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Detection of Hygiene-Relevant Parameters from Cereal Grains such as Mycotoxin based on Microscopic Imaging, Intelligent Image Processing and Data Mining

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Abstract. We present our work on a novel method for the detection of hygiene-relevant parameters from grains of cereal crops based on microscopic image acquisition, image processing, and interpretation methods as well as data mining method. Hygiene-relevant parameters are for example Mycotoxins. We describe the data acquisition, the image analysis and interpretation method as well as the reasoning methods that can map the automatic acquired parameters of grain to the relevant hygiene parameters. We compare the results to the conventional findings with DON-values determination obtained by Elisa tests. The results show that with imaging methods and the new computer science methods it is possible to develop new measurement method and to produce new insights into the quality control of food stuff.

Keywords: Grain Hygiene-Relevant Parameters, Grain Product Quality, Mycotoxin Detection, Microscopic Image Analysis, Image Interpretation, Data Mining

1 Introduction

Fungal contamination of cereals is a serious economic problem throughout the world. Several fungi cause a reduction of grain quality, especially changes in color and taste [1], [2], [3]. However, the main risks of fungal damage arise from the production of toxic compounds, known as mycotoxins. Mycotoxins can cause serious adverse health effects. Toxigenic fungi that produce mycotoxins in grains of cereals or oil seeds belong to the genera Aspergillus, Alternaria, Fusarium and Penicillium. The control of this problem is therefore of particularly interest in food safety and quality control programs.

The aim of the research was the development of an automatic image acquisition and image interpretation system for the fast recognition and interpretation of cereal grains damaged by fungi. We have developed a data acquisition method that allows to take the coverage from the grain and allows to place it under a microscope for the acquisition of a digital image. This image has been used to automatically determine the number and the kind of fungi spores contained on the grain. For that we have develop suitable intelligent image analysis and interpretation methods. Based on the enumeration of fungal spore classes we developed a method that can map this information to the hygiene-relevant parameters.

The work we present here, show that the proposed methods based on microbiological preparation of the sample, microscopic image acquisition and intelligent image analysis are very suitable to capture the desired information and allow to recognize formerly unknown information that can be helpful to determine the quality of food staff. Data Mining was used to map the acquired image information to the class of fungi species and the desired grain quality parameters. We distinct among good grain samples, fungi damaged samples, and gall-mosquito damaged samples.

In this paper we describe the material used for our study in Section 2. In Section 3 is described the microbiological preparation of the grain sample and the microscopic image acquisition. The image analysis methods are explained in Section 4. The mapping of image information to hygiene relevant parameter with Data Mining is shown in Section 5. Result of the classification into grain quality is presented in Section 6. Finally, we give conclusions in Section 7.

2 Material

For this work we have been using different quality classes of wheat grains:

1. visual optical perfect grains from a charge where no fungal grains were included,
2. fungal damaged grains,
3. gall-mosquito damaged grains, and
4. visual optical perfect grains taken from a charge of fungal damaged grains.

In total we had ten samples from each class. Thirty single grains have been taken from each sample for further evaluation.

3 Microbiological Preparation of the Grains and Image Data Acquisition

One main problem was to take off the coverage of the grains such as fungi and make the coverage visible under the microscope and by doing that make it useable for further digital processing. Therefore, we have developed a procedure for taking off the coverage from grains and bring it onto a medium that can be placed under a microscope. From there can be acquired a digital image with the help of a digital camera connected with the microscope.

The method of choice was a water-based extraction method. The grains were placed into a boil together with stones. This water-filled boil was shacked for 2 minutes, then the water was filled into a centrifuge and the sediment was put on a slide. This slide was placed under the microscope and a digital image was taken. The resulting digital images are shown in Figure 1a-4a.

4 Intelligent Image Analysis and Interpretation

4.1 Image Analysis

The main aim of the image analysis was to recognize possible fungi spores and process them further for further determination of the type of fungi species. Here we used our novel case-based object recognition method [4] developed for recognizing biological objects with high variation. For the architecture of such a system see Figure 5. The case-based object recognition method uses cases that generalize the original contour of the objects and matches these cases against the contour of the actual objects in the image. During the matching a score is calculated that describes the goodness of the fit between the object and the case. Note the result of this process is not the information what type of fungi spore is contained in the image. The resulting information tells us only if it is highly likely that the considered object is a fungi spore or not. The result of the image analysis is shown in the images of Figure 1b-4b. One of the main problems of such a case-based object recognition method is to fill up the case base with a sufficient large enough number of cases. We used our procedure described in [7] for that. For the study we have 10 different cases. The recognition quality is good enough to find most of the fungi objects as it can be seen in Figure 1b-4b.

In the next step we must classify the objects into fungi spore and what kind of fungi specie it is. Afterwards we must count the number of fungi spores for each fungi specie.

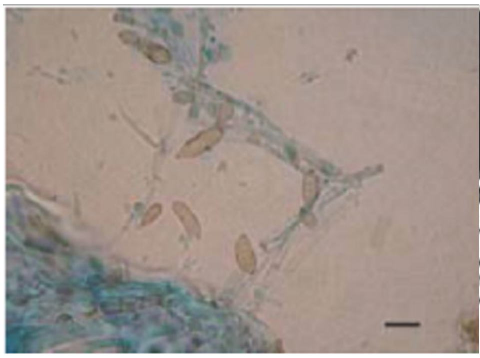


Fig. 1a. Coverage of Grains with Ulocladium

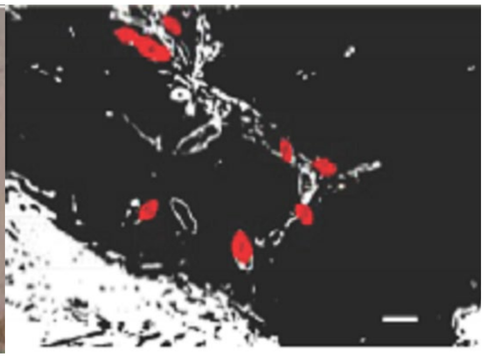


Fig. 1b. Segmented Image

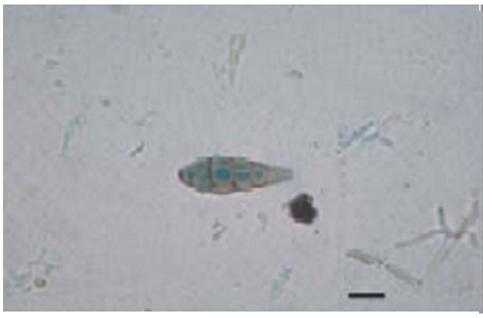


Fig. 2a. Coverage with Grains Alternaria Alternata

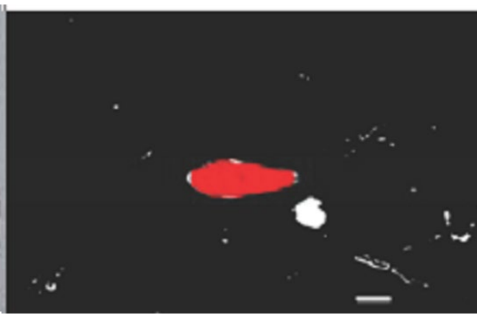


Fig. 2b. Segmented Image

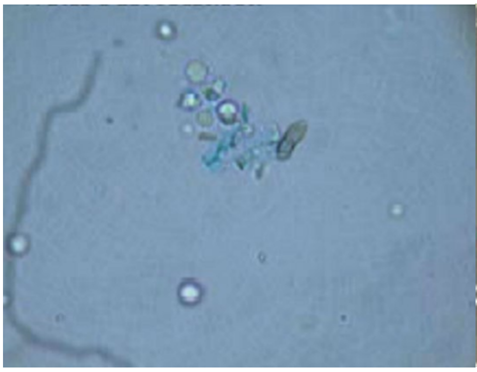


Fig. 3a. Coverage with Grains with Ulocladium

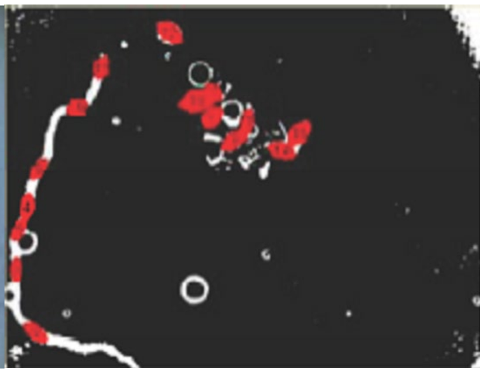


Fig. 3b. Segmented Image

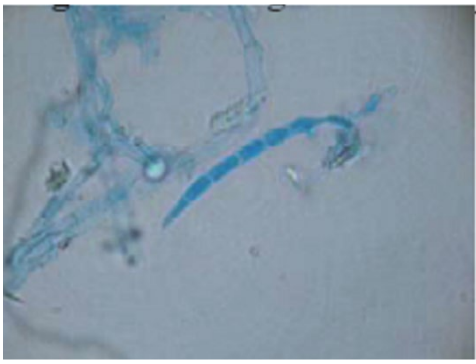


Fig. 4a. Coverage of Grains with Fusarium

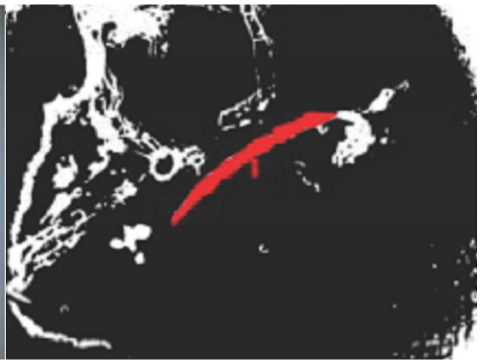


Fig. 4b. Segmented Image

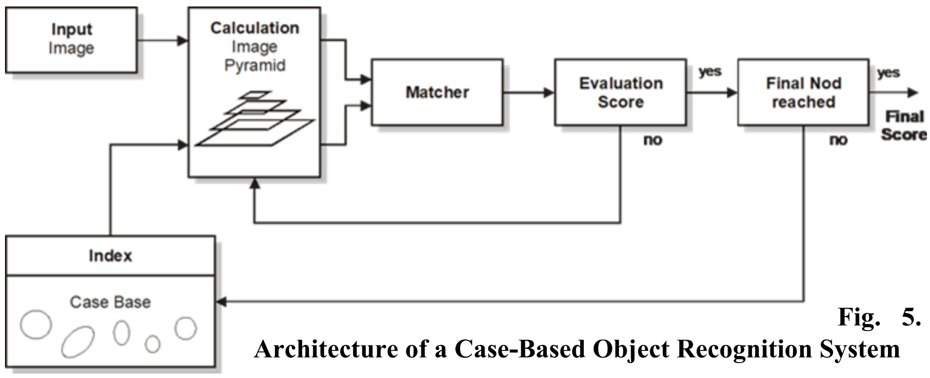


Fig. 5. Architecture of a Case-Based Object Recognition System

4.2 Image Interpretation and Data Mining

After the methods have been recognized potential objects that are highly likely fungi spores, we must extract features from the objects that distinguishes the object from background and different fungi species. Of course, one features is already the shape information used in the matching process but that is not enough for more detailed recognition. The features that must be calculated for this kind of objects is the inner structure, texture [8], and gray level information. The automatic feature extraction procedure extracts the features from the objects. These features describe the fungi spores. They are used for classification of the objects into different kind of fungi species.

We use decision tree induction [5] based on our tool Decision Master [6]. It allows easy learning of the classifier and the classification accuracy. We obtained an accuracy of 82.9%.

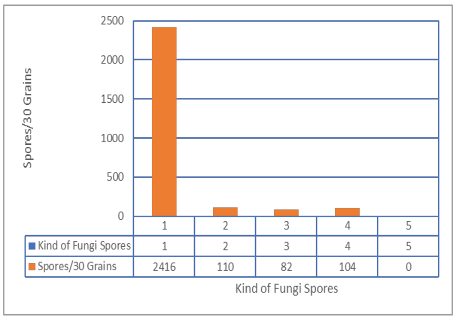
As the result we will get the information about the kind of fungi spores contained in the image and the number of fungi spores of different kind of fungi species (or classes).

5 Mapping of Image Information to Hygiene Relevant Parameter with Data Mining

The developed methods in Section 3-4 bring out information about the fungi species and the frequency of spores of a fungi specie. We will show now that this information can be used to control the quality of food stuff. We took into consideration the number of fungi spores of Fusarium (1), Alternaria/Ulocladium (2), Aspergillus/Penicillium (3), Cladosporium (4), and the number of fungi spores with unknown classification (5).

From the 4x10 different samples were created a data base where the column of each entry shows the fungi specie, the number of Fusarium spores, the number of Alternaria/Ulocladium, the number of Aspergillus/Penicillium, the number of Cladosporium, the number of fungi spores with unknown class and the total number of fungi spores.

Besides that, for each sample was determined the DON-value based on an ELISA test (see Figure 6). The concentration of a main mycotoxin producer of the genus Fusarium was determined by a commercial enzyme immunoassay screening (deoxynivalenol (DON)). To make sure that the kind and the number of fungi spores corresponds to the grain quality label.



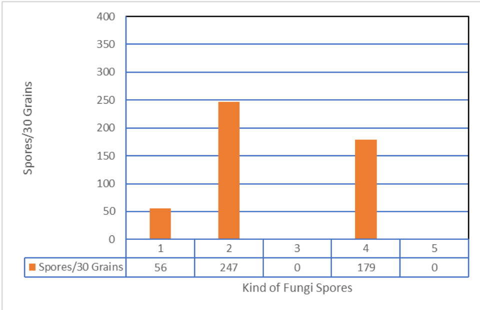
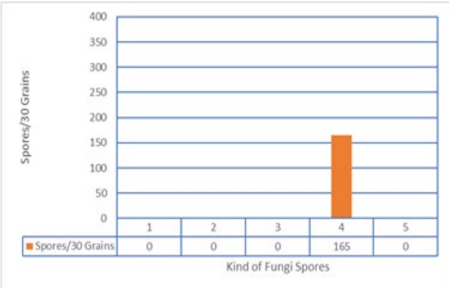


Fig. 6. Fusarium damaged Grain Sample **Fig. 7.** Gall-Mosquito damaged Grain Sample



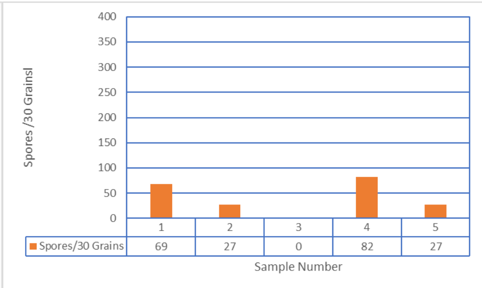


Fig. 8. Good Sample 1 **Fig. 9.** Good Sample 2

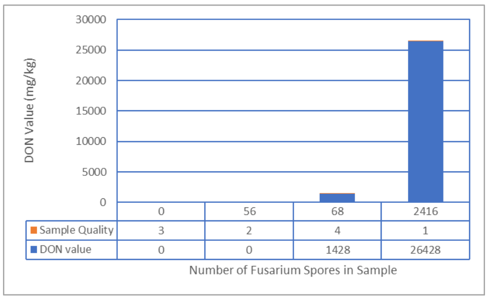


Fig. 10. DON Value versus Fusarium Spore Number for the four different Samples

Figure 6-9 show that there is a significant difference in the number and the kind of fungi species for the different charges Fusarium damaged grains (1), Gall Mosquito damaged grains (2), good sample 1 (3), and good sample 2 (4). Figure 6 shows that

DON value corresponds to the determined class labels. Grain with small number of Fusarium spores have low DON-value and grain charges with high number of Fusarium spores have high DON-value.

Decision tree induction based on an entropy-based criteria was carried out to find out the relation between the coverage of fungi spores in a sample and the class label of the quality of the grain samples (1,2,3,4) as named in Figure 6 to 9. We used our tool with Decision Master [Perner, 2003] for the decision tree induction experiment.

6 Result

The result of the induction experiment shows that there is a relation between the number of Cladosporium spores and Fusarium spores respective the class, see Figure 11. It says that grain charges with a high number of Cladosporium spores will have small number of Fusarium spores. The rule says a charge with "Cladosporium spores greater 125 AND Fusarium spores less or equal 28 items infer a good grain sample_1". The other rule says a charge with "Cladosporium spores greater 125 AND Fusarium spores greater 28 infer in Gall-Mosquito damaged sample".

Whereas samples with low Cladosporium spores can be either charges fungi damaged samples or good grain samples_2. The rules are:

> If Cladosporium less or equal 125 AND Fusarium less or equal 1236 THEN good sample_2.
> If Cladosporium less or equal 125 AND Fusarium greater 1236 THEN Fusarium damaged sample.

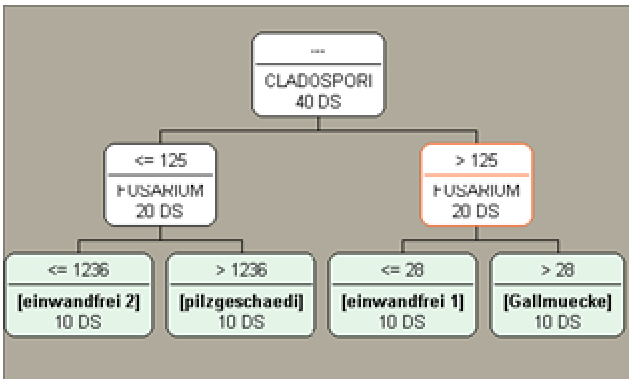


Fig. 11. Decision Tree for the Determination of Grain Quality based on the Number and the Type of Fungi Spores

It seems, that the number of Cladosporium spores indicate how many Fusarium spores are present.

The number of Alternaria and Aspergillus spores did not have a considerable influence for the hygiene-relevant parameters.

7 Conclusions

We have presented our results for the detection of hygiene-relevant parameters from cereal grains based on a microbiological method for sample preparation, microscopic image acquisition, and intelligent image analysis and interpretation methods. With these methods we were able to identify objects in the image that were highly likely fungi spores. After classification based on a classifier that used the extracted image features from the objects we could determine if the object is a fungi spore and what kind of fungi species it is. The classifier has built up by decision tree induction based on our tool *Decision Master*.

After a further evaluation with the number and kind of fungi spores in a sample we can decide about the quality of the grain sample. We used four classes "good grain sample_1", "good grain sample_2", "gall-mosquito damaged grain sample", and "fusarium damage grain sample". The classifier for that classification has also built up by decision tree induction based on our tool *Decision Master.*

It shows that decision tree induction is a flexible tool for building classifiers in an easy and quick way. You can also think about more classes for the grain quality as more as the influence and the frequency of fungi spores in grain samples were understood by using the developed techniques. However, more grain samples should be collected for the decision tree method to get good accuracy of the classifier.

The image acquisition method we have demonstrated in this paper works well and can be fully automated. It can also be constructed in such a way that the coverage from each single grain can be taken off and evaluated based on the intelligent image interpretation and data mining methods. The image analysis on case-based object recognition works well for this task but must be tuned so that a better object recognition rate can be achieved. From each single object can be extracted image features and these features can be used for classification. It is preferable to construct the classifier based on decision tree induction methods. Once the type and number of fungi spores has been determined this information can be set into relation with the hygiene-relevant parameters. We have shown that the number of Fusarium spores correlate with the DON-levels which is a value used for the determination of the mycotoxin concentration. However, when considering this experiment as a data mining experiment and applying decision tree induction to the created data base some other valuable information can be extracted which are hidden before. The next steps of our work will be to improve the image interpretation methods. With a fully automatic procedure we can apply our method to considerable number of grains sample. Formerly unknown relations or information based on the material in the coverage of the grain such as several types of fungi species can be discovered by our method.

The aim of our work was to develop a new measurement method for the determination of hygiene-relevant parameters on grains.

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