Effect of Image Resolution on Insect Detection in Wheat Radiographs

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ABSTRACT

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Wheat kernels were exposed for three days to adults of three insect species (*Sitophilus zeamais* Motschulsky [maize weevil], *Rhyzopertha dominica* (F.) [lesser grain borer], and *Sitotroga cerealella* (Oliv.) [Angoumois grain moth]); the kernels were incubated and X-rayed to produce contact films at three- to four-day intervals. The objective was to develop specifications for automated grain-inspection systems and evaluate the effectiveness of current radiographic methods. X-ray images of infested and uninfested kernels were presented to four trained subjects as original films and digitized video images at four magnifications corresponding to 32.8, 65.6, 131.2, and 262.4 μm of film per pixel. Insect detection was a sigmoidal function of insect age. The age required for 50% detection varied linearly with log pixel size (μm of film per pixel). Visual detection from the original films was similar to that from the 65.6 μm/pixel resolution. Ages for equivalent detection increased from the maize weevil to the Angoumois grain moth to the lesser grain borer, reflecting the rate of maturation of the individual species. Development of an automated image-acquisition system equivalent to visual inspection will depend on the availability of an X-ray sensor that is 65 μm or smaller.

Detection and control of insects during grain storage and commerce is a major problem (Storey et al 1982, Gecan and Atkinson, 1983). Internal infestation in commercial wheat received at mills is the principal source of insect fragments in flour (Farrell and Milner 1952). Three methods of analysis are used by the Food and Drug Administration to determine internal insect infestation in wheat (Russell 1988). These include the cracking and floatation method (AOAC 1984, methods 44.041 and 44.042), visual examination for wheat kernels damaged by insects, and X-ray examination (AACC 1983). Russell (1988) confirmed that visual examination, though more rapid, is not reliable for detecting hidden weevils that are developing internally, whereas the X-ray and flotation methods give equivalent results.

Schatzki and Fine (1988) briefly reviewed quality standards for insects in wheat and surveyed current and proposed methods for detecting hidden insects. Earlier reviews were published by LeTorc'h (1980), Arteman (1980), and Chambers (1987). Other methods include measurement of respired carbon dioxide (Bruce et al 1982, Sinha et al 1986a,b), acoustical detection in samples or silos (Vick et al 1988, Hagstrum et al 1990), nuclear magnetic resonance spectroscopy (Chambers et al 1984), and determination of uric acid in ground wheat by high-performance liquid chromatography (Wehling et al 1984). Each of these methods has advantages and limitations with respect to sensitivity, selectivity, speed, and cost.

Approximately 40% of U.S. millers and processors use X-ray radiography for quality control in accepting grain shipments (Arteman 1981). This method is subjective in that it relies on the judgment of a trained observer to find internally hidden insects and distinguish them from other grain features. Despite widespread use, it is not known how reliable X-rays are and how this method varies with the age of the insect. Using magnification and image enhancement of single wheat kernels, Schatzki and Fine (1988) attained 80% recognition of the maize weevil, Angoumois grain moth, and lesser grain borer larvae at 8.4, 15.7, and 27.2 days, respectively, after oviposition. That data made it appear worthwhile to determine whether computer images could be used for grain inspection.

The current study was designed to 1) quantify the detection of insects using current methods of visual inspection of X-ray films, 2) compare insect detection in digitized images of varying resolution with insect detection from the original X-ray films, and 3) estimate the image requirements of an automated X-ray grain-inspection system.

MATERIALS AND METHODS

Incubation and X-Rays

The methods used were generally the same as those of Schatzki and Fine (1988). Insect colonies of three species (*Sitophilus*

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zeamais (Motsch.) [maize weevil], *Rhyzopertha dominica* (F.) [lesser grain borer], and *Sitotroga cerealella* (Oliv.) [Angoumois grain moth]) were obtained from laboratories of the U.S. Department of Agriculture in Manhattan, KS, and Fresno, CA. Hard winter wheat (obtained from General Mills, Vallejo, CA) was tempered to 13% moisture, determined by AOAC method 14.003 (AOAC 1984). Wheat (50 g, plus 6 g of flour for *R. dominica*) was exposed to unsexed parent insects of a single species (50 maize weevils, 50 lesser grain borers, or five Angoumois grain moths) in an incubator (25°C, 70% RH) for three days, at which time the parents were removed. Using a minimum amount of rubber cement, kernels were mounted on two 1-ml polyethylene sheets in 17 × 17 kernel arrays for each insect. The plastic sheets were stapled to a wooden frame and placed in the incubator. (The wooden frame was secured with gaskets and clamps between a framed copper screen and a piece of plywood to prevent any emerging insects from escaping.) At three- to four-day intervals, contact X-rays were prepared using a Faxitron 43804N (Hewlett-Packard, McMinnville, OR) set at 25 kV for 0.5 min with Kodak M film. X-rays of individual kernels were transferred to an image-processing computer as described by Schatzki and Fine (1988) using a microscope (Nikon SMZ-10) and videocamera (Sony XC-57 CCD). This resulted in a total image data set (obtained over 10 time periods) containing 56 kernels infested with maize weevil, 41 with Angoumois grain moth, and 113 with lesser grain borer. Kernels containing two larvae that did not mature or any weevil egg holes were eliminated from the image set. Images of non-infested kernels (2,083) were obtained during one time period.

Recognition Tests

Kernels were classified as infested on the basis of mature insects observed in the later films. For recognition tests, composite images were created containing one infested kernel in an array of non-infested kernels. Formats were 2 × 2, 4 × 4, 8, × 8 or 16 × 16 kernel arrays for 32.8, 65.6, 131.2, and 262.4 μm of film per pixel, respectively. For each insect-format combination, an image data set was constructed for each of the 10 time periods consisting of 25 composites (14, 17, 21, 24, 28, 31, 35, 38, 42, and 45 days from start of oviposition) and an equal number of noninfested composites, bringing the total to 500 composites per image set. For the maize weevil, images from day 10 were also obtained, bringing these image sets to a total of 550 composites. Image-data sets were presented on a 13-in., diagonal, high-resolution, red-green-blue monitor (Hitachi HM-271-3C-01) to four trained subjects.

For training, the subjects viewed a set of known images for each insect, with 10 examples of infested kernels at each age. The subjects then responded to a practice set of 132 unknowns and reviewed the errors. Then the subjects were shown the test image sets and were asked whether or not a composite image contained an infested kernel. The image sets were shown in sequence, beginning with the highest resolution (32.8 μm/pixel) and ending with the lowest resolution, (262.4 μm/pixel) for each insect species. The subjects, allowed to work at their own speed, completed most image sets in 1–3 hr (including rest periods).

Data Analysis

The probit procedure of the Statistical Analysis System (version 6.04, SAS 1987,1988) was used to obtain maximum-likelihood estimates of B_0 and B_1 in the logistic model (equation 1, below) (Neter and Wasserman 1974) for each insect, format, and subject combination.

$$P = C + (1-C)\left[\frac{exp\,(B_0 + B_1 X)}{1 + exp\,(B_0 + B_1 X)}\right]$$

where P = the proportion of insects recognized, B_0 = the extrapolated intercept of the linearized logit function, B_1 = the slope of the linearized function, and X = the independent variable (days), and C (the lower asymptote) = the baseline or natural threshold response rate. For each equation, C was fixed at the proportion of uninfested composites identified as infested. The data set for each curve was restricted to eliminate excess data points in the asymptotic tails before fitting. In two cases (lesser grain borer, resolutions of 131.2 and 262.4 μm/pixel), where the data appeared to have an asymptote less than 1.0, the data were restricted to the region that could be fitted by the model. In these cases, it is not possible to extrapolate predictions beyond the data shown. There was no significant lack of fit ($P < 0.05$, Pearson chi-square) for 55 of the 60 fitted equations. The film data for the lesser grain borer from two subjects and for the Angoumois grain moth from three subjects showed a statistically significant ($P < 0.05$) lack of fit. This was due to a slightly steeper initial slope of the data compared with the prediction. Before being used as weights in subsequent calculations, the variances of the affected parameters were increased by a heterogeneity factor [h = chi-square/(k − 2)] (Finney 1971). The mean (μ) of a tolerance distribution can be calculated from equation 2:

$$\mu = -B_0/B_1,$$

where μ estimates the incubation time at which 50% of the infested kernels are recognized. For each insect, the values of μ and B_1 and their respective variances were used in a weighted analysis of variance (ANOVA) (Johnson and Milliken 1983) of resolutions and subjects to obtain the least-squares means of the parameters, which are independent of subject differences. The μ parameters were subjected to further analysis of covariance using $\log_2$ (pixel size) as a continuous variable, with insects and subjects as discreet variables.

RESULTS

ANOVA

Table I presents the observed values for the constant C and the maximum likelihood estimates of μ and B_1 obtained from the logit regressions. These were the input data for subsequent analysis.

The average C values from Table I for each of the four subjects were (A) 0.008 ± 0.002 (mean ± standard error, n = 15), (B) 0.030 ± 0.007, (C) 0.011 ± .003, C, and (D) 0.163 ± .029. Because the variance of subject D was much larger than that of the other subjects, ANOVA including all subjects was not possible. ANOVA using the remaining three subjects indicated no effect of insects ($P > F = 0.57$ in F test for insects), minor effects of resolution on C, and highly significant differences between subjects ($P > F = 0.001$ in F test for subjects).

Table II presents the results of ANOVA for μ and B_1. The upper portion of the table presents the subject-independent least-squares means by insect and resolution. Prediction curves calculated using $C = 0$ (representing no false-positive responses) and the parameters given in Table II are shown in Figures 1–3.

Highly significant ($P < 0.005$) differences among resolutions and subjects were observed for the μ parameter for all insects. These two variables accounted for 94.5–97.6% of the variation in μ. Figure 4 shows the least-squares means for μ in Table II, with the fitted regression lines obtained from the analysis of covariance. Also shown for comparison in Figure 4 are estimates of μ for one subject at 11.5 μm/pixel resolution from data presented by Schatzki and Fine (1988). Their data are not strictly comparable because the image was enhanced by computer to promote earlier recognition of the infested kernels. Nevertheless, these points are close to the extrapolated line based on the current results.

Fewer differences are seen in the slope parameter, B_1. The only important effects were associated with the three different insects. For this reason, an overall B_1 mean is shown in Table II for each insect. For B_1, the proportion of the variation that is explained by the different resolutions and subjects ranges from 0.55 to 0.81 between insects.

DISCUSSION

Insect Recognition

Recognition of the maize weevil, Angoumois grain moth, and lesser grain borer in film radiographs (representative of current practice) reached 50% after 15.6, 21.2, and 29.0 days incubation, respectively. In general, detection of insects in film was most similar to recognition from the 4 × 4 kernel composites corresponding to 65.6 μm/pixel. As pixel size was increased to raise the number of kernels inspected, the age for 50% recognition (μ) increased. μ is a linear function of the logarithm of pixel size over a large range of resolutions (Fig. 4). The increased number of kernels in lower-resolution composites may have contributed to this effect. However, any effect of the increased number of kernels per composite should be counterbalanced by the increased time the subjects spent examining those composites.

Detection of each insect followed a sigmoidal model with more developed larvae required for recognition at decreasing resolution (increasing pixel size). The highly significant effect of subjects on the parameters C and μ demonstrates the subjectivity of inspection results using current methods and suggests a potential gain in objectivity and standardization that could be achieved by a mechanized recognition system. The slopes of the recognition curves (B_1) corresponded to the rate of development of the insect species; the maize weevil produced the steepest slope, followed by the lesser grain borer and the Angoumois grain moth. Decreasing the resolution did not have a consistent effect on the slope parameter; in fact, the slopes for differing resolutions of the same insect were similar.

Requirements of an Automated Image-Acquisition System

An automated grain-inspection system would require the gen-

TABLE I

Effect of Insect, Pixel Size, and Subject on Threshold Constant C (Blank Response) and Logit Regression Parameters μ (Insect Age Required for 50% Detection) and B_1 (Linearized Slope)

		Insect								
		Maize Weevil			Angoumois Grain Moth			Lesser Grain Borer		
Pixel Size (μm)	Subject	C	μ (days)	B_1 (days^{-1})	C	μ (days)	B_1 (days^{-1})	C	μ (days)	B_1 (days^{-1})
Film	A[a]	0.002[b]	17.2 ± 0.36[c]	0.48 ± 0.051	0.001	22.7 ± 1.14	0.32 ± 0.081	0.001	28.8 ± 0.31	0.33 ± 0.046
	B	0.001	12.9 ± 0.49	0.43 ± 0.057	0.002	17.6 ± 0.74	0.25 ± 0.032	0.003	25.8 ± 0.62	0.22 ± 0.020
	C	0.002	16.6 ± 0.37	0.46 ± 0.051	0.002	23.7 ± 0.98	0.30 ± 0.058	0.001	31.0 ± 0.46	0.30 ± 0.025
	D	0.004	16.5 ± 0.68	0.46 ± 0.050	0.005	22.1 ± 1.24	0.19 ± 0.034	0.002	29.0 ± 0.36	0.24 ± 0.013
32.8	A	0.029	9.3 ± 1.33	0.30 ± 0.070	0.016	18.3 ± 0.93	0.24 ± 0.036	0.020	24.5 ± 0.75	0.28 ± 0.037
	B	0.020	11.0 ± 0.85	0.39 ± 0.083	0.012	17.8 ± 0.82	0.30 ± 0.046	0.040	22.2 ± 0.83	0.26 ± 0.037
	C	0.004	11.1 ± 1.54	0.21 ± 0.043	0.032	20.8 ± 1.02	0.26 ± 0.044	0.036	26.3 ± 1.72	0.23 ± 0.064
	D	0.160	10.9 ± 0.85	0.39 ± 0.081	0.192	19.2 ± 0.75	0.32 ± 0.048	0.428	25.5 ± 0.79	0.27 ± 0.037
65.6	A	0.012	15.2 ± 0.56	0.48 ± 0.081	0.000	24.4 ± 0.78	0.24 ± 0.029	0.000	31.0 ± 0.58	0.42 ± 0.057
	B	0.044	13.2 ± 0.64	0.47 ± 0.093	0.056	20.0 ± 0.86	0.26 ± 0.038	0.086	27.5 ± 0.79	0.33 ± 0.053
	C	0.016	18.8 ± 0.82	0.30 ± 0.047	0.016	21.0 ± 1.45	0.17 ± 0.028	0.020	34.2 ± 0.77	0.48 ± 0.108
	D	0.060	13.0 ± 0.62	0.46 ± 0.084	0.228	22.9 ± 0.75	0.28 ± 0.036	0.168	28.6 ± 0.70	0.31 ± 0.042
131.2	A	0.016	20.4 ± 0.69	0.32 ± 0.043	0.004	32.1 ± 0.87	0.20 ± 0.024	0.008	34.2 ± 0.60	0.42 ± 0.064
	B	0.032	17.4 ± 0.79	0.28 ± 0.039	0.080	26.9 ± 0.86	0.25 ± 0.032	0.052	32.8 ± 0.85	0.26 ± 0.045
	C	0.000	22.4 ± 1.07	0.25 ± 0.041	0.008	34.7 ± 1.40	0.15 ± 0.032	0.000	36.2 ± 1.37	0.33 ± 0.140
	D	0.232	17.0 ± 0.78	0.27 ± 0.036	0.216	29.9 ± 0.83	0.22 ± 0.025	0.196	34.4 ± 0.72	0.30 ± 0.046
262.4	A	0.000	26.2 ± 0.75	0.29 ± 0.045	0.004	40.6 ± 0.88	0.25 ± 0.038	0.012	40.6 ± 1.38	0.36 ± 0.125
	B	0.008	23.7 ± 0.62	0.40 ± 0.066	0.008	37.9 ± 0.85	0.24 ± 0.031	0.008	40.0 ± 0.70	0.38 ± 0.076
	C	0.012	27.2 ± 0.94	0.33 ± 0.080	0.008	37.6 ± 1.13	0.19 ± 0.034	0.004	44.0 ± 2.65	0.25 ± 0.125
	D	0.236	25.4 ± 1.95	0.34 ± 0.051	0.136	38.1 ± 1.01	0.19 ± 0.025	0.176	41.1 ± 0.80	0.41 ± 0.091

[a] Subject code.

[b] Values are the fraction of uninfested collages identified as infested.

[c] Values are expressed as maximum-likelihood estimates of μ and B_1 ± standard error of the mean.

TABLE II

Effect of Insect and Resolution on Least-Squares Means for Logit Model Parameters[a]

	Insect					
	Maize Weevil		Angoumois Grain Moth		Lesser Grain Borer	
Resolution Pixel Size	μ (days)	B_1 (days^{-1})	μ (days)	B_1 (days^{-1})	μ (days)	B_1 (days^{-1})
Film	15.6 a[b] ± 0.43	0.46 a ± 0.024	21.2 ab ± 0.80	0.23 a ± 0.022	29.0 a ± 0.30	0.27 a ± 0.008
32.8 μm	11.3 b ± 0.98	0.30 b ± 0.030	19.1 a ± 0.71	0.27 a ± 0.022	24.8 b ± 0.63	0.27 ab ± 0.017
65.6 μm	14.7 ab ± 0.60	0.40 ab ± 0.033	22.6 b ± 0.73	0.23 a ± 0.017	30.4 a ± 0.49	0.36 b ± 0.023
131.2 μm	19.1 c ± 0.75	0.27 b ± 0.019	30.4 c ± 0.77	0.20 a ± 0.015	34.4 c ± 0.60	0.32 ab ± 0.024
262.4 μm	25.5 d ± 0.80	0.32 b ± 0.027	38.8 d ± 0.78	0.21 a ± 0.016	41.5 d ± 0.73	0.38 ab ± 0.040
All	...	0.35 ± 0.012	...	0.23 ± 0.008	...	0.32 ± 0.011
	Proportion of Analysis of Variance Sum of Squares Attributed to Model Components					
Resolution (df = 4)[c]	0.830[d]	0.691	0.933	0.286	0.865	0.313
Subject (df = 3)	0.115	0.109	0.043	0.266	0.107	0.499
Error (df = 12)	0.055	0.200	0.024	0.448	0.028	0.188
Model R^2	0.945	0.800	0.976	0.552	0.972	0.812

[a] Values are expressed as least followed by square —LS means ± standard error of the mean.

[b] Means in each column followed by the same letter are not significantly different at the 0.005 level using Student's t test. This controls the experiment wide error rate at $P < 0.05$ for 10 possible comparisons ($P < 1-(1-0.005)^{10}$).

[c] Degrees of freedom.

[d] Values are the sum of squares for the model component divided by the corrected total sum of squares.

eration of a computer-readable X-ray image, backed up by appropriate image-recognition algorithms to select and possibly remove kernels containing insects. The system of choice would be a belt transport system passing an X-ray beam and a line-scanning X-ray detector array. To match the resolution of visual inspection, the array would require detectors no larger than 65.6 μm. The smallest line-scanning X-ray sensors currently available are 250 μm. To reduce the sensor size by a factor of 4 will require extensive physics and engineering development. Alternatively, one could use larger sensors at the cost of being able to recognize a smaller fraction of the hidden insect life cycle (Fig. 4).

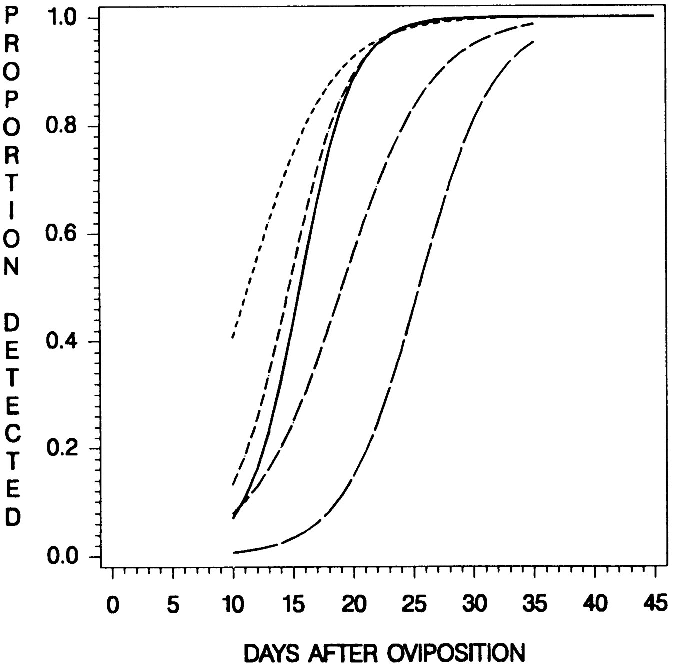

Fig. 1. Predicted recognition of maize weevils as a function of age and resolution. Solid line = film; dotted line = 32.8 μm/pixel; small, medium, and large dashed lines = 65.6, 131.2, and 262.4 μm/pixel, respectively.

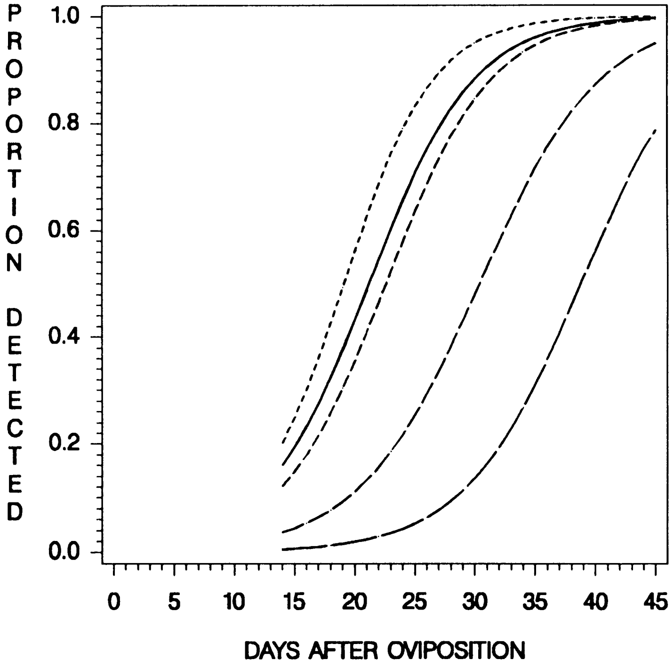

Fig. 2. Predicted recognition of Angoumois grain moths as a function of age and resolution. Solid line = film; dotted line = 32.8 μm/pixel; small, medium, and large dashed lines = 65.6, 131.2, and 262.4 μm/pixel, respectively.

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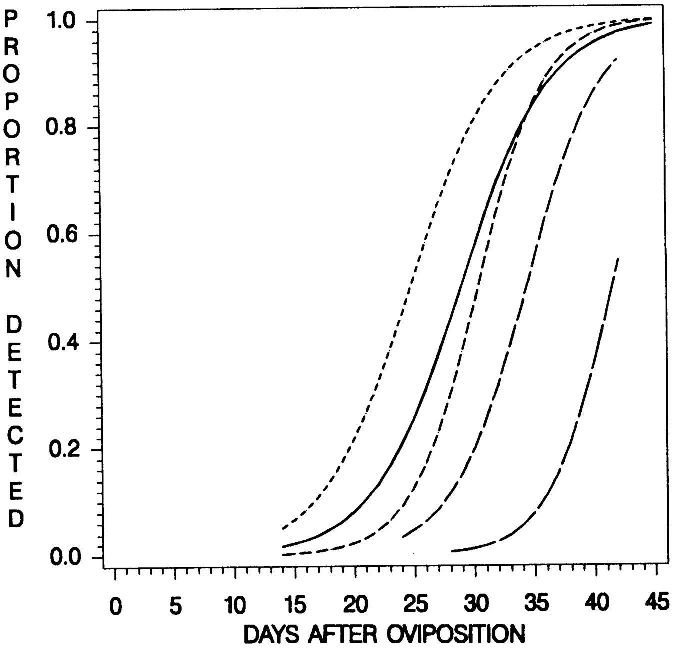

Fig. 3. Predicted recognition of lesser grain borers as a function of age and resolution. Solid line = film; dotted line = 32.8 μm/pixel; small, medium, and large dashed lines = 65.6, 131.2, and 262.4 μm/pixel, respectively. The 65.6 and 131.2 μm resolutions produced responses with asymptotes less than 1.0; the data were restricted to the region that could be fitted by the model. In these cases, it is not possible to extrapolate predictions beyond the data shown.

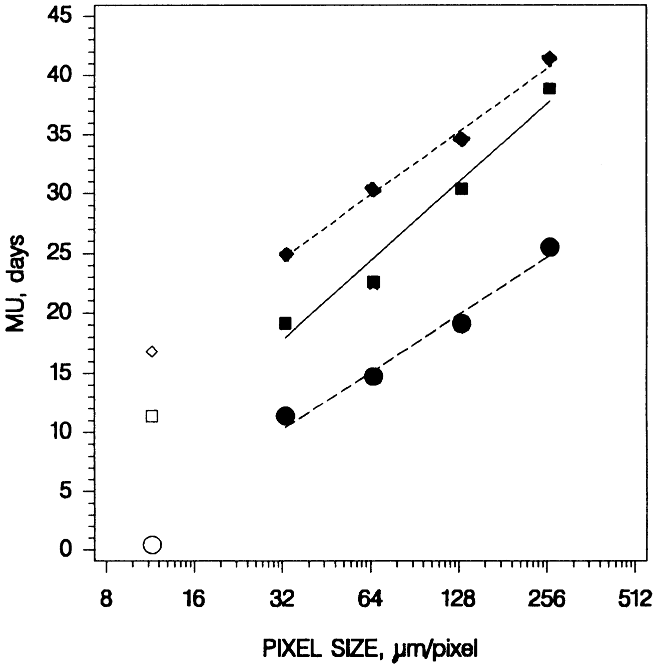

Fig. 4. Age at 50% recognition (μ) as a function of pixel size (μm/pixel). Solid symbols = weighted least-squares means from four subjects in the present work; open symbols = data from Schatzki and Fine (1988) for one subject who observed a computer-enhanced image. ●: Maize weevil regression $\mu = -13.9 + 4.82\cdot\log_2$ (pixel size); ■: Angoumois grain moth regression $\mu = -15.4 + 5.90\cdot\log_2$; ◆ : Lesser grain borer regression $\mu = -2.23 + 5.35\cdot\log_2$. Proportion of analysis of covariance sum of squares attributed to insect = 0.503 (df = 2), resolution = 0.442 (df = 1), insect × resolution = 0.008 (df = 2), subject = 0.023 (df = 3), error = 0.0251 (df = 39), model $R^2 = 0.975$.

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