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Boric acid adulteration detection in wheat flour using ATR-FTIR spectra and feed forward neural network

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Abstract

Adulteration in food commodities is common and thus easy, fast and reliable detection methods are essential to discourage it. Present study is focused on achieving this by using Attenuated Total Reflectance Fourier Transform Infrared Spectroscopy (ATR-FTIR) spectra collection along with Artificial Neural Network (ANN) classification of spectra for detection of boric acid added in wheat flour as adulterant. ATR-FTIR is non-destructive and fast method and gives signature spectra for many chemicals for easy identification and ANNs are very good at detection in pattern in data. It was found that combination of ATR-FTIR and ANN approach can detect more than 2% adulteration of boric acid in wheat flour successfully. The detection was on higher side and it was concluded that further studies using fast but effective sample preparation methods can improve detection levels by improving quality of signal.

Keywords: Adulteration, wheat flour, boric acid, ATR FTIR, neural network

Introduction

Adulteration in food commodities is on-going problem ¹. Boric acid is one such adulterant in wheat flour. It is added to wheat flour to extend its shelf life as it acts as antimicrobial agent and insecticide ². Boric acid is toxic in high doses and it may damages vital organs ³. Further boric acid is not approved as preservative in wheat flour in many countries including India ^{4, 5}. Thus boric detection in food products is required.

FTIR spectroscopic methods are non-destructive and rapid and thus lends well to easy adulteration detection. FTIR and chemometric based methods are already in use for adulteration detection in various products ¹. The molecular absorption and combination bands seen in the infrared spectroscopy typically give very complex spectra; it can be difficult to assign specific features to specific chemical components. Multivariate calibration techniques (e.g. principal components analysis, partial least squares, or artificial neural networks) are often employed infrared spectroscopy to extract the desired chemical information ^{1, 6}. Artificial neural networks are very good at pattern recognition and in general processing complex data which is difficult to understand by other methods ^{1, 7}. Thus in this study feed forward artificial networks along with principle component analysis was used as chemometric tools.

Materials and Methods

Sample Preparation

Pure wheat flour was obtained by grinding wheat grains of "MP Tukadi Sharbati" variety (obtained from local market) in flour mill. AR grade boric acid H_3BO_3 having 99.5% purity was obtained from Molychem, Mumbai, India. It was in fine dusty powdery form. Samples were prepared by weighing dry wheat flour and boric acid powders and then physically mixing them in beaker for 5 minutes to ensure homogeneous mixture. Pure boric acid powder and pure wheat flour samples were used as control samples.

Spectra Collection

Spectra was collected on Cray FTIR 640 spectrophotometer machine from Agilent Technologies in Attenuated Total Reflectance (ATR) mode. The reflectance crystal was of ZnSe (zinc selenium) and having single light reflection point and was of "Pike Miracle" brand. Beam splitter used in machine was made of KBR (potassium bromide).

Detector fitted in machine was "TE cooled DLaTGS" set at sensitivity value 4. Spectra collection speed was kept 5 kHz. For each sample presented to machine each spectra was taken 4 times and averaged by machine i.e. "number of scans" was set to value 4. Wave length range in which spectra collected was 6000 to 600 cm^{-1} . Interferogram sampling interval value was set to 2 and 1.28 kHz electronic low pass filter was kept active. Interferogram used was of symmetric double sided type. Processing of spectra was done by Resolution Pro Version 5 software and it applied ATR correction to spectra before putting it out in absorbance vs. wavelength from.

To collect spectra first background scan was performed without putting anything on ATR crystal. After that "wheat flour" or "boric acid" or "mix of both" as dry powder was kept on ATR crystal. The pressure arm was used to apply pressure on sample. Hatchet of the sample compartment was kept in open position during spectra collection process but was remained closed during background scan. Spectra was collected for various adulteration combinations as given in results and discussion section.

Adulterated samples were prepared having 8%, 6%, 4%, 2% and 1% boric acid in wheat flour along with 100% boric acid and 100% what flour used as control. For each sample 10 readings were taken by repeatedly applying sample and then removing it before applying same sample again. This resulted in total 70 spectra (5*10=50 for adulterated samples and 2*10=20 for control samples) which were used for training ANN. To test trained ANN slightly differing corresponding samples were prepared i.e. 7.5%, 5.5%, 3.5%, 1.5% and 0.5% boric acid was added to wheat flour along with 100% boric acid and 100% what flour as control samples. As each sample was applied 10 times this also resulted in 70 spectra and use to test ANN.

Artificial Neural Network Parameters

Neutral network used was of feed forward type fully connected network without bias inputs and with hidden layers. Number of neurons in input layer was equal to number of inputs used, hidden layer contained 32 more neurons than input layer and in output layer neurons were 2. Number of hidden layers was 1. Sigmoid function was used as output function. Training method used was stochastic gradient decent. All inputs were scaled up by dividing them with 0.002. Training algorithm was back propagation with

momentum value set at 0.9. Learning rate was varied according to data set. Learning epochs were based on target error i.e. learning was stopped when target error was achieved or sufficient epochs are done. Error function used was following.

$$E = \frac{1}{2} \sum_{k=1}^{k=n} (T_k - O_{o,k})^2$$

Where $k = 1$ to n indicate output neurons, T_k indicate target output on k^{th} neuron and O indicate actual output on k^{th} neurons in output layer. Target error for learning was set to 0.01 or 0.001 as per experimental need and if neurons shows less than 0.25 error on each individual neutron learning is considered successful. Trained networks was tested for correct identification of adulteration. The sample classification was considered incorrect when total average epoch error was more than 0.0625 which was based on above criteria as discussed in "results and discussion" section.

Statistical Analysis

Principle component analysis was done using R version 3.6.2 software available on www.R-project.org

Results and Discussion

FTIR spectra plot for 8% to 1% adulteration and control samples is given in Figure 1. Total 140 spectra are superimposed on each other to get clear comparative picture about results. In figure, one can see that FTIR spectra of boric acid is completely different compared to pure or adulterated wheat flour spectra. Though it is expected from observed differences in boric acid and wheat flour spectra that addition of boric acid should induces large changes in wheat flour spectra but this was not observed practically. Adulterated sample spectra remained more or less similar to pure wheat flour spectra though differences observed in some intervals. Spectral regions 700 to 800 cm^{-1} , 1380 to 1460 cm^{-1} and 3120 to 3200 cm^{-1} were particularly interesting as examination of plot shows that maximum differences appears in these regions. Thus this regions were selected for training and testing of ANN. The peaks at 3180 and 710 cm^{-1} can be attributed to OH groups in boric acid and 1420 cm^{-1} peak can be attributed to B-O bonds as suggested by literature [8].

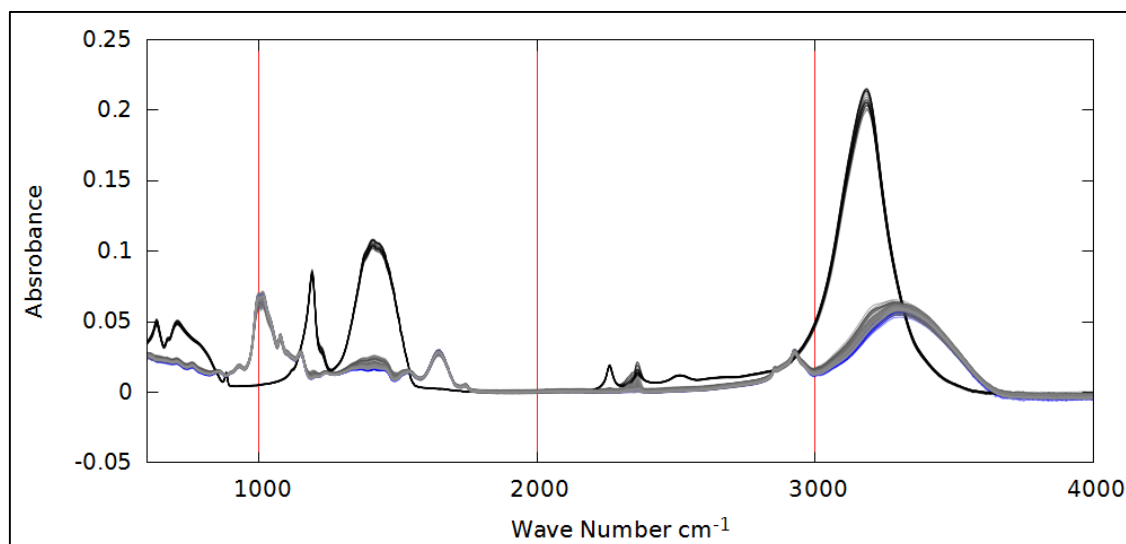


Fig 1: FTIR Spectra for 8% to 1% adulteration and control samples

From total spectra selected spectral regions (regions 700 to 800 cm^{-1} , 1380 to 1460 cm^{-1} and 3120 to 3200 cm^{-1}) are analyzed using Principle Component Analysis (PCA) technique. The results are shown in Figure 2. Note that PCA analysis involve only adulterated samples i.e. no pure boric

acid sample was used in PCA. The reason was distinct spectra of boric acid is already easily recognizable. Adding pure boric acid samples data hinders proper visualization of other samples in PCA graph.

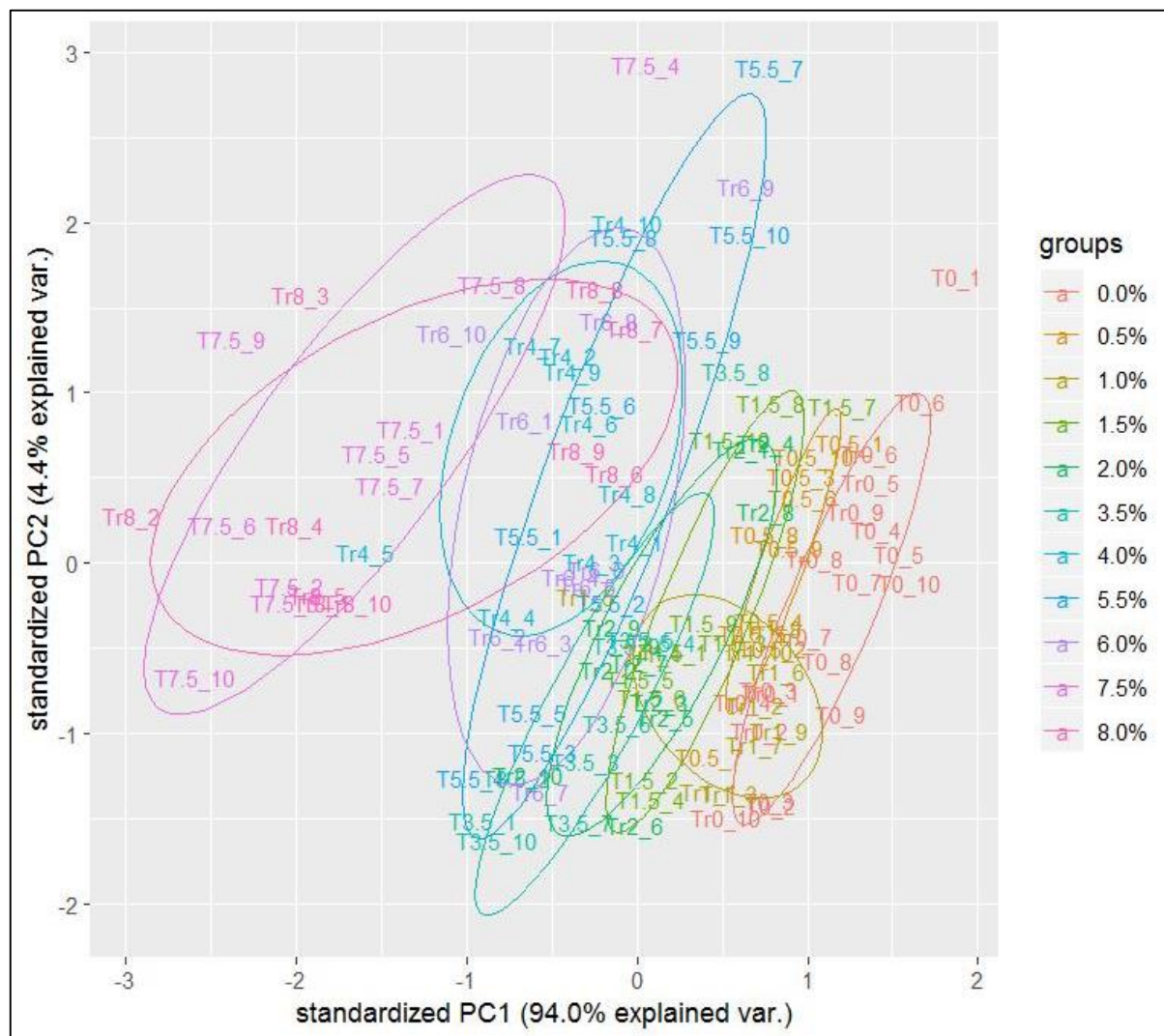


Fig 2: PCA plot

In Figure 2 "Tr" indicates sample is intended for training ANN while "T" indicate is for testing purpose. The figure immediately following "Tr" or "T" indicates level of adulteration of boric acid in wheat flour i.e. Tr5 means 5% boric acid was added in sample. The number after underscore indicate replication number i.e. Tr5_7 means 7th sample in Tr5 set.

It is clear from PCA analysis of data that some separation of adulterated samples is possible but separation is not very good particularly at lower levels of adulteration. This same result is confirmed when data was analyzed using artificial neural network. Use of PCA for data analysis along with chemometrics is well reported in literature [9].

Selection of hyper parameters of ANN is somewhat arbitrary choice as there exist no clear cut rules though some guidelines are suggested in literature [10]. Thus ANN some hyper parameters was chosen considering need of study i.e. inputs and outputs decided input and output layer neurons. Other hyper parameters can be considered empirical choices. Scaling of inputs was found necessary and helpful as otherwise all inputs were clustered around zero which gave

similar output when used as inputs of sigmoidal function making learning difficult. It was observed by authors that inputs varying between -4 to 4 in equally distributed way are ideal for sigmoid function output variation. Scaling in present study was step in this direction. Scaling of inputs was done by dividing inputs by 0.02 and taking results as inputs. Choice of 0.0625 as maximum allowed output error originates from criteria that maximum allowed error on each output neuron should not exceed 0.25. Considered two output neurons value of error function must be less than $\frac{1}{2}(0.25^2 + 0.25^2) = 0.25^2 = 0.0625$. At this stage maximum allowed error for training was 0.01 as this allowed training till error is fairly small.

It is clear from Figure 1 that all adulterated samples are very similar and boric acid is very distinct and thus boric acid spectra can be identified visually. For this reason boric acid samples were not used to train and test ANN.

Identification of wheat pure wheat flour vs. adulterated samples was tired. For this neural network was assigned to give output as given in Table 1. Maximum error for learning

was set to 0.01 and for testing was set to 0.0625. Learning rate ranging from 0.002 to 0.010 was tried and results are reported in Table 2.

Table 1: Neutral network output table

Output Neuron	Output Pure Sample	Output Adulterated Samples
First	1	0
Second	0	1

Table 2: Effect of changing learning rate on ANN

Learning Rate	Epochs to Reach Target Error	Train Errors %	Test Errors %
0.002	693	3.33	15.00
0.004	388	3.33	15.00
0.006	271	5.00	15.00
0.008	207	6.67	18.33
0.010	161	6.67	16.67

From this table it is evident at learning rate greater than 0.004 is not useful because after that, decrease in learning epochs occurs at expense of training ability. Thus 0.004 was chosen as learning rate for further examination. It must also be noted that higher learning rate also prevents over fitting but that can be done by limiting epochs. It is clear from the Table 2 that 388 epochs at 0.04 learning rate results in over fitting of data i.e. errors on train set are much lower than on test set. This indicates for limiting epochs.

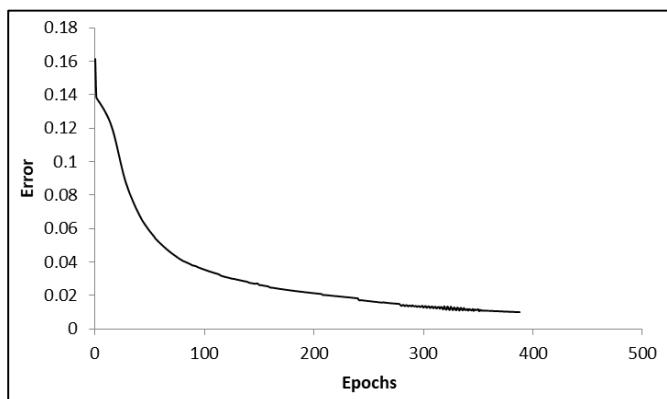


Fig 3: Error vs. epochs graph

Epochs can be decided from graph of epoch vs. error. The epoch vs. error graph for 0.004% learning rate is given in Figure 3. Smooth graph indicates that learning rate is not too much i.e. no too much overshooting and undershooting. It can be seen from the figure that learning is slows down after around 100 epochs. It was decided to test ANN using 500 to 300 epochs limit to see effect on over fitting.

Table 3: Effect of epoch limit on ANN

Epochs	Train Errors %	Test Errors %	Train Test Ratio
050	30.00	23.33	1.29
100	16.67	20.00	0.83
150	15.00	16.67	0.90
200	11.67	13.33	0.88
250	06.67	15.00	0.44
300	05.00	15.00	0.33

It can be concluded from Table 3 that around 150 epochs give equal train and test errors and over fitting of data can be

prevented successfully by limiting epochs to 150 epochs. Due to high error rate (15% in training set and 16.67% in testing) in 150 epoch training it was decided to train ANN to identify adulterated samples vs. control sample in one to one manner. For this adulterated sample data is compared against pure sample data one by one in separate manner i.e. "pure vs. 1% adulteration identification" and so on.

For one to one comparison maximum permissible error was reduced from 0.01 to 0.001 in order to increase number of training epochs and thus better training. Under this parameters ANN gave no training or testing errors till 4% level of adulteration. At 2% adulteration rate no training errors were reported but 15% test errors were reported after 1415 epochs, indicating need for limiting epochs to reduce possible over fitting of data. Same results were obtained at 1000, 500 epoch limits. At 250 epoch limit train error remained zero but test error shoot to 25%. At 100 epoch limit 40% test errors and 45% train errors were reported. From this results it was clear that ANN can be successfully employed to detect more than 2% adulteration only. At 2% adulteration level around 15% test samples fails even when training error is kept below 0.001 for all training samples. This was in harmony with PCA plot which indicate separation of samples at higher adulteration but at lower levels samples are mixed up.

Examination of literature reveals that use of ANN to detect adulteration with FTIR in wheat flour is not reported previously though many similar studies using various chemometric tools are reported [9, 11–13]. Thus finding of this study that "ANN can be employed to detect boric adulteration in wheat flour" is in harmony with reported literature.

Conclusion

Feed forward artificial neural networks can be successfully used to detect boric acid adulteration in wheat flour at more than 2% levels using FTIR spectra. It appears that detection level is on higher side and improvement scope remains on side of sample preparation changes rather than on tweaking ANN.

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