al., 2006). The Elisa of the PCV2 antibody detection process employed a commercially available PCV2 IgG ELISA kit (Biocheck®, UK), wherein S/P was calculated as follows:

$$\text{S/P} = \frac{[\text{OD}_{\text{sample}} - \text{OD}_{\text{mean negative control}}]}{[\text{OD}_{\text{mean positive control}} - \text{OD}_{\text{mean negative control}}]}$$

PCV2 antibody titer was established by applying the following equation with the S/P ratio of the sample set at a 1:50 dilution to an end point titer, which was equal to or greater than a cut-off value of 0.5:

$$\text{Log}_{10} \text{ Titer} = 1.1 * \text{Log}_{10} (\text{S/P}) + 3.361$$

For the PRRSv antibody titer, the serum samples were detected for total PRRSv-specific antibodies using a commercially available PRRSV ELISA kit (HerdCheck PRRS X3 Ab test, IDEXX Laboratories Inc., Westbrook, ME, USA). Serum samples were considered positive for the presence of PRRSv antibody when the average S/P ratio was ≥ 0.4 . For the PRRSv antibody titer, the same samples were tested for the presence of PCV2 antibodies using a commercial enzyme-linked immunosorbent assay (ELISA) kit (Porcine Circovirus type 2 Antibody Test, BioChek B.V., Reeuwijk, Holland) according to the manufacturer's instructions. Serum samples were considered positive for the presence of PCV2 antibody when the average s/p ratio was ≥ 0.5 .

Bacterial cultures

Tonsil swab cultures were collected using selective sheep blood agar plates (Tammer-Tutka Oy, Tampere, Finland). The plates were incubated at $+35^{\circ}\text{C}$ for 24 h for primary inspection and for an additional 24 hr. if the first readout was negative. The agar surface was examined for β -hemolytic and nonhemolytic colonies. Suspected colonies were identified as GBS by employing catalase tests and biochemical tests. They were then confirmed by the Christie, Atkins, Munch, Petersen (CAMP) test and the Latex Agglutination (Streptex B, Remel. Europe. Ltd. UK) (Ross, 1996). Moreover, tonsil swabs were streaked onto trypticase soy blood agar containing 5% sheep blood and incubated in a 5% carbon dioxide atmosphere at 37°C for 24-48 hr. *Streptococcus spp.* and *Pasteurella spp.* were then identified through standard biochemical characteristics (Carter, 1973).

Molecular technique for PRRSv and PCV2 detection

Serum samples obtained from fattening pigs were collected at 4, 8, 12, 16, 20 and 24 weeks of age (five sample pools for one sample) and RNA was extracted according to the method that was previously described (Wasilk et al., 2004). The primers for the virus were designed based on the highly conserved ORF7 region of PRRSv that had been used to detect PRRSv type by Yang et al. (2020). For PCV2, all samples in each age group were tested by PCR by following the protocol of Hjulsager et al. (2009).

Statistical analysis

Coughing indexes, ELISA results, bacterial culture and PCR results of each disease were determined by the Spearman's correlation coefficient and Kappa coefficient of R program version V3.2 (Therneau, 2013), for which the statistical significance was established when p-values were less than 0.05. Other relevant parameters were analyzed by descriptive analysis.

RESULTS

Systemic architecture and hardware

In this study, a recorder was created using the Voice Recognition Module V3 system that had been produced by Arduino[®]. The code used to classify sounds on pig farms involved software that was created with use of the python language, wherein the sound file was classified by three distinct parameters: wave plot, spectrogram and log power spectrogram.

Wave plot of python language was created as follows:

```
def load_sound_files(file_paths):
    raw_sounds = []
    for fp in file_paths:
        X,sr = librosa.load(fp)
        raw_sounds.append(X)
    return raw_sounds
def plot_waves(sound_names,raw_sounds):
    i = 1
    fig = plt.figure(figsize=(25,60), dpi = 40)
    for n,f in zip(sound_names,raw_sounds):
        plt.subplot(10,1,i)
        librosa.display.waveplot(np.array(f),sr=22050)
        plt.title(n.title())
        i += 1
    plt.suptitle("Figure 1: Waveplot",x=0.5, y=0.915,fontsize=18)
    plt.show()
```

Spectrogram of python language was created as follows:

```
def plot_specgram(sound_names,raw_sounds):
    i = 1
    NFFT = 1024 # the length of the windowing segments
    Fs = 22050 # the sampling frequency
    fig = plt.figure(figsize=(25,60), dpi = 40)
    for n,f in zip(sound_names,raw_sounds):
        plt.subplot(10,1,i)
        specgram(np.array(f), Fs=22050)
        # specgram(np.array(f), NFFT=NFFT, Fs=Fs, noverlap=900)
        plt.title(n.title())
        i += 1
    plt.suptitle("Figure 2: Spectrogram",x=0.5, y=0.915,fontsize=18)
    plt.show()
```

Log power spectrogram of python language was created as follows:

```
def plot_log_power_specgram(sound_names,raw_sounds):
    i = 1
    fig = plt.figure(figsize=(25,60), dpi = 40)
    for n,f in zip(sound_names,raw_sounds):
        plt.subplot(10,1,i)
        D = librosa.amplitude_to_db(np.abs(librosa.stft(f))**2)
        librosa.display.specshow(D,x_axis='time', y_axis='log')
        plt.title(n.title())
        i += 1
    plt.suptitle("Figure 3: Log power spectrogram",x=0.5,
y=0.915,fontsize=18)
    plt.show()
```

The sound file obtained from the recorder was converted to an image (Figure 1) that was composed of a wave plot (Figure 1A), a spectrogram (Figure 1B) and a log power spectrogram (Figure 1C). The wave plot depicts the attitude of sound and time, while the spectrogram is a concise ‘snapshot’ of an audio wave. It is an image that is used to plot frequency (y-axis) and time (x-axis) and uses different colors to indicate the amplitude of each frequency. The brighter the color and the higher the line would be indicative of the degree of energy of the signal of the sound. The log power spectrogram indicates the relationship between hertz, decibel (Bar scale) and time.

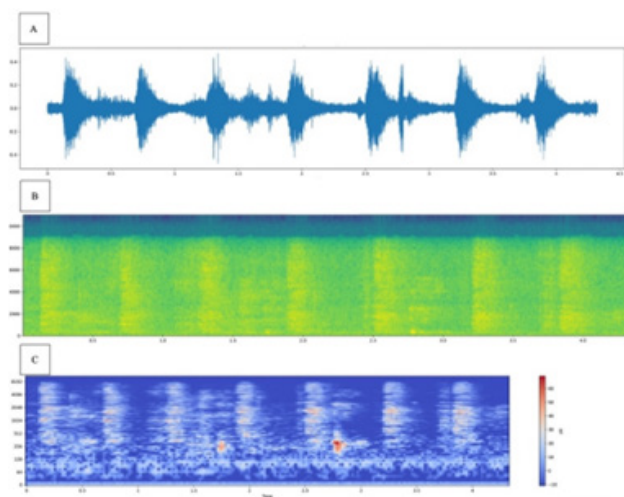


Figure 1 Images obtained from the sound file composed of a wave plot (A), a spectrogram (B) and a log power spectrogram (C), respectively.

For the purposes of classification of the different sounds, the sound files obtained from the recorder were analyzed to distinguish between coughing and other sounds recorded on these pig farms. These sounds were classified according to three parameters: wave plot, spectrogram and log power spectrogram, as are shown in Figure 2. Consequently, pig cough sounds were found to differ from the other sounds in all three methods of analysis. Thus, the wave plot for coughing (Figure 2 A) revealed a stable rhythm of the attitude of sound and time, but not for the other sounds (Figures B, C, D). Moreover, the spectrogram and log power spectrogram of coughing indicate that different colors are representative of the amplitude of each frequency, which was in accordance with the pattern presented on the plot wave. Accordingly, this would indicate that there was a stable rhythm of the attitude of sound and time for the coughing sounds but not for the other sounds.

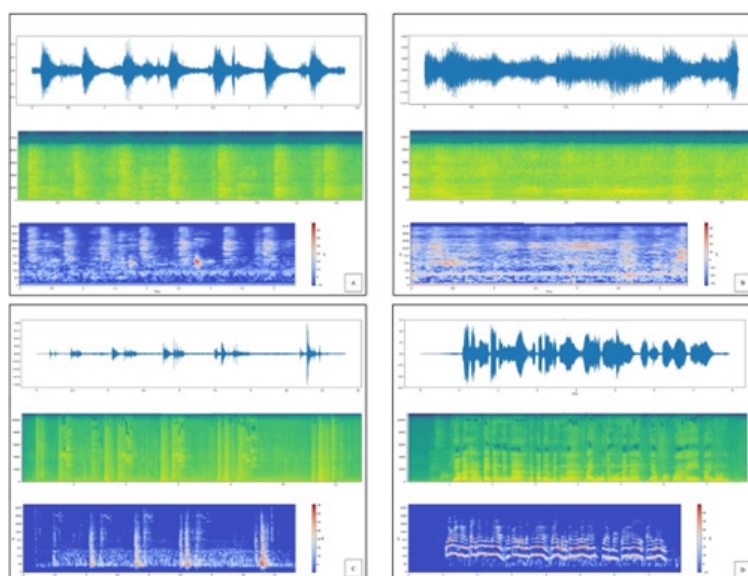


Figure 2 The image character of each type of sound: coughing (A), machine sounds (B), walking steps of laborers (C) and other sounds made by laborers (D). Each type of sound is represented by a wave plot, a spectrogram and a log power spectrogram.

Farm and clinical examinations

For growth performance, the experiment involved three replications. Each one was performed according to the same protocol, but the initial period of time for each replication was different (January, April and September). The growth performance parameter was composed of initial weight, final weight, body weight, average daily gain and %loss, all of which are showed in Table 1. Initial weight and final weight for each replication group were not found to be significantly different ($p>0.05$), of which the final weight of members of replication group 3 gave the best result when compared with the other replications. However, the average degree of growth performance of all replications was too low, especially in Replication 2, when compared with the growth performance standard (Wüstholtz et al., 2017).

Table 1 Growth performance of each replication group of piglets, of which the last column depicts the average of all parameters (mean±SD) (Replication1-3; n=45, Average of all group; n=3).

Parameters	Replication 1 (n=15)	Replication 2 (n=15)	Replication 3 (n=15)	Average (n=3)
Initial weight: kg.	6.63±0.37	6.24±0.52	6.54±0.40	6.47±0.20
Final weight: kg.	82.31±2.97	81.68±5.63	85.88±4.25	83.29±2.27
Body weight gain: kg.	74.65	74.48	78.87	76.00±2.49
ADG: gram/day	600.59	598.77	629.67	609.68±17.34
%loss	13.33	13.33	6.67	11.11±3.85

kg. = kilogram, ADG = Average Daily Gain

For coughing index, in each replication, the sounds of fifteen pigs per pen were recorded every month and these sounds were then classified to establish a coughing index (CI). The CI of each age of all replications was calculated as an average and is presented in Figure 3. CI was determined after 8 weeks of age. Moreover, at 16, 20, and 24 weeks of age, the pigs displayed high CI values when compared with other ages indicating the possible presence of a respiratory problem (Davis, 1983). This was particularly true at ages between 12 weeks to 16 weeks, as is indicated by the high slope of the graph.

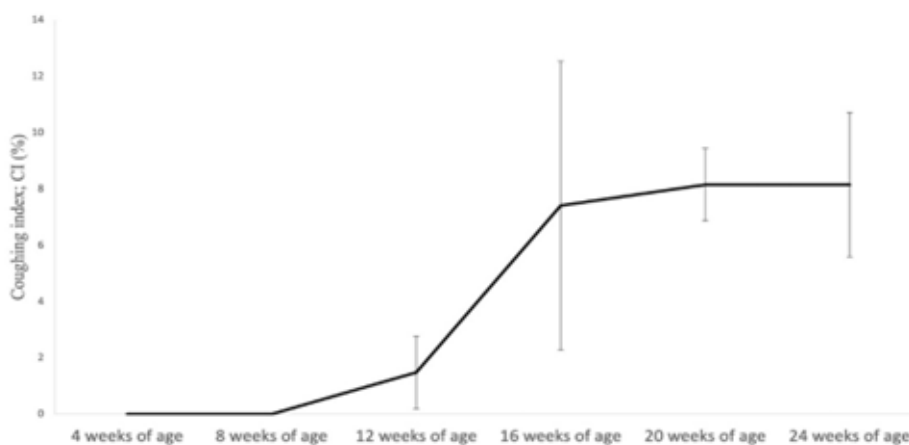


Figure 3 Average coughing indexes and standard deviation at each age of the pigs.

Seroprevalence of *M.hyo*, PRRSV and PCV2 from ELISA

Each replication involved five pigs per group during each month of the experiment to produce a graph (Figures 4, 5, 6) that was representative of the seroprevalence of the disease. These seroprevalence profiles were transformed from the serum as a positive ratio (s/p ratio) for each disease. The results of the seroprevalence profile of *M.hyo* for Replications 1-3 are shown in Figure 4 (A, B, C). The resulting pattern indicated a degree of similarity of 20% positive as was established by ELISA at 12 weeks of age. Furthermore, both the percent positive value and the average s/p ratio at 16 – 24 weeks of age were higher than for the other ages. The results of the seroprevalence profile of PRRSV and PCV2 were similar to the previous results for the disease. A negative result at an early age was observed, but a positive result was recorded at 12 weeks of age or more (Figures 5. A, B, C). The results of the seroprevalence profile of PCV2 differed from the results of both *M.hyo* and PRRSV, wherein the positive results were observed to be high at 4 and 8 weeks of age, and after 16 weeks to 24 weeks of age (Figures 6. A, B, C).

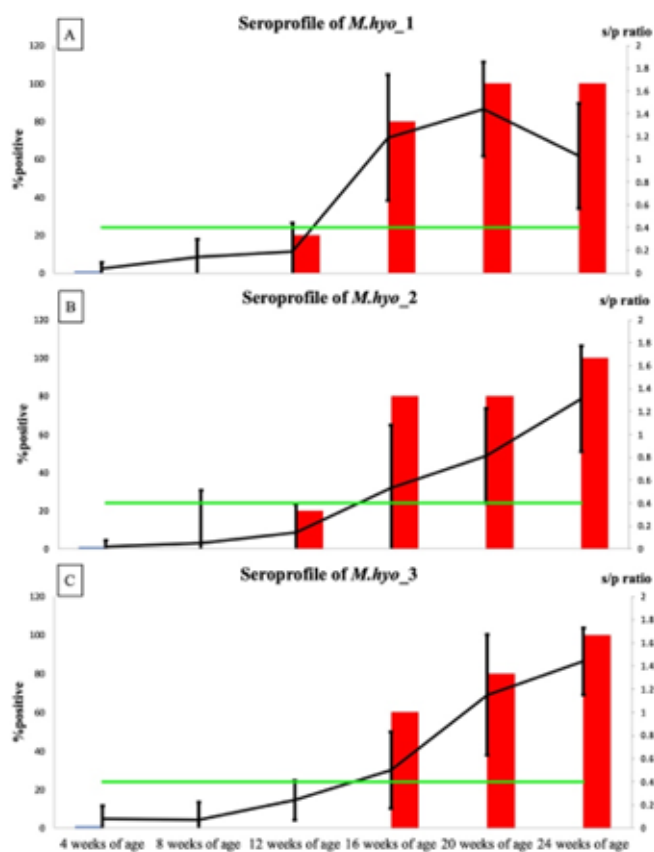


Figure 4 Seroprevalence profile of *M.hyo* (A, B, C) from the ELISA of each replication group; graphs plotted as percent positive of s/p ratio as shown on the left y-axis and average s/p ratio of each age is shown on the right y-axis. The x-axis is representative of the age of the pig, whereas the green line represents the cut-off value of each disease in the ELISA test.

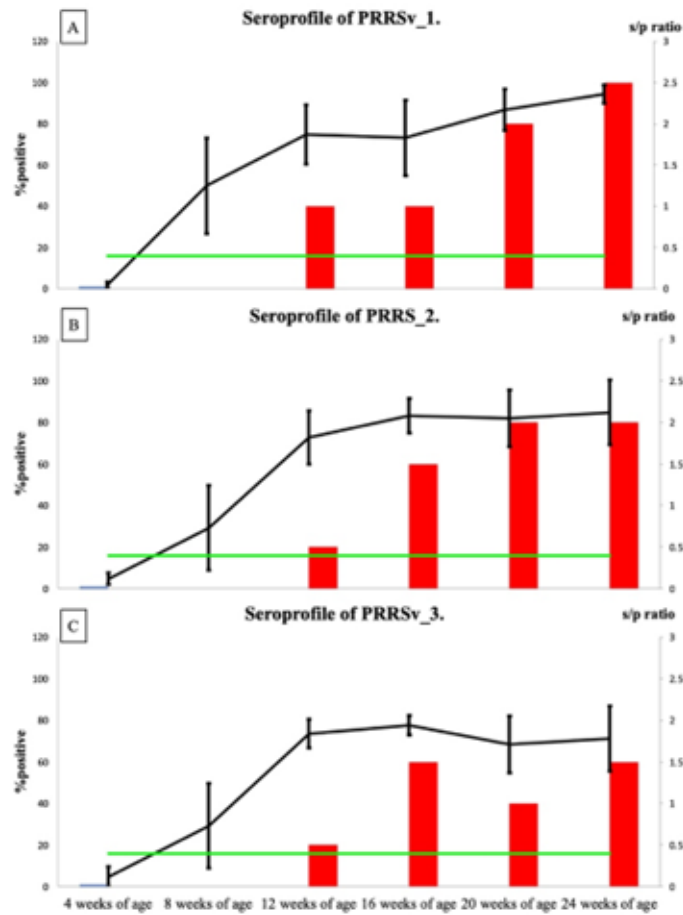


Figure 5 Seroprofile of PRRSv (A, B, C) from the ELISA of each replication group; graphs plotted as percent positive of s/p ratio as shown on the left y-axis and average s/p ratio of each age is shown on the right y-axis. The x-axis is representative of the age of the pig, whereas the green line represents the cut-off value of each disease in the ELISA test.

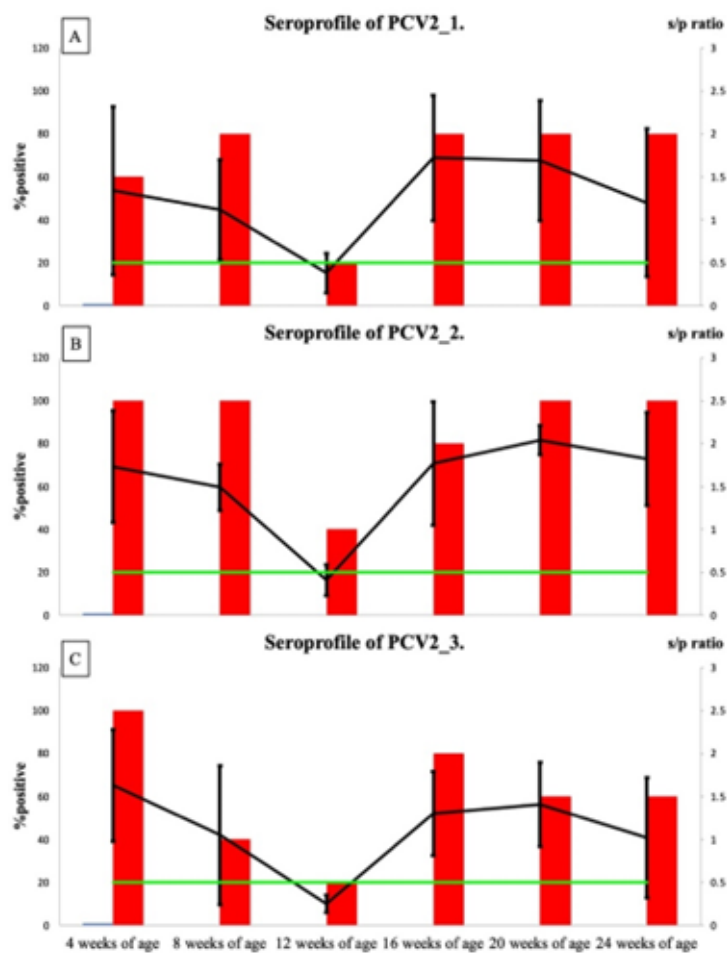


Figure 6 Seroprofile of PCV2 (A, B, C) from the ELISA of each replication group; graphs plotted as percent positive of s/p ratio as show on the left y-axis and average s/p ratio of each age is shown on the right y-axis. The x-axis is representative of the age of the pig, whereas the green line represents the cut-off value of each disease in the ELISA test.

***M.hyo* by ELISA, bacterial culture and PCR results**

According to the bacterial cultures (Table 2), we found that the indicative results of the *Streptococcus spp.* culture were detected at 12, 16 and 20 weeks of age in all replications, while results were detected at 8 and 24 weeks of age in two replications and detected at 4 weeks of age in only one replication, respectively. In terms of the results of the *Pasteurella spp.* culture, an indicative result was observed at 12 and 16 weeks of age in only one replication. With regard to the results of PRRSv by PCR, indicative results were detected at 12, 16, 20, and 24 weeks of age in all replications, while an indicative result was detected at 8 weeks of age in only one replication and was not detected at 4 weeks of age in all replications, respectively. According to the results of PCV2 by PCR, indicative results were detected at 18 and 24 weeks of age in all replications, while they were detected at 16 weeks of age in two replications and were detected in only one replication, respectively. However, they were not detected at 4 and 8 weeks of age in all replications. According to results of *M.hyo* by ELISA, the bacterial culture results and the results of the PCR of each replication were calculated and are presented in Table 2.

Table 3 presents a correlation of the coughing index with the results of respiratory disease detection performed in the laboratory using Spearman Rank Correlations and Kappa statistics, all of which were assessed according to the information presented in Table 2. It was found that both Spearman Rank Correlations and the Kappa values produced consistent results when comparisons were made between the coughing index with the respiratory disease detection results obtained from the laboratory. This was particularly relevant with regard to the ELISA result of *M.hyo*, as well as the PCR result of PRRSv and PCV2, which indicated high correlation coefficient and agreement values (closely 1) and a p-value of least than 0.05. However, these results were different from the results of the *Streptococcus spp.* and *Pasteurella spp.* cultures, which produced low correlation coefficient and agreement values (far from 1) and a p-value of more than 0.05.

Table 2 depicts the mode of the ELISA of *M.hyo* results, the mode of the bacterial culture results and the mode of PCR results for all replications in each age group (positive/the number of the replication group).

Age of pigs in all replications	Coughing index	ELISA of <i>M.hyo</i>	Bacterial culture (positive/sample)		PCR result (positive/sample)	
			<i>Streptococcus spp.</i>	<i>Pasteurella spp.</i>	PRRSv	PCV2
4 weeks of age	0/3	0/3	1/3	0/3	0/3	0/3
8 weeks of age	0/3	0/3	2/3	0/3	1/3	0/3
12 weeks of age	1/3	1/3	3/3	1/3	3/3	1/3
16 weeks of age	3/3	3/3	3/3	1/3	3/3	2/3
20 weeks of age	3/3	3/3	3/3	0/3	3/3	3/3
24 weeks of age	3/3	3/3	2/3	0/3	3/3	3/3

Table 3 Correlation of coughing index with respiratory disease detection results by Spearman Rank Correlations and Kappa Statistics.

Laboratory test	Spearman Rank Correlations		Kappa	
	Correlation coefficient; r	p-value	Agreement; a	p-value
ELISA result of <i>M.hyo</i>	0.78	<0.001	0.78	0.001
<i>Streptococcus spp.</i> culture	0.33	0.18	0.29	0.16
<i>Pasteurella spp.</i> culture	0.31	0.20	0.18	0.18
PCR result of PRRSv	0.69	0.001	0.64	0.003
PCR result of PCV2	0.77	<0.001	0.78	0.001

DISCUSSION

Classification code developed from sound from recorders

Python is a computer programming language that is often used to build websites and software, automate tasks and conduct data analysis (Jadhav et al., 2021). Our study used python to distinguish pig sounds from others sounds on the pig farms and then classify them. Accordingly, sound files were converted to images to allow researchers to further differentiate between all relevant sounds. A wave plot (Figure 1(A)) is often used to compare differences in sound characteristics (Boddapati et al., 2017); however, in our study, both a spectrogram and a log power spectrogram were also used (Figure 1(B, C)). Both represent a visual depiction of the scope of frequencies of a sign as it contrasts with time. This can serve as an effective strategy when spectrograms are employed as a stacked point of view on periodograms across a specific stretch of time for electronic signs (Dewi et al., 2019). This method can allow researchers to clearly

distinguish the pig coughing sounds from other sounds on farms (Figure 2, A-D). Pig coughing sounds (Figure 2, A) produced a steady rhythmic wave when compared with other sounds recorded on the farm such as machine sounds (Figure 2, B), the walking sounds of laborers (Figure 2, C) and other sounds generated by laborers (Figure 2, D). In a previous study, Chedad et al. (2001) reported that the wave plot can be employed to distinguish pig coughing sounds from metal clanging, grunting and other background noises. Shi et al. (2018) reported that audio-based cough detection systems are now increasingly being applied in clinical research. Furthermore, more sophisticated automated cough algorithms are now being developed with improved processing speeds so that audio-based cough monitors can advance the assessment of a patient's response to treatments. In conclusion, our developed python code can be used to classify pig coughing sounds from others sounds on pig farms.

Growth performance

The experiment involved three replications, of which each one was performed according to the same protocol. However, the initial of time was different in each replication (January, April and September). According to Table 1, initial weight and final weight in each replication group were not determined to be significantly different ($p>0.05$), wherein the final weight of Replication group 3 produced the best result when compared with the other replications. It was determined that the temperature recorded during the starter period for the pigs affected the growth performance in this case because all other factors were the same in all replications such as those of feed and management (Millet et al., 2005). Replication group 3 was initiated in September, while the duration of pig growth was from November to December. During that time period, the temperature was low as it was the winter season; however, that was the period with the highest growth rate. That is likely why Replication group 3 gave the best growth performance result. Alternatively, Replications 1 and 2 were initiated in January and April, respectively. Accordingly, the starter periods were in April and June, which occurred in the summer and rainy seasons, respectively. The season was found to have affected the growth performance of the pigs (Chmielowiec et al., 2012). However, the average growth performance of all replications was low when compared with the growth performance standard. This was likely due to the existing status of the disease and any applicable vaccination program, especially with regard to the *M.hyo* vaccine (Nathues et al., 2012).

Correlation of coughing index with seroprofile of diseases from the ELISA results, bacterial cultures results and PCR results

The coughing index (CI) established in our study was calculated from the number of coughing bouts, the number of examined pigs and the total time of observation as they relate to incidences of respiratory problems (Nathues et al., 2012). Figure 3 shows that a CI result was achieved after 8 weeks of age, while subsequent CI results were achieved at more than 2, 16, 18, 20 and 24 weeks of age. This would indicate that a respiratory problem had occurred in pigs at those ages. Pigs between 12 to 16 weeks of age were represented by a higher of slope (the sero-conversion), which was indicative of possible infections of *M.hyo*, PRRSv and PCV2, as well as secondary infections with

various other bacterial pathogens (e.g., *Pasteurella multocida* and *Streptococcus suis*) (Thacker, 2006). In our study, the results of CI correlated with the results of seroprofile of *M.hyo* that had been established by ELISA, PRRSv and PCV2 by the ELISA test (Figure 4, 5, 6: A-C). Accordingly, those diseases were found on this farm because positive results were clearly detected during the fattening period (12-24 weeks of age). Positive results of *M.hyo* and PRRSv were observed after 12 weeks of age but a positive result of PCV2 was observed after 16 weeks of age (Figures 4, 5, 6: A-C). Importantly, at 4 and 8 weeks of age, positive results may have been due to maternal immunity (Baekbo et al., 2012). Serology can be used to show the presence of diseases at the herd level; however, it is not suited for diagnosis among individual animals (Sørensen et al., 1997). Therefore, the monitoring of coughing is necessary to identify respiratory problems on pig farms. With regard to *M.hyo* monitoring, the results of CI were significantly correlated with the results of the serology test of *M.hyo* by ELISA, which revealed high correlation values ($r=0.78$, $a=0.78$, $p<0.05$) for both Spearman rank correlations and Kappa analysis (Table 3).

Bacterial cultures obtained from tonsil swabs were used to determine the presence of *Streptococcus spp.* and *Pasteurella spp.* because they are known to be the most common causes of coughing and respiratory problems among fattening pigs. Both of these pathogens had previously been identified as the cause of coughing and respiratory problems in pigs (Zimmerman et al., 2019). Our study found that *Streptococcus spp.* was detected in all ages of pigs; however, *Pasteurella spp.* was only detected in pigs at 12 and 16 weeks of age. In fact, *Streptococcus spp.* is a commensal type of bacterial infection, which is known to be a frequent early colonizer in the upper respiratory tract of pigs (Vötsch et al., 2018). Notably, *Pasteurella spp.* are opportunistic pathogens that can cause atrophic rhinitis and pneumonia in pigs (Bessone, 2019). Previous studies have reported that both bacteria can cause coughing, respiratory problem and secondary infections of porcine respiratory disease complex or PRDC (Thacker, 2006). According to the results of our study, both *Streptococcus spp.* and *Pasteurella spp.* were not observed to be correlated with coughing, as was determined by Spearman rank correlations and Kappa ($p>0.05$).

A PCR was used to determine the PRRSv and PCV2 infection status of pigs by examining serum samples (Table 2). Positive results were found in at least one replication from 8 weeks of age and 12 weeks of age for PRRSv and PCV2, respectively. The occurrence of a positive PCR result for both PRRSv and PCV2 in pigs would indicate the presence of an endemic disease on the farm. This outcome was in accordance with the previously recorded history on farms and with the outcomes of previous studies (Kittiwon et al., 2019). Consequently, it is not surprising that both positive PCR results of PRRSv and PCV2 correlated with incidences of coughing, wherein the correlation value was validated as $p<0.05$ for both Spearman rank correlations and Kappa analysis (Table 3). In previous studies conducted by Kristensen et al. (2004) and Kixmüller et al. (2008), it was reported that coughing is an important clinical sign of PRRSv and PCV2 infection. However, the time intervals of the coughs recorded in our study were too significant to correlate with the previous laboratory results. Accordingly, the qPCR method should be employed to detect the copy number of the PCV2 genetic materials in further studies.

As has been explained above, Spearman rank correlations and Kappa analysis were used to establish a correlation value between coughing and the other results of the laboratory tests (Table 3). We found that CI revealed significant correlations with the serologic ELISA results of *M.hyo* ($p < 0.05$) for both Spearman rank correlations and Kappa analysis. Moreover, a high correlation coefficient of the Spearman test ($r = 0.78$), and a high degree of agreement of Kappa test, were also observed ($a = 0.78$) when the results were compared with the standard (Landis & Koch, 1977). Accordingly, CI indicated significant correlations with the PCR results of PRRSv and PCV2 ($p < 0.05$), which revealed a high correlation coefficient of Spearman test ($r = 0.69$, $r = 0.77$, respectively) and a high degree of agreement of the Kappa test ($a = 0.64$, $a = 0.78$, respectively). Nonetheless, CI did not indicate significant correlations with cultures of *Streptococcus spp.* and *Pasteurella spp.* ($p > 0.05$), which exhibited low correlation coefficients of the Spearman test ($r = 0.33$, $r = 0.31$, respectively) and a low degree of agreement of the Kappa test ($a = 0.29$, $a = 0.18$, respectively). Consequently, our findings indicate a correlation of CI with the results of ELISA for *M.hyo*, as well as with the PCR results of PRRSv and PCV2, but these outcomes did not correlate with the cultures of *Streptococcus spp.* and *Pasteurella spp.* Therefore, veterinarians and/or swine practitioners should express concern about the potential emergence of those diseases when pig coughing is observed on the farm.

CONCLUSIONS

This paper outlines how we have employed the python language to build an audio deep learning recognition system for pig cough sounds. This system could be used to classify one sound from another on pig farms. Sound files obtained from a recorder were converted to images composed of a wave plot, a spectrogram and a log power spectrogram. The identified sounds could then be checked by a human to confirm their differentiation. Moreover, our study determined that the sound of pig coughing is an early abnormal sign of respiratory problems. The advanced indication of this type of respiratory problem would be preferred over the need to wait for results from laboratory tests. Consequently, the monitoring of coughing on pig farms would be well-suited for the detection of respiratory problems. This would be a much faster method of detection leading to even earlier diagnoses. Coughing can indicate to a producer that an illness may be present in the pig house and, therefore, would be well-suited for the monitoring of respiratory problems on pig farms. Coughing is not only an effective way to monitor the presence of a respiratory problem on pig farms, but it can also be correlated with certain respiratory diseases such as *M.hyo*, PRRSv and PCV2 infections. It was very surprising to us that coughing has not previously been correlated with *Streptococcus spp.* and *Pasteurella spp.* infections. Therefore, this paper endeavors to combine new technology in the detection of abnormalities in pig herds. We hope to promote the use of this system for the early detection of respiratory problems and illnesses on pig farms through syndromic surveillance.

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CONFLICT OF INTEREST

The authors declare that they have no conflicts of interest.

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