JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Logistic Regression Modeling of Cropping Systems To Predict Fumonisin Contamination in Maize

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The aims of this research were to monitor the presence of fumonisins in maize crops in northern Italy over a 6 year period, to study the role of the cropping system on fumonisin levels, and to contribute to the development of a predictive system for fumonisin contamination. In the 6 year period from 2002 to 2007, 438 maize samples were collected in five regions, supported by agronomic data, and analyzed for fumonisin content. Fumonisin was detected in almost all of the grain samples, but 2007 was less and 2005 more contaminated compared to the other years. Preceding crop, maturity class of hybrids, nitrogen fertilization, sowing and harvest week, and grain moisture significantly affected the level of contamination. The logistic regression developed explained around 60% of variability with major roles for longitude, maturity class, and growing weeks. The function can be used to quantify the effect of these factors in a predictive system.

KEYWORDS: Cropping system; maize; mycotoxins; fumonisins; modeling; logistic regression

INTRODUCTION

Fusarium verticillioides Sacc. (Niremberg) is the most common toxigenic fungus in maize; it causes ear rot disease, typically occurring on random kernels, groups of kernels, or physically injured kernels, and consists of a white or light pink mold (*1*). In addition to causing ear rot symptoms, *F. verticillioides* is also frequently found in symptomless kernels (*2*). *F. verticillioides* produces primarily fumonisin toxins; these are a group of at least 15 compounds, the most prevalent being fumonisin B_1. Mycotoxins represent a serious, multifaceted economic problem, and maize crops are the most commonly affected, economic losses in maize being due to yield loss caused by diseases induced by the fungi, direct loss of grain which is unfit for sale due to mycotoxin contamination, losses in animal productivity because of mycotoxin-related health problems, and human health costs (*3*).

Approximately 10 million tons of maize per year are produced in Italy, primarily in the Po Valley (northern Italy); a high percentage of this production (88%) is directly consumed as animal feed. In Italy, fusarium ear rot is the most common disease associated with maize ears, and late in the season it can be found at low levels in nearly all maize fields; consequently, fumonisins are the main mycotoxins that affect maize in this geographic area (*4, 5*). Very high levels of fumonisin contamination were recorded in northern Italy over the period from 2004 to 2006 (*6*).

The usual route of mycotoxin exposure is consumption of contaminated food or feed by humans or animals; however, dermal exposure and inhalation may also be important. The primary concern with regard to fumonisin exposure is its possible connection with human esophageal cancer and with a higher incidence of neural tube defects; furthermore, it shows acute toxicity for reared animals (*7, 8*). Worldwide food legislation safeguards the health of consumers and the economic interests of producers and traders, imposing limits on the concentrations of specific mycotoxins in foods. A European Union regulation sets maximum levels for *Fusarium* toxins in foodstuffs (*9*). The maximum tolerated level for total fumonisins ($FB_1 + FB_2$) in raw maize is 4000 μg/kg, and lower levels are fixed for maize-derived foods for direct human consumption. Recommendations have also been made at the European level regarding fumonisin ($FB_1 + FB_2$) content in animal feeds (*10*); the lowest concentration is 5 mg/kg, tabulated for complete feeds destined for pigs and horses.

Fusarium ear rot is more common in warm and dry areas, and it is generally favored by warm, dry weather during the grain-filling period (*1, 11*). In several studies cited by Munkvold (*12*), fumonisin levels were negatively correlated with season-long rainfall or with rainfall in June.

Any management practice able to maximize plant performance and decrease plant stress reduces mycotoxin contamination (*13*). Most of the fungi causing ear rot survive in crop residues; good management of residues is suggested as a control

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10.1021/jf801809d CCC: $40.75
Published on Web 10/09/2008

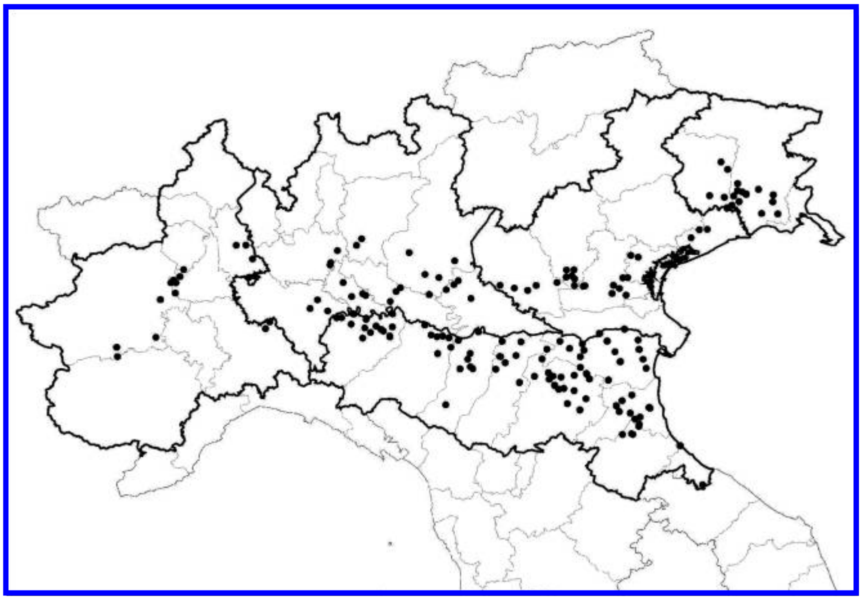

Figure 1. Sampling points in the 2002−2007 maize survey carried out in northern Italy.

measure for this disease (*14*), but there is little direct evidence of the success of this approach. Studies on the survival of *Fusarium* species causing fusarium ear rot suggest that tillage and crop rotation are unlikely to affect this fungus and its mycotoxins (*15*), and tillage practices did not affect the incidence of ear rot caused by *F. verticillioides* or *Fusarium graminearum* in South Africa (*16*). Previous cropping history has been shown to influence soil populations of toxigenic fungi (*17*), but the importance of initial inoculum has not been established. Sowing dates can influence the risk of ear rot; earlier sowing results in a lower risk, greatly depending on annual meteorological conditions. With regard to toxin levels, crop nutrition stress and weed-related stress have been associated with high contamination (*18*). Jones and Duncan (*19*) reported that a higher rate of nitrogen fertilizer consistently reduced aflatoxin levels.

The aims of this research were to monitor the presence of fumonisins in maize crops of northern Italy over a 6 year period, to study the role of the cropping system on fumonisin levels, and to contribute to the development of a predictive system for fumonisin contamination at harvest.

MATERIALS AND METHODS

Sampling and Data Collection. In the 6 year period from 2002 to 2007, samples were collected in five regions where maize is an important crop destined for food or feed: Piedmont, Lombardy, Veneto, Friuli Venezia Giulia, and Emilia Romagna. The monitored area was between 43.8589 and 46.1667 latitude north and between 7.4989 and 13.3327 longitude east.

A total of 98, 98, 84, 77, 37, and 44 maize samples, grown in 2002, 2003, 2004, 2005, 2006, and 2007, respectively, were collected during harvesting from maize crops managed using the ordinary cropping system for each area (**Figure 1**). Sampling of maize was performed according to EC Directive 76/371, Commission of the European Communities (*20*). Samples were dried at 45 °C and analyzed for fumonisins; fumonisin B_1 (FB_1) was determined in all of the samples and FB_2 starting from 2005.

A questionnaire was prepared and completed for each sample to collect the relevant information on the cropping system: soil texture, previous crop, debris management, tillage and other field operations, hybrid seeded, sowing period and investment, mineral nutrition, weeds control, irrigations, flowering period, crop injuries (borers, hail, and wind), chemical control of ECB, harvesting period, and moisture of kernels at harvesting.

A database was built up, including data from the questionnaires and on fumonisin content in all of the samples. All data were georeferenced using GIS Arc View 8.2 [Environmental System Research Institute (ESRI), Redlands, CA].

Fumonisin Analysis. Fumonisins were determined according to the method of Visconti et al. (*21*). Fumonisins were extracted from 10 g of sample in a plastic centrifuge bottle with 50 mL of acetonitrile/methanol/water (25:25:50, v/v/v). After extraction for 45 min using a rotary-shaking stirrer and centrifugation at 4500*g* for 6 min, the supernatant was poured into a flask; another 50 mL of the same solution was added to the residue in the centrifuge bottle, and a second extraction was performed for 30 min. The combined extracts were filtered through a folded filter paper. An aliquot of 2 mL was diluted with 20 mL of 0.1 M phosphate-buffered saline (PBS, pH 7.4) and purified through an immunoaffinity column (R-Biopharm Rhône Ltd., Glasgow, Scotland); after the column had been washed with PBS (2 mL), the fumonisins were slowly eluted (0.5 mL/min) with methanol (6 mL) into a graduated glass vial; subsequently, the eluate was concentrated to 2 mL under a gentle stream of nitrogen. Analysis was carried out using a LC-MS/MS system, consisting of an LC 1.4 Surveyor pump (Thermo-Fisher Scientific, San Jose, CA), a PAL 1.3.1 sampling system (CTC Analytics AG, Zwingen, Switzerland), and a Quantum Discovery Max triple-quadrupole mass spectrometer; the system was controlled by Excalibur 1.4 software (Thermo-Fisher). After dilution of the extract (0.1 mL brought to 1 mL) with acetonitrile/water (30:70, v/v, acidified with 0.4% acetic acid), the fumonisins were separated on a 150 mm × 2.1 mm i.d., 5 μm, Betasil RP-18 column (Thermo-Fisher) with a mobile phase gradient of acetonitrile/water (both acidified with 0.4% acetic acid) from 25:75 to 55:45 in 9 min, then isocratic for 3 min; the flow rate was 0.2 mL/min. Ionization was carried out with an ESI interface (Thermo-Fisher) in positive mode as follows: spray capillary voltage, 4.0 kV; sheath and auxiliary gas, 35 and 14 psi, respectively; temperature of the heated capillary, 270 °C. The mass spectrometer analysis was operated in selected reaction monitoring (SRM). For fragmentation of $[M + H]^+$ ions (*m/z* 722 for FB_1, *m/z* 706 for FB_2), the argon collision pressure was set to 1.5 mTorr and the collision energy to 36 V. The detected fragment ions were *m/z* 704, 352, and 334 for FB_1 and *m/z* 688, 336, and 318 for FB_2. Quantitative determination was performed using LC-Quan 2.0 software.

Fumonisin standards were obtained from Sigma-Aldrich (St. Louis, MO). FB_1 and FB_2 (1 mg) were separately dissolved in 10 mL of acetonitrile/water (1:1, v/v); the concentration was calculated using the weight indicated by the manufacturer. These solutions were diluted to obtain HPLC calibrant solutions in acetonitrile/water (30:70, v/v, acidified with 0.4% acetic acid) at individual concentrations of FB_1 and FB_2 between 2.5 and 50 μg/kg.

Table 1. Index Groups Defined for Agronomic Traits of Maize Samples

index group	soil sand content	preceding crop	maturity class[a]	sowing week	nitrogen	harvest week	grain moisture
1	20	maize	110	≤13	≤200	≤35	≤20
2	40	wheat	118	14−16	200−324	36−37	20−23
3	60	others	125	≥17	≥325	38−39	≥24
4			130			≥40	
5			135				

[a] Maturity class was expressed as mean number of days from emergence to ripe.

The recovery values were estimated by spiking a blank sample with a measured volume of fumonisin standards, so as to obtain contamination levels of 1000 μg/kg. The average recovery values were 95.5 ± 1.9% for FB_1 and 93.6 ± 2.1% for FB_2. The results of the analyses were not corrected for recovery. The limits of detection for both FB_1 and FB_2 (LOD, signal-to-noise ratio 3:1) and quantification (LOQ, signal-to-noise ratio 10:1) were 10 and 30 μg/kg, respectively.

Agronomic Factors and Data Analysis. Statistical data analysis was managed on FB_1 because these data were available for the whole period considered in the study. Four thousand micrograms per kilogram was used as the separation level between good and bad quality maize.

Data collected for each agronomic factor were grouped according to the indices reported in **Table 1**.

Soil texture was expressed as a percentage of sand: coarse, medium, and fine represented 60, 40, and 20% of sand. Maturity class of hybrids was expressed as the mean number of days from emergence to ripe.

A further parameter, growing weeks, was calculated as the difference between harvest and sowing week; it is similar to maturity class but based on the effective seeding and harvesting time.

Univariate analysis of variance was applied to these data, and means were separated using least significant difference (LSD; $P \leq 0.05$). Data analysis was managed with univariate generalized linear model function in SPSS (ver. 13.0.1, 2004).

Development of Models. Logistic regression and a classification and regression tree (CRT) were used as assessment models.

Sensitivity and specificity were calculated for the binary response models. Sensitivity was defined as the percentage of correctly classified cases in class 1 ($FB_1 \geq 4000$ μg/kg) and specificity as the percentage of correctly classified cases in class 0 ($FB_1 < 4000$ μg/kg). The estimated probabilities were compared to the observed data as a measure of goodness of fit of the model.

Logistic Regression. Logistic regression is a multivariate technique for estimating the probability that an event occurs; it was applied to data using the Logistic Regression Module of SPSS. This regression is suggested when the dependent variable is binary and when the assumption of the normal distribution of data, requested by parametric techniques, is not respected. It enables the estimation of the probability of an event occurring (dependent variable) based on independent variables (a given set of conditions). The logistic regression model can be written as follows:

$$\text{Prob(sample\_contaminated)} = \frac{1}{1+e^{-Z}}$$

where e is the base of the natural logarithms and Z is the linear combination

$$Z = B_0 + B_1X_1 + B_2X_2 + ... + B_PX_P$$

where B_0, B_1, B_2,..., B_P are coefficients estimated from the data, X_1, X_2,..., X_P, the independent variables (agronomic traits in this case). The logistic regression was run including longitude and all agronomic traits considered for ANOVA, except preceding crop (it is not a continuous variable as requested by the logistic regression), as independent variables and both observed and yearly standardized data on FB_1. The logistic regression was run two times, using the data set 2002−2006 and with the data set also 2007 included.

Table 2. Mean Yearly Content and Minimum/Maximum Levels of Fumonisin B_1 and B_2 in Samples Collected in the 6 Year Survey in Northern Italy

	FB_1 (μg/kg)		FB_2 (μg/kg)		FB_2/FB_1	
year	mean	min/max	mean	min/max	mean	min/max
2002	5132 (55/98)[a]	<LOQ/19596				
2003	5415 (49/98)	<LOQ/19687				
2004	6303 (44/84)	<LOQ/22174				
2005	6910 (53/77)	358/27418	2561	<LOQ/12572	0.31	0/0.87
2006	4018 (10/37)	<LOQ/21324	1902	<LOQ/8607	0.50	0/0.67
2007	662 (0/44)	<LOQ/3006	374	<LOQ/1920	0.40	0/1.00

[a] The number of samples containing ≥4000 μg/kg of fumonisin B_1, over the total number of samples considered, is reported in parentheses.

The probability values are based on a 0−1 scale; when the value is higher than 0.5, the event is considered to be occurring and vice versa. The parameters of logistic regression are estimated using the "maximum-likelihood method".

Classification and Regression Trees (CRT). In this approach, classification trees are built by using a binary partitioning algorithm to recursively split the data in each node into increasingly homogeneous subsets until the response data are "pure", that is, all of the cases belong to the same class (*22*). CRT attempts to maximize within-node homogeneity, and several non-negative functions are used to determine the purity of the node.

The method used to measure impurity and the minimum decrease in impurity required to split nodes can be selected. For scale-dependent variables, the least-squared deviation (LSD) measure of impurity is suggested, and it was used here for data analysis; it is computed as the within-node variance, adjusted for any frequency weight or influence value (SPSS, ver. 13.0.1, 2004).

The CRT was run with the data set 2002−2007.

RESULTS AND DISCUSSION

During the 6 year survey in northern Italy, FB_1 was detected in almost all of the maize samples; the lowest yearly mean FB_1 contamination was 662 μg/kg in 2007 and the highest was 6910 μg/kg in 2005; the maximum contamination of a sample was around 20000 μg/kg in almost every year, except 2007. Ten samples (27%) exceeded the legal limit in 2006 and 0 in 2007, whereas around 50% of samples were over the threshold in the other years. The rate FB_2/ FB_1 varied between 0.31 in 2005 and 0.50 in 2006 (**Table 2**).

The maize cropping system in northern Italy was managed in the usual way, and some practices monitored, such as soil tillage, debris management, weed control, and plant density, were very similar for all of the samples collected; also, irrigation is a common practice in all mean-long season maize, with very similar volumes. Data on flowering period were reported in few questionnaires; ECB control was not common and applied in several crops only in 2007. Therefore, the cited traits were not included in the data analysis.

All of the parameters included in the ANOVA were significant, except sand content in soil with 5021, 5334, and 4925 μg/kg of FB_1 of kernels, respectively for coarse, medium, and fine soils (**Table 3**).

Fumonisin B_1 content in grain was significantly higher with maize as preceding crop (5940 μg/kg) with respect to wheat (4193 μg/kg), with more or less than 325 kg/ha of nitrogen added with manuring (6940 μg/kg with respect to about 4900 μg/kg), and in long- with respect to short-season hybrids (around 6000 with respect to 4000 μg/kg). Maize seeded before the 16th week (around the 20th of April) showed a FB_1 contamination around 5000 μg/kg, whereas with later sowing it increased to

Table 3. Mean Content of Fumonisin B_1 (Micrograms per Kilogram) in Maize Kernels Obtained from Different Cropping Systems

index group	preceding crop	maturity class	sowing week	nitrogen	harvest week	grain moisture
1	5940 (171)[a] a[b]	4285 (67) b	4860 (221) b	5128 (155) b	3538 (103) b	4257 (132) b
2	4193 (119) b	3933 (45) b	5109 (174) b	4691 (198) b	5541 (161) a	5320 (179) ab
3	5006 (141) ab	4928 (142) ab	8226 (36) a	6940 (75) a	5579 (89) a	5884 (82) a
4		6051 (139) a			5971 (75) a	
5		5991 (43) ab				

[a] The number of observations for each group is reported in parentheses. [b] Different letters indicate significant differences between mean fumonisin B_1 content in the groups of each cropping system ($P \leq 0.05$).

8226 μg/kg; harvest time (later harvest−higher contamination) and grain moisture (higher moisture−higher contamination) also played a significant role.

Development and Validation of Models. Results obtained running the logistic regression were very similar using observed and standardized data, and only nonstandardized data are reported.

The regression coefficients of the logistic equation computed with the data set 2002−2006 were in the range between −0.010 and 0.180. Longitude and maturity class were significant parameters. All of the parameters, except soil sand content, were positive, which means that increasing their values increased the risk for FB_1 contamination.

When the data set 2002−2007 was considered, parameters were in the range between −0.009 and 0.174, very similar to those previously reported. Longitude and maturity class were confirmed, but also sowing week and growing weeks were significant parameters. The sign of parameters was confirmed.

The effect of the significant parameters on the probability of a sample being contaminated, taking into account the 60% of variability explained with the 2002−2007 data set, is 9, 24, 10, and 17%, respectively, for longitude, maturity class, sowing week, and growing weeks. The greatest effect is played by the maturity class, which can affect ±24% the probability of contamination.

According to CRT, the threshold between negative and positive effects corresponds to 10.8806° longitude, 128 days maturity class, and 24 growing weeks.

Fifty-eight percent of samples were correctly classified by the model, 17% with low contamination and 41% with FB_1 above 4000 μg/kg, when the 2002−2006 data set was considered. Fourteen percent of samples were underestimated (predicted <4000 and observed ≥4000), and 28% of incorrectly estimated cases were false alarms, which means samples below the limit but predicted over the limit. The underestimated samples were 9, 13, 7, 21, and 5% and the overestimated 31, 25, 26, 14, and 19%, respectively, in the years from 2002 to 2006. The mean probability of underestimated cases, computed considering the probability associated with each sample by the logistic regression, was 0.42, and only 14 samples (3% of total) were hardly contaminated, with FB_1 >10000 μg/kg: 5 in 2003, 2 in 2004, and 7 in 2005. The mean probability of overestimated cases was 0.59, and only 54 samples (12% of total) were almost safe, with FB_1 <2000 μg/kg.

Model validation with data collected in 2007 (data not included in the model development) showed 41% of samples correctly classified and 59% overestimated; a mean probability of 0.56 was associated with the latter cases with a mean (min/max) FB_1 content of 785 (<LOQ/3006) μg/kg.

Sixty-four percent of samples were correctly classified by the model, 29% with low contamination and 35% with FB_1 >4000 μg/kg when the 2002−2007 data set was considered. Sixteen percent of samples were underestimated, and 20% of incorrectly estimated cases were false alarms. The underestimated samples were 8, 18, 11, 34, 8, and 0% and the overestimated 29, 14, 15, 10, 11, and 9%, respectively, in the years from 2002 to 2007. The mean probability of underestimated cases was 0.39, and only 19 samples were hardly contaminated, with FB_1 >10000 μg/kg, 6 in 2003, 2 in 2004, and 11 in 2005. The mean probability of overestimated cases was 0.59, and only 41 samples were almost safe, with FB_1 <2000 μg/kg.

Results obtained with the CRT run using the 2002−2007 data set were very similar to those from the logistic regression. In fact, the total number of correctly classified cases was 61%, the number of underestimated cases was 17 versus 18%, and the number of false alarms 22 versus 19%.

The role of parameters defined in these two approaches was confirmed, with maturity class, longitude, and growing weeks as more relevant (**Figure 2**). Many cultural practices that discourage disease require decisions to be taken prior to crop seeding. The need for such measures has traditionally been evaluated using informal methods of risk assessment, which usually are very imprecise. Quantitative, site-specific risk assessments or predictive models for mycotoxin accumulation could significantly contribute to crop safety, but limited efforts have been devoted to this issue in the past 10 years.

In a series of studies, Marin et al. (*23*) described the conditions that favor fumonisin production by *F. proliferatum* and *F. verticillioides* and provided a framework for possible prediction of fumonisin accumulation in the field. A mathematical simulation of ear rot development, following inoculation with *F. graminearum* and *F. verticillioides*, was presented by Stewart et al. (*24*). Although this work did not include a component for the prediction of initial infection, it provided a detailed quantitative assessment of environmental influences on postinfection disease development. More recently, a conceptual model for the dynamic simulation of the life cycle of *F. verticillioides* in maize and the production of FB_1 in kernels was developed, following the principles of "systems analysis" (*25*). Attention was focused on fungal ecology, and almost all aspects of the disease cycle were described. Mathematical equations relating relevant meteorological parameters and the infection process were developed, but the cropping system was not considered.

The data set included in this research, obtained in a 6 year survey, showed mean fumonisin levels always exceeding the legal limit for unprocessed maize destined for human consumption, and contamination 6 times higher was sometimes found. Fumonisins are definitely the main problem for maize growers in northern Italy, and the importance of meteorological factors on contamination levels has been confirmed by differences between years in similar growing conditions. The role of the cropping system has also been confirmed; it plays a minor role with respect to meteorology, but the great advantage is that it is manageable by farmers.

The maize hybrid maturity class had an important effect on FB_1 contamination. Late-maturing hybrids were potentially more at risk in the area under study. Furthermore, it is well-known that

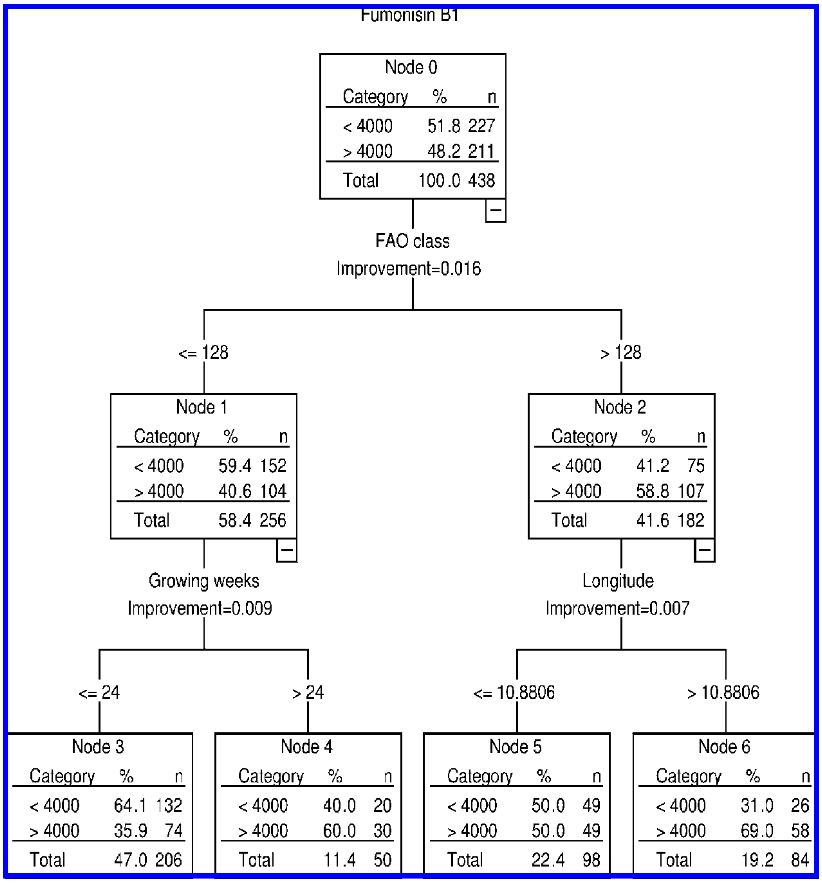

Figure 2. Classification and regression trees for data on fumonisin B_1 contamination in kernels in relation to agronomic traits (data set 2002−2007).

fumonisins are influenced by other genotype traits such as grain hardness or by physical barriers to ear infection (*26*−*28*) and that genetic resistance is actually one of the most studied aspects.

Sowing dates seem relevant to reduce the risk of severe ear rot, but scientific evidence is not yet available.

High nitrogen fertilization significantly increased fumonisin content in maize, in agreement with Marocco et al. (*29*); on the contrary, a reduction of aflatoxin level was reported by Jones and Duncan (*19*). The shared vision is that balanced fertilization generally assures lower contamination.

The real length of the vegetative period (growing weeks) was positively correlated with fumonisin accumulation, indicating more fumonisins in late-harvested maize. Early or prompt harvest at maturity is critical to obtain a crop with minimal fumonisin contamination. The delay in harvesting a crop that is known to have some contamination can only result in a higher amount of fumonisin in grain. Because contamination is cumulative, delay can only exacerbate the problem on infected ears, even when some resistance to contamination is present.

Mycotoxin production in maize can be affected by several cultural practices, partly because of the relationship between drought stress and susceptibility to *F. verticillioides* and fumonisin accumulation. Cultural practices that tend to expose plants to greater drought stress will lead to higher levels of fumonisins. The combined effects of several cultural practices on toxin development in maize have been investigated and summarized (*12*). Earlier planting and harvest along with irrigation or deep tillage to alleviate drought stress all reduced infection and toxin concentration.

Our results indicate that the prevalence of *F. verticillioides* is affected by longitude. Fusarium ear rot is favored in warmer and drier areas (*1, 11*), conditions associated with optimal temperature for *F. verticillioides*. In several studies cited by Miller (*30*), fumonisin levels were negatively correlated with season-long rainfall or with rainfall in June. Furthermore, fusarium ear rot was negatively correlated with rainfall during June and July, but positively correlated with rainfall during the period from August to October. These results are consistent; a dry period during grain filling favors more severe fusarium ear rot, as also stated by Munkvold (*12*).

Fumonisin B_1 was not correlated with soil sand content; probably, the irrigation schedule masked the effect of different soil textures. Although data have so far not demonstrated a significant contribution of soil texture toward the reduction of fumonisins, correlations between soil texture and incidence of *Aspergillus flavus* strains and aflatoxin production are known (*31*).

The agronomic traits significantly related to FB_1 level (ANOVA) were considered to describe their quantitative effect on fumonisin contamination at harvest, using an operative logistic regression focused on the role of the cropping system. Agronomic traits explained about 60% of contamination variability, which is relevant in practice, these being the factors managed by farmers. The years considered in the study were very different for contamination and meteorological conditions, and the validation of the logistic model developed from 2002−2006 data with information collected in 2007 (external validation) did not give satisfactory results; in fact, 56% of samples were overestimated. This overestimation is strictly related to the meteorological conditions of 2007 and represents a weak point of this approach, meteorological data not being included in the model. These results suggested the inclusion of 2007 in the data set for regression, even if validation would have not been possible, because a wider range of years, with meteorological conditions from very conducive to disadvantageous for fumonisin synthesis, improved the accuracy of the logistic model.

The logistic regression run with the wider data set (2002–2007) increased the percentage of explained data. The CRT approach confirmed the results and, moreover, it defined the threshold between high and low risk for the significant agronomic traits.

The role of traits was shown with statistical evidence; the less conducive condition for fumonisin contamination was associated with hybrids with maturity class lower than 128 days and a number of growing weeks lower than or equal to 24, with only 36% of samples over the fixed limit. In contrast, hybrids with maturity class higher than 128 days seeded in areas with a longitude higher than 10.8806 are associated with high risk, with 69% of samples over the fixed limit of 4000 μg/kg of FB_1 (**Figure 2**).

In conclusion, the data reported in this paper give significant support to predictive models for fumonisin contamination in maize, quantifying the role played by agronomic traits.

ACKNOWLEDGMENT

We appreciate the expert assistance provided by Luca Pasini.

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Received for review June 12, 2008. Revised manuscript received September 5, 2008. Accepted September 5, 2008. Work supported by grants Progetto Strategico MIUR- MiPAF 2002–2005 and Progetto CRPV 2004–2006 related to the development of an integrated support system to improve crop production quality and reduce the risk of mycotoxin contamination in food; National MICOCER project 2006–2008 supported by MIPAAF.

JF801809D