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MACHINE VISION DETECTION OF TETRAZOLIUM STAINING IN CORN

by

Wei Xie and Marvin R. Paulsen
Graduate Assistant and Professor
Agricultural Engineering Department
University of Illinois at Urbana-Champaign
Urbana, IL. USA

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Summary:
A machine vision algorithm was developed to detect and successfully quantify tetrazolium staining in sectioned corn kernels. The tetrazolium-machine vision algorithm was used to predict heat damage in corn due to drying air temperature and initial moisture content. Corn harvested at 20% and 25% moisture was negatively affected by drying at 70°C. Corn harvest at 30% moisture was negatively affected by heat at all drying temperatures above 25°C, and was much more severely affected as drying temperature increased.

Keywords:
Corn, machine vision, germination, inspection.

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ABSTRACT

A machine vision algorithm was developed to detect and successfully quantify tetrazolium staining in sectioned corn kernels. The tetrazolium-machine vision algorithm was used to predict heat damage in corn due to drying air temperature and initial moisture content.

The machine vision tetrazolium test was able to predict viability loss and therefore detrimental effects of heat on corn to be used for wet milling. Corn harvested at 20% and 25% moisture was negatively affected by drying at 70°C. Corn harvest at 30% moisture was negatively affected by heat at all drying temperatures above 25°C, and was much more severely affected as drying temperature increased.

INTRODUCTION

Corn is one of the most important grain crops and needs considerable care to preserve its quality (Aldrich and Leng, 1965). Viability is an important index of corn seed quality. It indicates the probability of future seedling growth and has a direct relationship to production. Farmers need seeds with a high viability for planting. Thus, it is necessary to test the viability of seeds before planting. The traditional method was to take a samples of seeds, plant it, then observe growth. This method requires a minimum of 5 days and temperature and relative humidity must be controlled to provide the corn sample with favorable conditions (AOSA, 1988). Furthermore, the work is tedious and is mostly all manual. It is desirable to find an easier way to do viability tests if accuracy can be maintained.

Grain drying is necessary to reduce the possibility of the corn spoilage after harvest. Unfortunately the heat from the drying process also reduces the viability of corn seeds (Baker et al, 1991). Research was done to find optimum drying conditions to quickly dry corn before fungal growth starts and to preserve high viability. A method for providing close inspection of seed viability is required. Thus, an effective and quick method for testing corn viability would also be helpful for maintaining high quality grain after grain drying processes.

OBJECTIVES

The objectives of this study were:

a) to develop a tetrazolium testing procedure for corn that best accommodates automated

1. Acknowledgement: This Study was a part of Project No. 28230 of the Illinois Agricultural Experiment Station, College of ACES, University of Illinois at Urbana-Champaign.

evaluation using machine vision;

b) to develop and implement a machine vision algorithm for corn viability classification based on the tetrazolium test; and

c) to study the relationship between initial corn moisture and drying air temperature on the loss of viability using machine vision classification of tetrazolium-stained corn kernels.

LITERATURE REVIEW

In 1941, it was discovered that tetrazolium(TZ) salts were reduced from colorless forms to insoluble, colored formazans in living tissue (Delouche et al., 1962). Lakon (1942) initiated research on rapid viability testing with tetrazolium. He found that when tetrazolium salts came in contact with living tissue in seed embryos, the chemical was reduced to an insoluble pigment and stained the tissue. Non-living tissue did not effect reduction of the tetrazolium salts (Delouche et al., 1962). Of several tetrazolium salts, Lakon found that 2, 3, 5-triphenyl tetrazolium chloride was the most suitable (Delouche et al., 1962). The level of viability of corn can be judged by the stained area of the kernel section, color saturation, and location of the stained areas.

Tetrazolium salt is an oxidation-reduction indicator and it has been well established that the development of a non-diffusible red color in tissue is the result of reduction of the chemical by enzymatic action. One or more of the dehydrogenase systems appear to be involved in the reaction.

Howarth and Stanwood (1992) developed a machine vision system to interpret TZ test results. The basic procedure was to grab a RGB image of the TZ stained corn seed section, segment the whole kernel out of the image in the red plane, and calculate the area of the whole kernel A_w. Then, the algorithm subtracted the green plane from the red plane, scaled the subtracted image between 0 and 255, thresholded the image by 128, and calculated the area of the thresholded image, A_t, which represented the stained area. Then, the algorithm computed the ratio between the stained area and the area of the whole kernel:

$$R = A_t/A_w$$

The Howarth and Stanwood (1992) algorithm was trained with corn seeds of known viability. The seeds were either viable or non-viable. It obtained a R for each corn seed. It computed a classifier R_C with least error by assuming that the probability density function for each class was normally distributed. Each corn seed was classified as viable if $R > R_C$ and classified as non-viable otherwise.

Digital Color Image Acquisition

Image acquisition was a process of sensing and converting the electromagnetic energy spectrum into digital form (Gonzales and Woods., 1993). The electromagnetic energy spectrum was reflected from the object of interest. The digital image was two-dimensional and discrete. The value at each pixel in two-dimensional coordinate space represented the electromagnetic energy level. One number was able to represent the gray level for a black and white image while at least three numbers were needed to represent a color image (Gonzales and Woods., 1993). A RGB color model was usually used with a digital color camera. Howarth and Stanwood (1992) used a Cohu RGB camera to acquire an image of TZ stained corn seed sections into a frame buffer of 512 x 480 pixels. The frame buffer was inside an ATVista 4-MEG color board. The spacial resolution was approximately 0.015 mm/pixel (Howarth and Stanwood, 1992).

Image Enhancement

The original images were usually processed to become suitable for a specific application. The first few operations of TZ test interpretation were done to segment the whole kernel from the background as an object and to segment the stained parts from the whole kernel as an object (Howarth and Stanwood, 1992). The segmentation was based on the distinguished gray level range of the background, the whole kernel, and the stained parts. Noises involved with image acquisition caused sparse pixels in the original image to be random gray levels. These pixels randomly appeared inside the image. They were likely to be segmented from their neighbor pixels and to be mistaken as objects. Under such a situation, the appropriate image enhancement was noise reduction for TZ test interpretation.

Image Segmentation

Image segmentation is the first step in image analysis. Segmentation subdivides an image into its constituent parts or objects. The level to which this subdivision is carried depends on the problem being solved. The image is usually subdivided until the objects of interest are isolated (Gonzales and Woods, 1993). There are generally two approaches for segmentation algorithms. One is based on the discontinuity of gray-level values and the other is based on the similarity of gray-level values. The first approach is to partition an image based on abrupt changes in gray levels. The second approach is thresholding, region growing, and region splitting and merging.

Thresholding is one of the most important approaches to image segmentation. A digital image can be expressed by a two-dimensional function f(x, y), where (x, y) are the coordinates of each pixel in the image and f(x, y) is the gray-level value. A well designed image acquisition system acquires images in which object and background pixels have gray levels grouped into two dominant modes. The thresholding approach is to select a threshold, T, that separates the two modes.

Gonzales and Woods (1993) defined a thresholded image g(x, y) as:

$$g(x,y) = \begin{cases} 1 & \text{if } f(x,y) > T \\ 0 & \text{if } f(x,y) < T. \end{cases}$$

Thus, pixels labeled 1 (or any other convenient intensity level) correspond to objects, whereas pixels labeled 0 correspond to the background. When T depends only on f(x, y), the threshold is called global. If T depends on both f(x, y) and p(x, y) the threshold is called local. If, in addition, T depends on the spatial coordinates x and y, the threshold is called dynamic.

For example. Liao et al. (1994) developed a real-time machine vision system to detect color and surface defects of maize kernels. The machine vision system took images of several corn kernels at each time. The image contained separate corn kernels and a dark blue background. The objects of interests were individual corn kernels since the final products of the processing were the quality-related features. Each corn kernel was isolated from background and other corn kernels inside the same image during the segmentation process (Liao et al., 1994). The segmentation process was global thresholding. The histogram of the whole image was generated. The thresholding value was generated according to the result of the histogram analysis and was constant for the same imaging condition and application (Liao et al., 1994).

Representation

The result of segmentation is usually a binary image. A binary image contains only two types of pixels, pixels of either 0 or 1 gray-level value. Objects of interest are isolated out in pixel aggregations in gray-level 1, which are still raw data needing a suitable description for further computer processing. The process for finding representative characteristics of an object and expressing them with numbers, which are able to be further processed by a computer, is called representation (Duda and Hart, 1973). The numbers expressing the characteristics of an object are called the features of the object. The techniques to find representations of the objects of interest are referred to as feature extractions.

Convex Hull

Convex hull of a area is an important feature of the area. It was usually used to describe the whole region that bounds the area. Gonzales and Woods (1993) defined convex hull of an arbitrary set S as the smallest convex set containing S. For a section inside an image, all of the section pixels comprise a pixel set. Thus, its convex hull is the smallest convex area containing all of the section pixels.

MATERIAL AND METHODS

Tetrazolium Staining Procedure

Tetrazolium staining procedures for corn consisted of preconditioning the sample, preparing the 2, 3, 5-triphenyl tetrazolium chloride (TTC) solution, and staining. The choice of an appropriate procedure ultimately depended on the speed, accuracy, and for the convenience of ensuing processes. The Association of Official Seed Analysts (AOSA, 1970) stated that the purpose of preconditioning seeds was to make the seed tissue less fragile and to start the germination processes. The general preconditioning method was to either place samples on top of or between moist blotters or paper towels overnight or to place them in water for 3-4 hours at a temperature of 30°C. Howarth and Stanwood (1992) preconditioned corn seed samples by placing them for 10 hours in a plastic container between moist blotters. Submerging the samples in water requires less time, however, soaking will often cause dry brittle seeds to fracture (AOSA, 1970). Also, this method can easily saturate the corn seeds making the absorption of TZ difficult.

Delouche et al., (1962) reported that embryos of corn seeds need to be sectioned to be exposed to tetrazolium solution since their waxy pericarps are not readily permeable to tetrazolium solution. The usual methods were to section or pierce the corn seeds (AOSA, 1970). Howarth and Stanwood (1992) bisected each seed longitudinally using a razor blade. After cutting, only one half of each seed was used to avoid repetition of results on the same seed.

Triphenyl tetrazolium chloride was the most suitable tetrazolium salt for the TZ test (Delouche et al., 1962). It is a white powder. TZ solution was developed with TTC powder and distilled water. The Association of Official Seed Analysts (AOSA, 1970) recommended TTC solutions should have a concentration of 0.1% to 0.5%. Howarth and Stanwood (1992) used a 0.1% solution while Delouche et al., (1962) specified a 0.5% solution for corn seeds.

Half of each corn seed was placed into the TZ solution while the other half was discarded. Approximately 30 to 60 minutes were needed to stain corn seeds satisfactorily at 40°C, but at room temperature it takes longer (Delouche et al., 1962). In general, staining occurs twice as fast at 30° C than at 20°C, and two times faster at 40°C than at 30°C. However, temperatures higher than 45°C were not allowed (Delouche et al., 1962).

Corn seeds were placed on four sheets of wet paper towels. Six sheets of wet paper towels were used to cover the sample. The moistening process took 6 hours. Then, corn seeds were sectioned and ready for staining. Solutions of 0.1%, 0.2%, and 0.5% TZ were tried, but 0.1% TZ solution was chosen since it gave better segmentation between the stained and unstained areas. The TZ solution was removed after the corn seed section was stained to the desired intensity. The duration of staining was 3 hours. Then, the sectioned and TZ stained corn seeds were washed several times with water, for about 50 seconds.

Tetrazolium Test Interpretation

Living tissue inside an embryo stains bright red. Each stained corn seed was interpreted as as highly viable to non-viable based on how large of an area was stained bright red and the location of the stained areas. (AOSA, 1970). Some areas of the embryo, such as the radicle, seminal root, and plumule, were more important to be stained than other areas, such as the scutellum (Delouche et al. 1962).

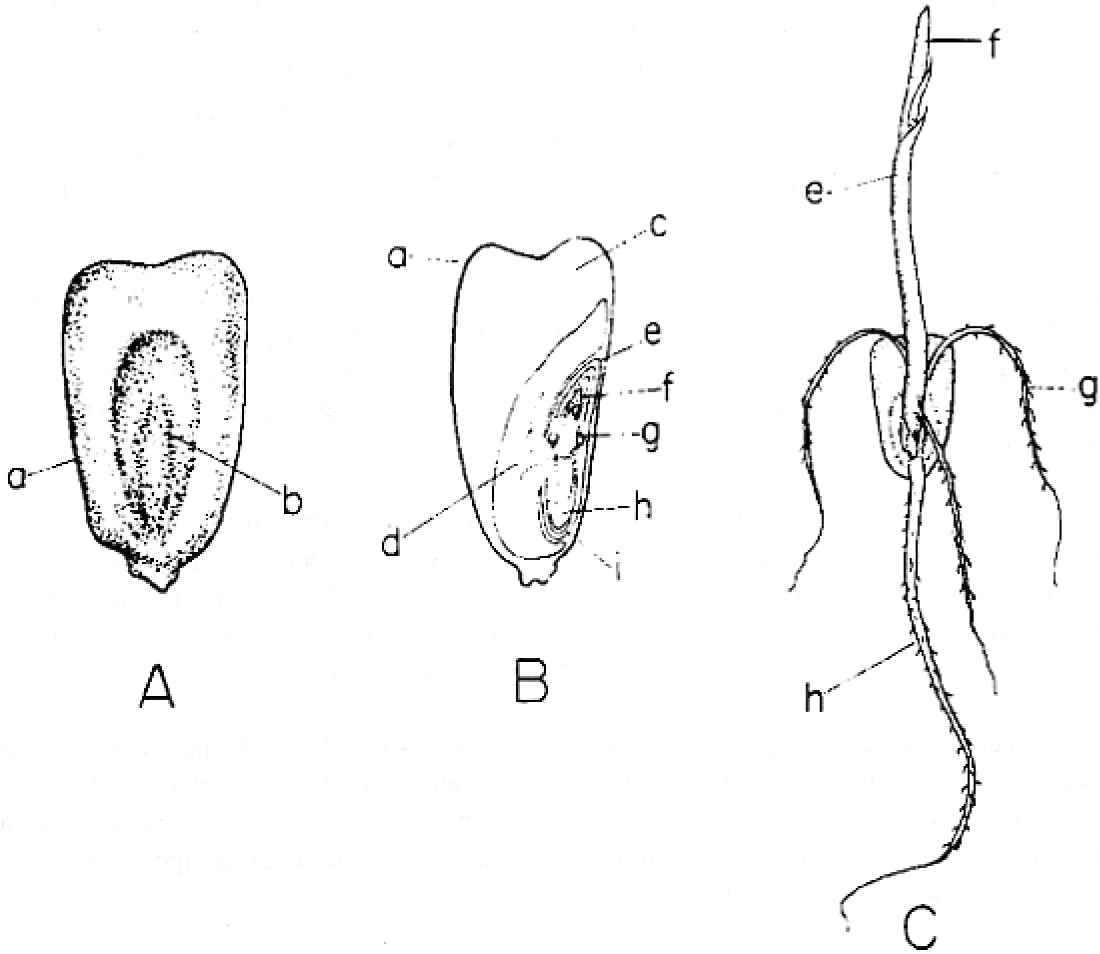


Figure 1. Corn seed Structure, from AOSA (1970)
A. External view of caryopsis. B. Bisectional view of caryopsis. C. Seedling
a. pericarp; b. embryo; c. endosperm; d. scutellum; e. coleoptile; f. plumule; g. seminal root; h. radicle; i. coleorhiza.

Delouche et al. (1962) specified some general rules of interpretation. First, the larger the unstained area inside the embryo, the more likely it is that the corn seed is not viable. And, the radicle of the corn seed could be unstained up to about three-fourth of its length (from the tip) without affecting the germinability. Also, if the seminal root of a corn seed was unstained, it would be classified as non-germinable. Unstained area on the plumule of a corn seed prevents the seed from germinating normally. The identification of the radicle, seminal root, plumule, and other areas of the stained corn seed sections was subtle and difficult. The seed corn TZ analyst must be well trained and experienced to be familiar with the morphology of seed corn sectioning. It was beyond the current image processing and pattern recognition technology to identify the accurate location of crucial cell division areas of the embryo. The current TZ test interpretation was mainly based on how much area was stained inside the embryo since the radicle, seminal root, and plumule were more likely to be stained if a large portion of corn seed was well stained (Howarth and Stanwood, 1992).

The measurement of the area of stained parts was complicated with traditional schemes since the stained area was of random shape and randomly distributed inside the embryo. Howarth and Stanwood (1992) developed a machine vision system to measure the TZ-stained area of each corn seed. The corn seeds were classified into viable or non-viable categories based on the ratio between the stained area and the area of its whole kernel. The algorithm was successful with an error rate of about 10%.

However, Howarth and Stanwood's algorithm employed only a one-feature-two-class classification function. The feature was the ratio between the stained area and the total area of the whole kernel. This single feature interpretation accuracy could be improved on by using more features. Corn seeds of different varieties had embryos of different sizes, independent of their viability. The exact feature should be the ratio between the stained area and the area of the embryo though the area of the embryo could not be extracted with current machine vision techniques. More features, such as the approximation of the embryo area, could be added to improve the accuracy of the interpretation. With more features added, more reliable discriminant functions could be developed to classify the corn seeds into more categories including a third class, called weakly viable.

Development of a Machine Vision Algorithm for Tetrazolium Viability Classification

A corn seed viability classification algorithm was developed to interpret the results from tetrazolium test. The algorithm used image processing and classification techniques after grabbing the image of a stained corn seed section. The processes included image enhancement, image segmentation, feature extraction, and classification. The algorithm implemented the general criteria for interpreting tetrazolium test results on corn seed. It evaluated corn seed viability based on how predominate the stained portion of the embryo part was.

Image Acquisition

We used a perspective camera model to acquire images. The corn seed sections were situated in a plane perpendicular to the optical axis. The bottom surface of the half corn seed was cut flat in order for the corn seed to remain in a controlled fixed position under the camera..

The resolution of the RGB CCD camera was 512 x 480 pixels. The field of view was 30.2 x 28.3 mm. The spatial resolution was approximately 0.059 mm/pixel. Up to four corn seed sections could be grabbed inside one image with resolution less than 256 x 240 for each. The sections could be situated in any random orientation and at any position inside the field of view but they could not touch one another. The background was a flat wooden surface painted black. The black background with low reflectance exposed the objects of interest with higher R, G, and B value at every pixel. The black background did not give higher R, G, and B values to the objects. The distant gray level separation between background and objects made the image segmentation easier.

The RGB image with 512 x 480 resolution was saved momentarily in the Matrox on-board frame buffer during the running of the program. The RGB image took one-fourth of the total space of the frame buffer. The other space of the frame buffer was used to save intermediate results of the image processing operations. The sections were processed and classified one by one in each step of the program.

Image Enhancement

Some pixels in the background had abnormally high gray levels within the gray level range of pixels of objects due to the noise. Those sparse points were taken as separate objects with global segmentation techniques. So, the original images could not be used directly. Noise reduction was done for the purpose of later image segmentation. The image noise reduction was done in the spatial domain by calling a Matrox on-board spatial filter kernel. The filter was set as an 3x3 averaging filter:

$$\frac{1}{9}\begin{bmatrix} 1 & 1 & 1 \\ 1 & 1 & 1 \\ 1 & 1 & 1 \end{bmatrix}$$

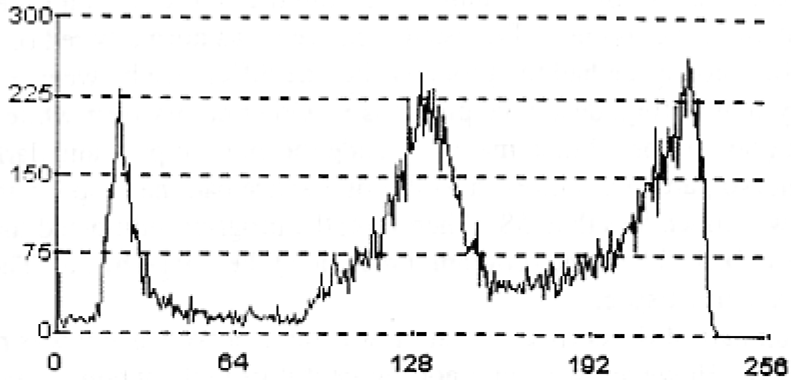
The filter was run through the entire image at a speed of 61 frames/second.

Image Segmentation

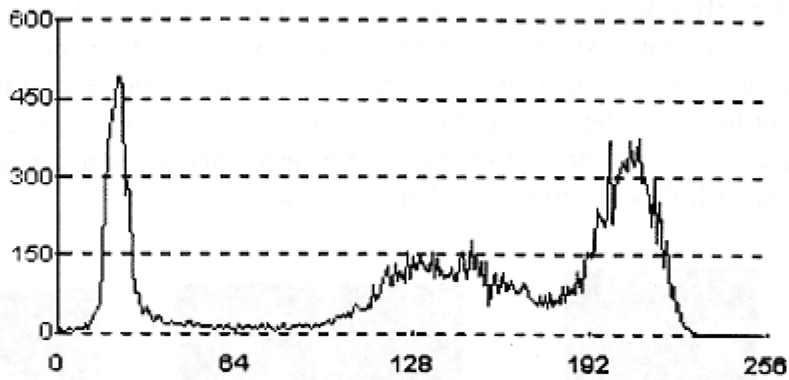
The objects of interest in this study fell into three levels of information details. The first was the sections out of the whole image. The second was the individual corn seed sections out of all the sections. The third was the stained area out of the whole kernel for each corn seed section. Therefore, the segmentation was done in three steps. First, we found a fixed threshold value from the histogram of the red plane. Since the background had a very low reflectance, a fixed value could separate the objects from its background. This threshold value selection was done with a separate program. The typical histograms of the red plane of a stained corn seed image are shown in Figure 2. The background pixels always dominated the area less than 65 and took a shape of normal distribution while object pixels dominated the area of higher gray levels and took different shapes due to variant staining status. The threshold value for this program was set at 60. It worked well as long as the image acquisition system remained unchanged. This meant that the same illumination, camera setting, and the same background was used.

The program globally thresholded the image into two parts. All the pixels with a gray level less than 60 were assigned the value 0, and all pixels with a gray level greater than or equal to 60 were assigned the value 255. The 0 gray level area was the background and the 255 gray level area was the corn seed sections.

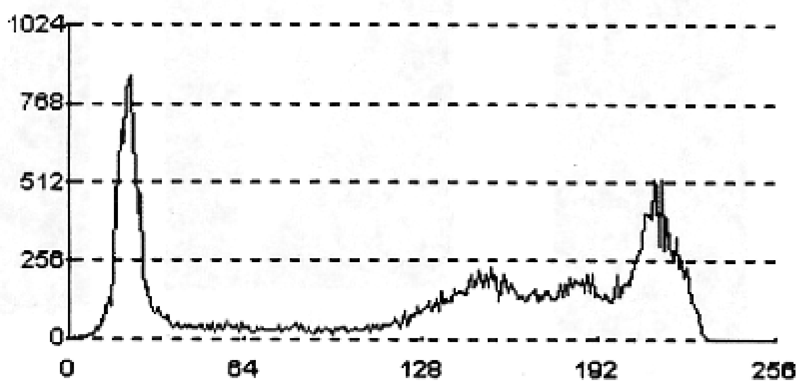
The foreground particles were divided into separate objects, which corresponded to individual corn seed sections. Here, the region-oriented segmentation techniques were used. The foreground pixels were grouped together with their 8-connected neighbors. Each grown region had a unique gray value. The unique gray value started at 1. Thus, the last region had the gray value, n, assuming there were a total of n corn seed sections inside the image. Usually, n is 4. The region of the ith section could be segmented by a single value threshold, i. The result was a 512 x 480 binary image with only ith section region. The ith section was isolated by ANDing the binary image with R and G planes of the smoothed image. The B component of the image was not needed for the next processing operation.



Histogram of red plane of a well stained corn seed section.



Histogram of red plane of a partly stained seed corn section.



Histogram of red plane of a poorly stained corn seed section

Figure 2. Typical histograms of red plane of stained seed corn sections. Horizontal axis is the gray level, vertical axis is the number of pixels

The third step was to isolate the sound area from the whole section for each corn kernel. The sound color of an active tissue of TZ tested corn seed was normally red or bright pink. The sound color in RGB color space had the property that the difference between R and G was greater than 58 with the system setting, described previously. Other colors were white or yellow for the endosperm, bright gray for the whitish material inside the embryo part, and dark red for the heavily stained embryo part. The colors in RGB color space had the property that the difference between R and G was much less than 58. Therefore, the program subtracted the G plane from the R plane pixel to pixel. Then, the program thresholded the image by 58. The foreground was the sound area with active tissue.

A significant part of the corn seed section was the embryo since it was related to the corn seed viability directly. However, we could not segment it from the whole section when the corn seed was not fully and soundly stained. We approximated the embryo part with the convex hull of the sound area. The convex hull was computed by calling the Matrox on-board morphological routine. For many partly stained sections, the approximation was very close. Figure 3 shows images that are typical of partly stained corn whose convex hull of their sound area closely approximate the embryo area. The approximation was poor for some section that stained very little. Since another feature independent of the embryo part was also used and the embryo independent feature was the dominant factor for the little stained section, this poor approximation brought little error to the final classification.

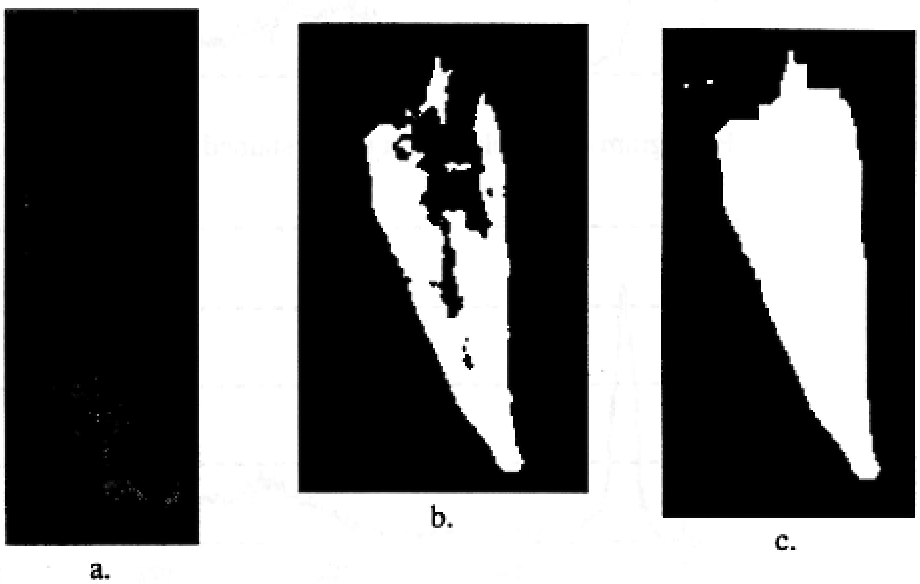


Figure 3. Convex hull of the stained area of a TZ stained seed corn section
a. stained seed corn b. stained area segmented c. convex hull of stained area

Feature Extraction

The whole section was segmented and the embryo area and the sound area were approximated. Each segmented object was analyzed to obtain its area. The pixel area was computed by calling the Matrox on-board statistical routine to count the number of pixels for each object.

The first feature x_1 was the ratio between the area of the sound area and that of the whole section. The second feature x_2 was the ratio between the area of the sound area and that of the approximated embryo part. The second feature was the most important of the two if the approximation of the embryo part was accurate. The two features were combined for the classification.

Classification

Initially, the two features of a test sample of 100 kernels were computed. The viability level of the corn seeds was evaluated by the TZ test interpretation standard. The 100 feature vectors were draw in the two-dimensional space with x_1 as x coordinate and x_2 as y coordinate. (Figure 4) The corn seeds with same viability level tended to fall into the same regions of the vector plane. Minimum distance classifiers are derived from the 2-d feature space. The corn kernels are linearly separated into three categories by two discriminant functions:

$$d_1 = 0.233x_1 + 0.4808x_2 - 0.3595$$
$$d_2 = 0.1654x_1 + 0.2896x_2 - 0.2927$$

The two discriminant functions were used to classify corn seeds. If $d_1 < 0$, the corn seed was classified as non-viable. Otherwise, if $d_2 < 0$, the corn seed would be classified as weakly viable. Or if $d_2 \geq 0$, the corn seed would be classified viable. There is a case when no area of the seed corn section is stained; and therefore, the corn kernel will be classified as totally dead.

Samples Tested

A series of experiments were conducted to test the two-feature viability classification algorithm. Three varieties of corn seeds were used. They were hard endosperm, medium endosperm, and soft endosperm. All three varieties were dried to 14% from initial moisture contents of 30%, 25%, or 20%. The drying air temperatures used were 25 (ambient), 50, 60, and 70°C.

The total number of test samples was 36 with 3 varieties, 3 initial moisture contents, and 4 drying air temperatures. There were 50 kernels tested for each of the 36 samples.

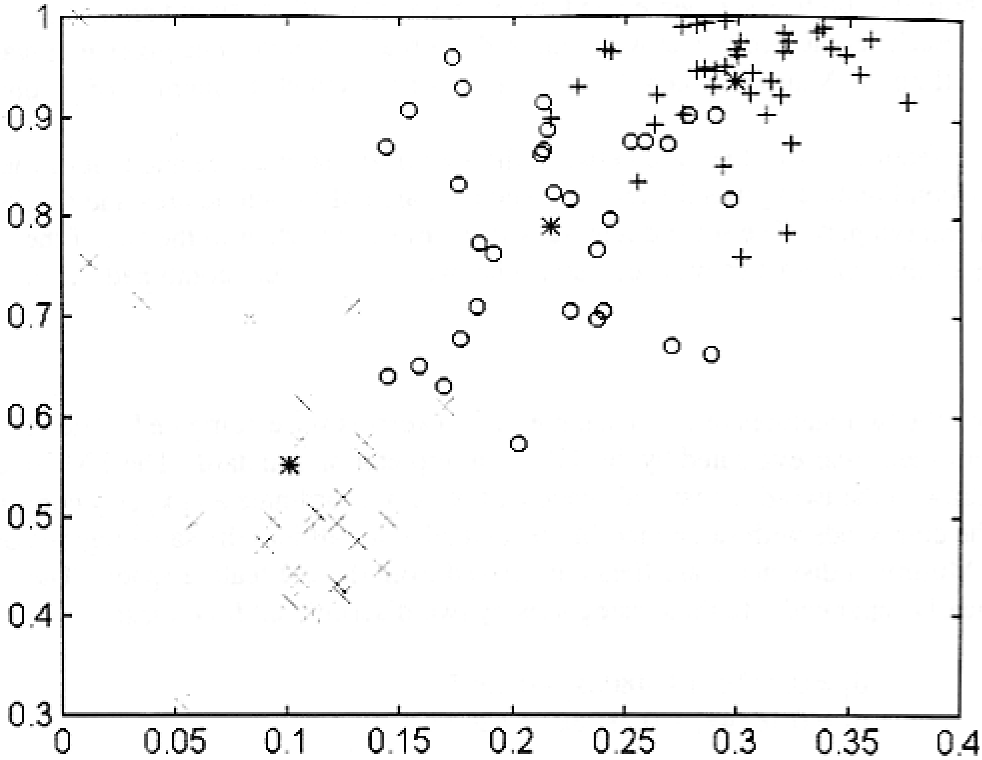


Figure 4. Feature space of stained seed corn section. Horizontal axis is x_1 and vertical axis is x_2. X represents non-viable seed corn kernels, o represents weakly viable, + represents viable. ✱ represents the center of each class

RESULTS AND DISCUSSION

Machine Vision Algorithm for Tetrazolium Viability Classification

The machine vision program classified each corn seed kernel into one of four classes. The four classes were: viable, weakly viable, non-viable, and totally dead. The results are listed in Table 1.

Independent tetrazolium and warm germination tests were performed by the Illinois Crop Improvement Association (ICIA). The ICIA tests used portions of the same 36 samples that were tested with machine vision tests, as shown in Table 2.

Figures 5, 6, and 7 compared machine vision viability percentages with Illinois Crop Improvement Association's tetrazolim test and warm germination test results.

Table 1. Mean machine vision classification results for 50 kernels tested from each of the 36 samples.

Variety	Initial moisture, %	Temperature °C	Viable %	Weakly Viable %	Non-Viable %	Totally dead %
hard	20	25	86	14	0	0
		50	72	28	0	0
		60	66	34	0	0
		70	12	24	58	6
	25	25	60	40	0	0
		50	44	46	10	0
		60	56	30	14	0
		70	2	14	74	10
	30	25	82	18	0	0
		50	64	36	0	0
		60	24	46	30	0
		70	4	8	52	36
medium	20	25	96	4	0	0
		50	80	20	0	0
		60	58	42	0	0
		70	34	60	6	0
	25	25	78	20	2	0
		50	57	39	4	0
		60	62	36	2	0
		70	20	24	46	10
	30	25	82	18	0	0
		50	64	36	0	0
		60	16	60	22	2
		70	0	10	82	8
soft	20	25	82	16	2	0
		50	76	18	6	0
		60	78	20	2	0
		70	70	28	2	0
	25	25	86	14	0	0
		50	70	28	2	0
		60	70	28	2	0
		70	8	46	46	0
	30	25	88	12	0	0
		50	76	24	0	0
		60	34	36	30	0
		70	0	4	60	36

Table 2. Illinois Crop Improvement Association tests for warm germination and tetrazolium for the 36 samples.

Variety	Initial moisture %	Temperature °C	TZ test %	Warm germination %
hard	20	25	95	89
		50	95	91
		60	88	90
		70	23	28
	25	25	86	84
		50	82	79
		60	95	70
		70	13	15
	30	25	90	79
		50	77	80
		60	75	47
		70	4	2
medium	20	25	81	88
		50	87	87
		60	86	88
		70	56	55
	25	25	89	85
		50	71	78
		60	78	86
		70	17	41
	30	25	95	77
		50	94	75
		60	78	34
		70	3	3
soft	20	25	88	81
		50	93	92
		60	95	87
		70	89	79
	25	25	95	80
		50	80	86
		60	92	82
		70	39	45
	30	25	92	71
		50	54	61
		60	15	25
		70	0	2

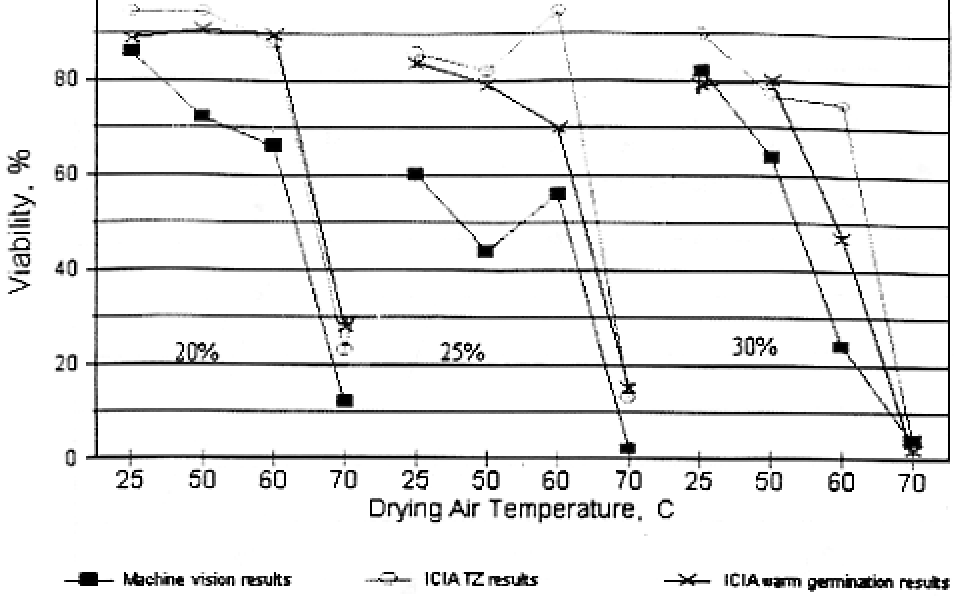


Figure 5. Percentage of viability as a function of drying air temperature for hard endosperm corn at initial moistures of 20, 25, and 30%.

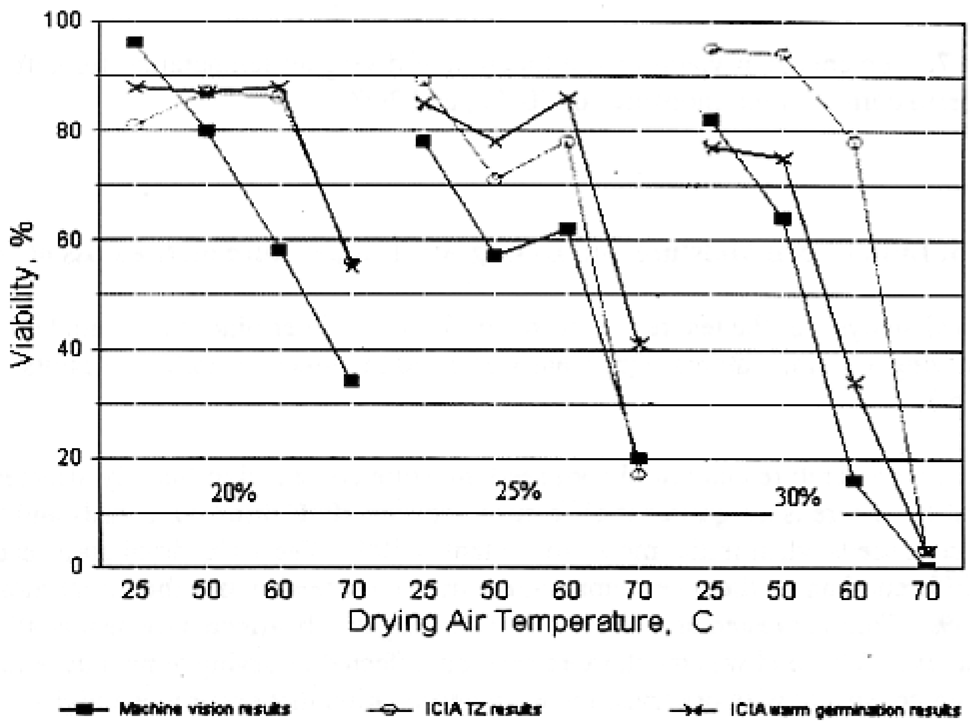


Figure 6. Percentage of viability as a function of drying air temperature for medium endosperm corn at initial moistures of 20, 25, and 30%.

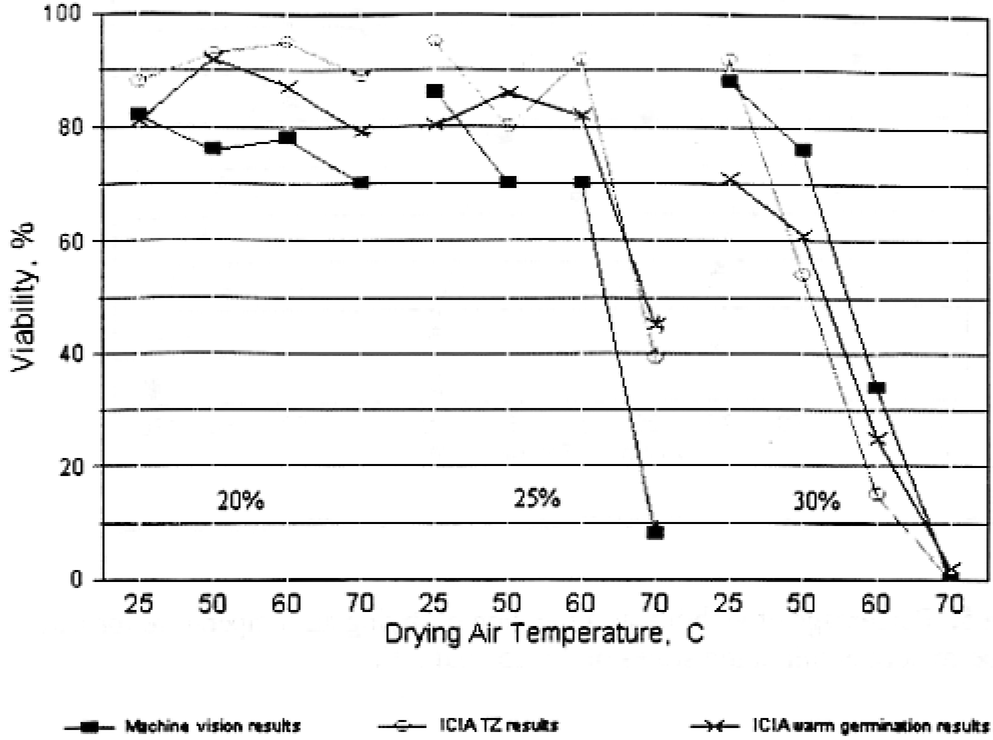


Figure 7. Percentage of viability as a function of drying air temperature for soft endosperm corn at initial moistures of 20, 25, and 30%.

Relationship of Initial Corn Moisture and Drying Air Temperature on the Loss of Viability

Statistical analysis of the test results were conducted to determine if the initial corn moisture and drying air temperatures significantly affected the loss of viability. Results are shown in Table 3.

Drying air temperature and initial corn seed moisture content significantly affected final corn seed viability. There is a significant difference between 70°C dried corn seeds and the 50° to 25°C dried corn seeds when initial moisture content is 20%. The 70°C dried corn seeds were significantly different than all the other temperature dried corn seeds when the initial moisture content was 25%. Corn harvested at 30% moisture was negatively affected by heat at all drying temperatures above 25°C, and was much more severely affected as drying temperature increased. These results are consistent with observed losses in germination that occur when high-temperature air is used to dry very wet seed corn, Baker et al. (1991).

Table 3. Statistical analysis for determining the effect of drying air temperature on viability as measured by machine vision on tetrazolium-stained corn seeds

Moisture %	Temperature	N	Mean	Duncan Grouping
20	ambient	3	88.0	A
	50	3	76.0	A
	60	3	67.3	A B
	70	3	38.7	B
25	ambient	3	74.7	A
	50	3	62.7	A
	60	3	57.0	A
	70	3	10.0	B
30	ambient	3	84.0	A
	50	3	68.0	B
	60	3	24.7	C
	70	3	1.3	D

SUMMARY AND CONCLUSIONS

The tetrazolium staining procedure was used with a 0.1% 2, 3, 5-triphenyl tetrazolium chloride solution. The corn seeds were first moistened in paper towels for 6 hours, then sectioned and placed in the TZ solution for 3 hours at 20°C in a dark environment.

A machine vision algorithm was developed using two ratios, first the sound area to the whole kernel area, and secondly the sound area to the approximate embryo area. When the features were combined, the predicted seed viability compared favorably with actual tetrazolium and warm germination tests.

The machine vision tetrazolium test was able to predict viability loss and therefore detrimental effects for wet milling due to heat damage. Corn harvested at 20% and 25% moisture was negatively affected by drying at 70°C. Corn harvested at 30% moisture was negatively affected by heat at all drying temperatures above 25°C, and was much more severely affected as drying temperature increased.

REFERENCES

Aldrich, Samuel R. & Leng, Earl R. 1965. Modern Corn Production. The Farm Quarterly, Cincinnati, OH.

Association of Official Seed Analysts (AOSA). 1970. *Tetrazolium Testing Handbook.* The Association Official Seed Analysts.

Association of Official Seed Analysts. 1988. *Rules for testing seeds.* The Association of Official Seed Analysts.

Baker, K.D., Paulsen, M.R., Zweden, J. Van. 1991. Hybrid and Drying Rate Effect on Seed Corn Viability. *Transactions of ASAE. 34(2):499-506.* St. Joseph, MI 49085-9659 USA

Copeland, L.O. and M.B. McDonald. 1985. *Principles of Seed Science and Technology.* Burgess Publishing Company, Minneapolis, MN.

Delouche, J.C., T.W. Still, M. Raspet and M. Lienhard. 1962. *The Tetrazolium Test for Seed Viability.* Agriculture Experiment Station, Mississippi State University. Starkville, MS

Duda, P.O. and P.E. Hart. 1973. *Pattern Classification and Scene Analysis.* John Wiley and Sons, New York, NY.

Gonzales, R.C. and R.E. Woods. 1993. *Digital Image Processing.* Addison-Wesley Publishing Company. New York, NY.

Howarth, M.S. and P. C.Stanwood. 1992. Tetrazolium staining viability seed test using color image processing. ASAE Paper 92-3578 ASAE. St. Joseph, MI.

Lakon, G. 1942. Topographical detection of the viability of cereal seeds by tetrazolium salts. Ber. Deut. Bot. Ges. 60:299-305. Saatgut-Wirtsch.

Liao, K., M.R. Paulsen, and J.F. Reid. 1994. Real-time detection of color and surface defects of maize kernels using machine vision. *Journal of Agricultural Engineering Research 59: 263-271.*

Reid, J.F., C. Kim, and M.R. Paulsen.. 1991. Computer vision sensing of stress cracks in corn kernels. *Transactions of ASAE. 34(5):2236-2245.*

Zayas, I, J.L. Steele, and R.E. Dempster. 1990. Pattern recognition of corn defects by color image analysis. *1990 AACC Abstract No. 267.* American Association of Cereal Chemists, St. Paul, MN.