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Poultry health, bird sound classification, *Clostridium perfringens* type A, data mining, Artificial neural network.

An Intelligent Procedure for the Detection and Classification of Chickens Infected by *Clostridium Perfringens* Based on their Vocalization

ABSTRACT

In this study, an intelligent method was implemented for the detection and classification of chickens by infected *Clostridium perfringens* type A based on their vocalization. To this aim, the birds were first divided into two groups that were placed in separate cages with 15 chickens each. Chickens were inoculated with *Clostridium perfringens* type A on day 14. In order to ensure the absence of secondary diseases and their probable effect on bird vocalization, vaccines for common diseases were administered. During 30 days of the experiment, chicken vocalization was recorded every morning at 8 a.m. using a microphone and a data collection card under equal and controlled conditions. Sound signals were investigated in time domains, and 23 features were selected. Using Fisher Discriminate Analysis (FDA), five of the most important and effective features were chosen. Neural Network Pattern Recognition (NNPR) structure with one hidden layer was applied to detect signals and classifying healthy and unhealthy chickens. Firstly, this neural network was trained with 34 samples, after which eight samples were tested for accuracy. Classification accuracy was 66.6 and 100% for days 16 and 22; i.e., two and eight days after the disease, respectively. The results of this study demonstrated the usefulness and effectiveness of intelligent methods for diagnosing diseases in chickens.

INTRODUCTION

Today, some diseases are of particular importance due to the heavy economic losses they cause in poultry production. Necrotic enteritis is one of these diseases, which was discovered by Parish in 1961. After the antibiotic ban imposed since January 2006, the prevalence of this disease increased, with the annual losses of over \$ 2 billion. This disease is caused by *Clostridium perfringens* type A, which is a gram-positive, rod-shaped, spore-forming anaerobic bacterium. This disease causes atrophy of intestinal villi, decreased absorption of nutrients, and ultimately worse feed conversion ratio; this ratio is critical, because feed accounts for 60-70% of poultry production costs. This disease emerges when broilers are two to three weeks old and causes unspecific symptoms, such as reduced growth (Ficken, 1997), reluctance to move (Helmboldt, 1971), ruffled feathers (Van et al., 2004), anorexia (Bernan et al., 2003), and eventually death. The symptoms may not be evident until bird death, and necropsy findings include lesions in the jejunum and ileum (Ficken, 1997), which may also be present in the duodenum and cecum (Van et al., 2004). The small intestine is distended with gas, and the large intestine becomes thin and fragile. Considering the mentioned symptoms, necrotic enteritis is a silent disease, which often can only be diagnosed after the birds die, and this is one of the reasons for its late diagnosis and irreversible economic damage.



Today, non-destructive tests have a critical role in people's lives. These tests refer to the technology by which the health or its absence in target of a treatment is detected without any effects on its functional properties (Cherfaoui, 2012). These tests have been increasingly used all fields of animal production and agriculture. For example, detecting the ripening or quality of fruits is among the applications of non-destructive testing (Chen & Sun, 1991; Sun *et al.*, 2010). When non-destructive tests are used in live organisms, it will be possible to obtain information from those organisms which otherwise could only be extracted by destructive tests under normal circumstances. Inflammation is a silent disease (lack of accurate and reliable symptoms for disease confirmation) and thus requires a system for the timely diagnosis and treatment of the disease.

Vocalization provides useful information on their health status, species, and body size (Kasten *et al.*, 2010). Acoustic plays an important role in communication in many animals. Virtually all birds and animals are able to vocalize. Diseases of the respiratory system can directly interfere with acoustic generation and resonance, which are involved in sound production (Gerhard *et al.*, 2004). Bio-acoustics can help farmers to early detect respiratory disease by evaluating animal cough sounds through real time monitoring (Ferrari *et al.*, 2008). Animal vocalization can communicate different messages. For example, a call may be used to signal readiness to mate, to warn conspecifics of a predator, to keep in touch with other members of the group, or it may be an expression of pain or need. It has also been suggested that vocalization may be an expression or communication of an emotional state or reaction to an event, eliciting emotional states in others. Thus, analysis of vocalizations has been suggested as a non-invasive method for studying the emotional state of an animal (Emiear *et al.*, 2014). The analysis of vocalization, may one of the most reliable and least invasive methods of assessing distress in cattle. Alternatively, it can be used as an indication of stress, because it is relatively insensitive to low and moderate degrees of distress and it is only observed in any significant degree when the animal perceives itself to be in serious difficulty (Jon & Joseph, 1999).

Diagnosis means identifying a disease existing in a group of animals, which are maintained together. Therefore, by applying several methods like treatment, destruction of diseased birds, and vaccination of healthy birds, damage to other birds can be prevented.

Automation is the most conventional human response to activities that are repeated several times (Gaston & O'Neill, 2004). In this regard, analyzing

animal vocalization has been recently considered by researchers (Huang *et al.*, 2009). Signal detection means extracting sound from the noisy ambient during sound recording. According to the features of the extracted signal, sounds can be classified and related to the species (Skowronski & Harris, 2006).

Learning algorithms, such as linear discriminant analysis (Simmonds *et al.*, 1996), decision tree (Parsons & Jones, 2000), artificial neural networks (ANN) (Boddy *et al.*, 1994; Chesmore *et al.*, 2001), and support vector machines (SVM; Fagerlund, 2007), are the best choices for automatic detection of species, since they have very high accuracy compared with species classification by humans, which is both time-consuming and inaccurate (Acevedo *et al.*, 2009).

Chedad *et al.* (2001) tried to classify pigs based on their coughing sound. In that study, it was necessary to discriminate coughing sounds from other noises such as grunting or metal sounds, and therefore the researchers designed an algorithm based on probabilistic neural network with the accuracy of 91.9% (Chedad *et al.*, 2001).

Acevedo and Miguel (2009) automatically examined and classified three birds and nine frog species using SVM. In this study, animal sounds were classified according to minimum and maximum frequency, sound length, and maximum power. Then, three methods of linear discriminant analysis, support vector machine, and decision tree were used to classify the sound signals. Classification accuracy of the mentioned species in this study was obtained using linear discriminant analysis, decision trees, and support vector machine as 71, 89, and 95%, respectively. High accuracy of these three methods allowed sound-based identification of the species (Acevedo *et al.*, 2009).

Huang *et al.* (2009) designed an automated system for frog sound identification. Based on the results of this study, there seem to be certain frog species that can be easily recognized by their vocalization. Sound samples were first divided into syllables. Then, three features of spectral centroid, signal bandwidth, and threshold velocity were extracted from the parameters of k-nearest-neighborhood (KNN) and SVM classifiers, which accuracy was 89.0 and 90.30%, respectively (Huang *et al.*, 2009).

In the present study, an intelligent system was designed to detect and classify healthy and *C. perfringens* type A infected chickens. Firstly, the sound of both healthy and unhealthy chickens was recorded, and twenty-three features were extracted from the time domain signal. These features were then applied



to an artificial neural network for diagnosis and classification.

METHODOLOGY

This study was performed in February and March, 2014 at the Faculty of Agriculture of Tarbiat Modares University. Thirty 14-d-old Ross chickens were divided into two groups of 15 chickens each: the first group did not have any contact with the bacterium until the end of the study and were considered the healthy control samples. The second group was inoculated with the bacterium *C. perfringens* type A on day 14. Thus, 50% of the chickens were healthy, and 50% were unhealthy. The two groups were maintained in separate rooms. In order to prevent the emergence of other diseases, all birds were vaccinated against common diseases, using Newcastle-Bronchitis, infectious bursal disease, LaSota-Newcastle, and B1 Newcastle vaccines. Vaccination schedule is given in Table 1.

Table 1 – Vaccines and routes.

Type of injection	Type of vaccine
Spray	Newcastle-Bronchitis
Eye drop	B1 Newcastle
Eye drop	LaSota-Newcastle
Drinking water	Gambro

Birds were daily removed from their cages at 10 a.m. on days 16, 17, 18, 19, 20, 21, 22 and placed individually in boxes, which consisted of 50 cm³ cubes. Sound started to be recorded three mins after birds were placed in the boxes in order to allow birds to recover from handling stress. Sounds were captured using a microphone with the following specifications (Figure 1): 9.7×6.7 mm diameter, less than 2.2 KΩ impedance, 100~16kHz frequency, and -58 dB ± 3 dB sensitivity. Sounds were recorded in a computer and in “wav” format, at a frequency rate of 44000 Hz. Sounds were subsequently analyzed using the MATLAB 2013a software.

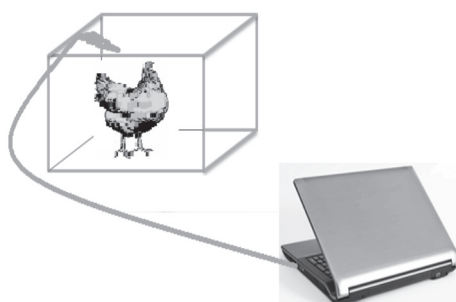


Figure 1 – Scheme of sound recording.

DATA MINING

Feature extraction

Since it is not possible to visually extract information from raw signals, for precise investigation it was necessary to extract the features from these signals that could be used as input for the neural network. Feature functions were numerical and had specific values; also, each one stated a characteristic or mode of a signal. Twenty-three features were extracted from the raw data in the present study (Table 2). These indices were the feature functions that have been used by other researchers in the field of data mining and can be used to obtain the information required for data mining (Khazaei *et al.*, 2013; Lei *et al.*, 2008). In this table, $x(n)$ is signal time-series and N is the number of data points.

Feature selection: Fisher discriminating analysis (FDA)

Selecting good features leads to time savings, reduced size of classifier's input data, less calculation, and enhanced accuracy of trouble-shooting. If there are few feature functions, characteristics of a particular signal cannot be adequately determined, and the classifier cannot distinguish between two different signals. Selecting a large number of feature functions also makes the classifier fail to make a distinction between two groups of extracted features. Thus, at the stage of dimensionality reduction using FDA, five of the most essential extracted features in the previous step were chosen. To use this method, data were first classified. In this process, features that differentiate two classes are selected so that there is a minimum difference between the members of one class and a maximum difference between the members of two classes (Chiang *et al.*, 2000; Diaf *et al.*, 2013; Sugiyama *et al.*, 2010).

In this study, five features with minimum difference within classes and maximum differences between classes were selected. FDA identified some features daily, out of which five were similar on all days. These five features included maximum signal value, standard deviation, root mean square, third central moment divided by the cube of the standard deviation, and crest factor.

Classification: Artificial Neural Network

The features selected in the data mining step should be used as the classifier input. There are numerous methods for the detection and classification of chickens



Table 2 – Primary features extracted.

Name	Formula	Name	Formula
Mean	$T1 = \frac{\sum_{n=1}^N x(n)}{N}$	Geometric mean	$T13 = \sqrt[N]{\prod_{n=1}^N x(n)}$
Maximum signal value	$T2 = \max x(n) $	Correlation coefficient	$T14 = \frac{T3}{T2} \times 100$
Standard deviation (stdv)	$T3 = \sqrt{\frac{\sum_{n=1}^N (x(n) - T1)^2}{N - 1}}$	The average deviation from the mean	$T15 = \frac{\sum_{n=1}^N (x(n) - T1)}{N}$
Quadratic mean square	$T4 = \left(\frac{\sum_{n=1}^N \sqrt{ x(n) }}{N} \right)^2$	Skewness	$T16 = \frac{\sum_{n=1}^N (x(n) - T1)^3}{T3^3}$
Root mean square	$T5 = \sqrt{\frac{\sum_{n=1}^N (x(n))^2}{N}}$	kurtosis	$T17 = \frac{\sum_{n=1}^N (x(n) - T1)^4}{(N - 1)(std)^4}$
Third central moment divided by the stdv	$T6 = \frac{T18}{T3^3}$	The third central moment	$T18 = \frac{1}{N} \sum_{i=1}^N (x_i - T2)^3$
Crest factor	$T7 = \frac{T2}{T5}$	The fourth central moment	$T19 = \frac{1}{N} \sum_{n=1}^N (x(n) - T2)^4$
Maximum signal value divided by the mean	$T8 = \frac{\frac{T2}{\left(\frac{\sum_{n=1}^N \sqrt{ x(n) }}{N} \right)^2}}$	The fifth central moment	$T20 = \frac{1}{N} \sum_{i=1}^N (x_i - T2)^5$
Root mean square divided by the mean	$T9 = \frac{T5}{T1}$	The sixth central moment	$T21 = \frac{1}{N} \sum_{n=1}^N (x(n) - T2)^6$
Impulse factor	$T10 = \frac{T2}{\frac{1}{N} \sum_{n=1}^N x(n) }$	The fourth central moment divided by the square of the variance	$T22 = \frac{T19}{T11^2}$
variance	$T11 = \frac{\sum_{n=1}^N (x(n) - T1)^2}{N - 1}$	The sum of squares	$T23 = \sum_{n=1}^N (x(n))^2$
Harmonic mean	$T12 = \frac{N}{\sum_{n=1}^N \frac{1}{x(n)}}$		

infected by *Clostridium perfringens*, some of which included neural networks (Boddy *et al.*, 1994), decision tree (Parsons & Jones, 2000), linear discriminant

analysis (Xanthopoulos *et al.*, 2013), and support vector machine (Neri, 2014). In this study, neural network pattern recognition (NNPR) was applied to detect signals



and classify healthy and unhealthy chickens. For this purpose, data were divided into three groups: the first category included 70% of data for network training, the second category included 15% for the validation of the network structure, and the third category included 15% for testing the neural network in terms of new data detection and classification. The feed-forward neural network structure was defined with three layers (input, hidden, and output) and tan-sigmoid activation function. In the input layer, one neuron was selected per feature. Since five features were chosen in data mining step, the input layer of neural network had five input neurons. The output layer had two classes and therefore consisted of two neurons. The number of neurons in the hidden layer strongly influences the performance of neural networks. Table 3 shows the performance of the neural network on days 16 to 22. On day 22, when the neural network presented maximum accuracy, four neurons were obtained in the hidden layer. The number of neurons in the hidden layer to obtain maximum accuracy was determined by trial-and-error. Table 4 presents the performance results of the neural network on day 22 when different numbers of neurons were used. In addition, the optimal number of neurons was similarly obtained on the other days. The number of neurons in the input and output layers was constant and results shown in Table 4 were obtained by changing the number of neurons in the hidden layer. Since the existence of four neurons in the hidden layer maximized the accuracy of neural network, the structure of the neural network on day 22 was determined as $5 \times 4 \times 2$.

Table 3 – Performance of the neural network.

Evaluation Day	Number of Hidden Neurons	Classification Accuracy	
		Testing Data	Training Data
16	7	66%	71.60%
17	9	77.34%	82%
18	7	80.42%	88.33%
19	8	80%	90%
20	5	90%	92.50%
21	4	95%	96%
22	4	100%	100%

Table 4 – Performance of neural network for day 22 with different numbers of neurons in the hidden layer.

Number of Hidden Neurons	Classification Accuracy	
	Testing Data	Training Data
2	91.20%	99%
3	97.1%	99%
4	100%	100%

RESULTS AND DISCUSSION

Obtained signals

In this section, two examples of sound signals from healthy and unhealthy chickens are shown. As given in Figure 2 and 3, the vocalization of healthy chickens presented higher intensity and uniformity in the shape of vocalization than the unhealthy ones. This was also reported in the literature (Engel *et al.*, 2000; McKay *et al.*, 2005).

These figures demonstrate that the vocalization of infected chickens presented lower sound intensity than healthy ones, but presented higher frequency within the main low frequency range. The mentioned differences in the feature extraction section were also confirmed.

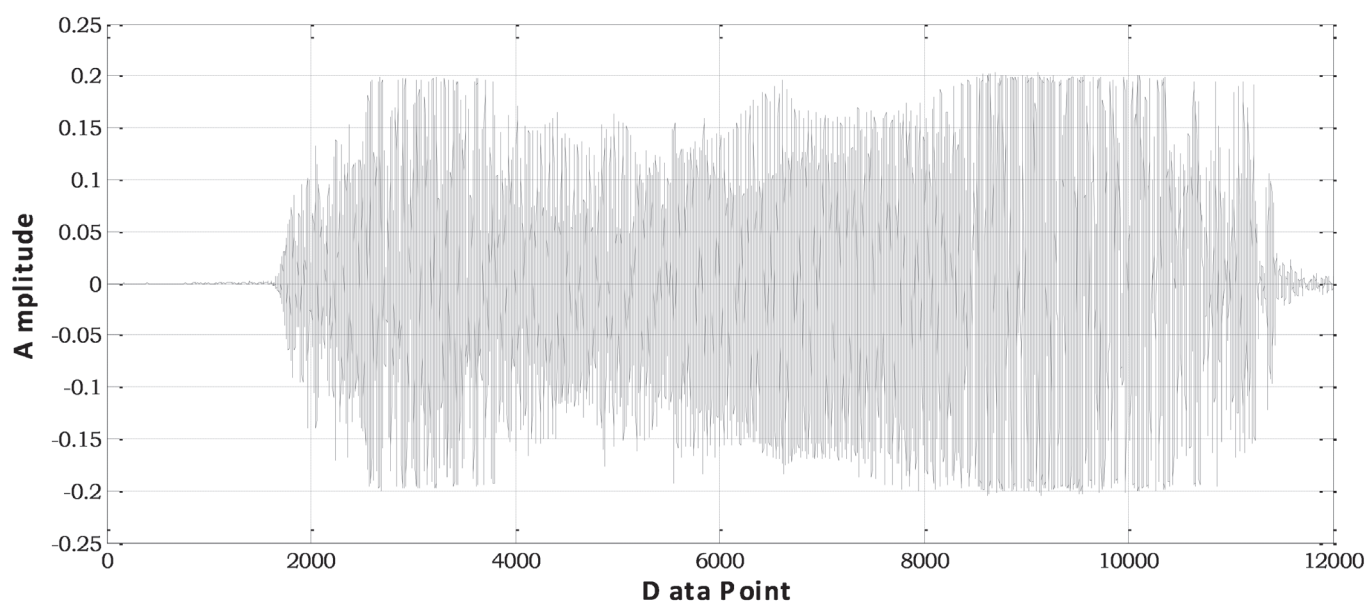


Figure 2 – Healthy chickens sound signals.

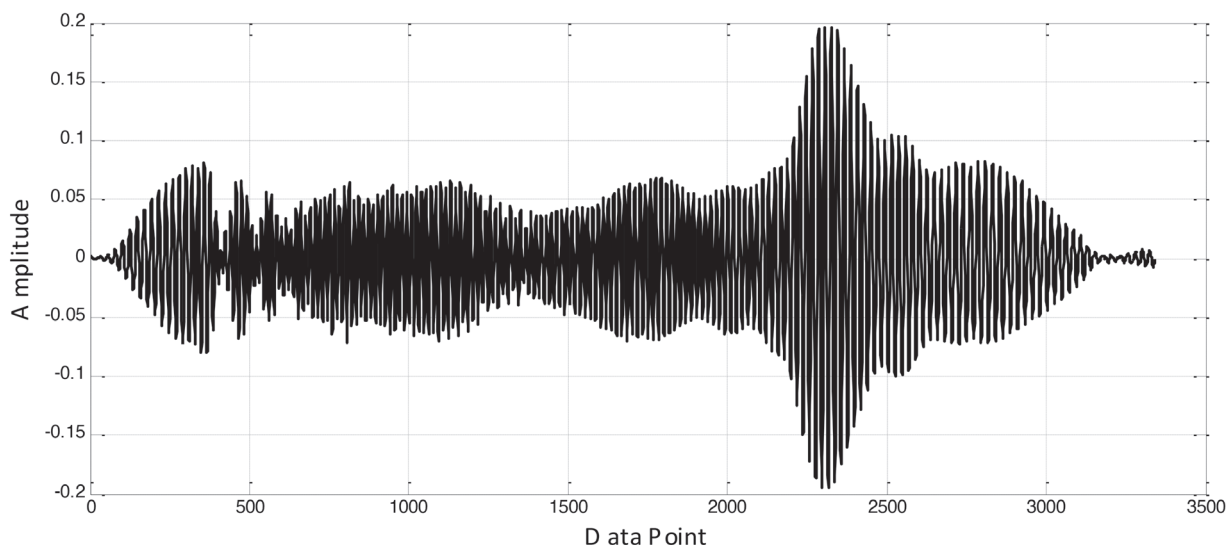


Figure 3 – Infected chickens sound signals.

Data mining results

Figure 5 shows the importance of selecting a good feature using FDA. Part a shows the signal value for a not-so-good feature. As can be seen in part a, there

was not much difference between the two classes. However, in part b, which shows a good feature, the distinction between two classes is evident.

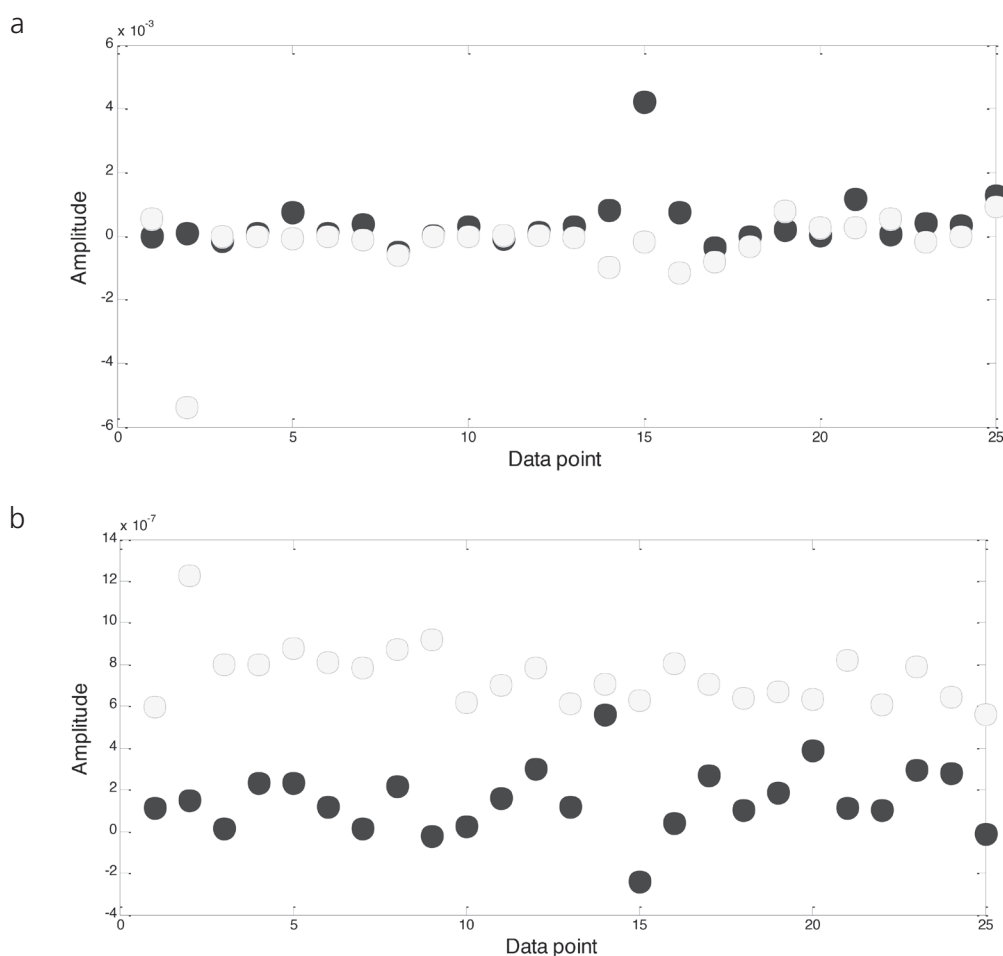


Figure 5 – The effect of FDA method on signal feature selection – a: worst features, b: best features.



Table 5 shows the mean values of some selected features of two healthy and unhealthy birds. Among the features mentioned in this table, maximum signal value and root mean square were the functions that detected the intensity and quantity of a signal. The obtained values demonstrated that maximum values and root mean squares of vocalization were higher in healthy than unhealthy chickens. These values indicate that vocalization intensity of healthy birds was higher than that of unhealthy ones. SD and crest factor showed signal uniformity. In fact, the higher the standard deviation and crest factor of a signal, the less uniform and wavier is the signal (Mckay *et al.*, 2005). These values determined that vocalization of the infected birds was less uniform and more dispersed than that of the healthy ones. Therefore, the results obtained in the feature extraction step confirmed the results obtained during the signal step. Indeed, these values clearly showed that the sound intensity of the infected birds was higher and less uniform than that of the healthy ones (Nowak *et al.*, 1997).

Table 5 – Mean values of the selected features.

Row	Feature	Healthy sample	Infected sample
1	Maximum signal value	+ 111%	-----
2	Root mean square	+ 107%	-----
3	Standard deviation	-----	+ 99%
4	Crest factor	-----	+ 98%

Classification accuracy of the detection of infected chickens

This study included 50 vocalization samples, out of which 34 were used to train the classifier (70% of data), eight to validate the classifier structure (15% of data), and finally eight ones for testing the classifier (15% of data). Maximum accuracy of the neural network was obtained for day 22. Thirty four samples were used to train the neural network. Out of these 34 samples, 15 were related to healthy birds sand 19 to infected ones. The confusion matrix of the neural network for data training shows that the neural network was able to differentiate vocalization samples of healthy chickens from those of unhealthy ones with 100% accuracy. Out of the eight samples used in the test, the neural network was able to detect five healthy and three unhealthy samples with 100% accuracy.

According to Table 4, the accuracy of the neural network was 66%, 77%, 80%, 80%, 90%, and 95% on days 16 to 21, respectively. Based on the detection of neural network, the disease appeared on days 17, 18, and 19, and reached its peak on days 20, 21, and 22. Thus, the neural network was able to detect and

classify unhealthy and healthy chickens on day 22 with 100% accuracy.

CONCLUSION

This study analyzed the vocalization of healthy and unhealthy broilers, and proposes an intelligent system for the diagnosis and classification of birds. Twenty-three features were extracted from the 50 vocalization samples and five were selected using the FDA method for data mining and dimensionality reduction step. These features were used as neural network inputs to determine its performance. Accuracy of the neural network during the training and testing phases was 100%. The results of this study demonstrate the usefulness and effectiveness of intelligent methods to diagnose poultry diseases, and therefore, allows for the early treatment of sick birds before the disease is further disseminated. For further application of this method, other common diseases should be evaluated.

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