***Short Communication***

# Hepatocellular Carcinoma and Aflatoxin Exposure in Zhuqing Village, Fusui County, People's Republic of China[1]

**Jia-Sheng Wang,[2] Tiren Huang, Jianjia Su, Feng Liang, Zhongliang Wei, Yongqiang Liang, Haitao Luo, Shuang-Yuan Kuang, Geng-Sun Qian, Guiju Sun, Xia He, Thomas W. Kensler, and John D. Groopman**

Department of Environmental Health Sciences, School of Hygiene and Public Health, Johns Hopkins University, Baltimore, Maryland 21205 [J-S. W., G. S., X. H., T. W. K., J. D. G.]; Guangxi Cancer Institute, Nanning, Guangxi, People's Republic of China [T. H., J. S.]; Fusui Liver Cancer Institute, Fusui, Guangxi, People's Republic of China [F. L., Z. W., Y. L.]; and Shanghai Cancer Institute, Shanghai, People's Republic of China [H. L., S-Y. K., G-S. Q.].

## Abstract

**Hepatocellular carcinoma (HCC) is a common cause of cancer morbidity and mortality in Asia and Africa. Epidemiological studies have found that dietary exposure to aflatoxin $B_1$ ($AFB_1$) and chronic infection with hepatitis B virus are two major risk factors for HCC. We have collated the incidence and mortality data of malignant tumors from 1973 to 1999 in Zhuqing Village, Fusui County, an area with very high HCC rates, and found that this cancer accounted for 64% of the total cancer incidence. Dietary intake of $AFB_1$ was monitored for 1 week in a study group consisting of 15 males and 14 females from different households in this village. Four of 29 participants (13.8%) and 3 of 15 (20%) male participants were hepatitis B virus surface antigen positive. $AFB_1$ was detectable in 76.7% (23 of 30) of ground corn samples (range, 0.4–128.1 ppb), 66.7% (20 of 30) of cooking peanut oil samples (range, 0.1–52.5 ppb), and 23.3% (7 of 30) of rice samples (range, 0.3–2.0 ppb) collected from each household. Mean levels of serum $AFB_1$-albumin adducts in this group were 1.24 ± 0.31 pmol/mg of albumin at the beginning of the study and 1.21 ± 0.19 pmol/mg of albumin at the end of the period. Urinary $AFB_1$ metabolites were detectable in 88.9% (24 of 27) samples (range, 0.9–3569.7 ng/24-h urine). These data provide the exposure and disease risk information for establishing intervention studies to diminish the impact of aflatoxin exposure in this high-risk population.**

## Introduction

HCC[3] is one of the most common cancers in Asia, Africa, and in groups of Asian- and Hispanic-Americans, and is a leading cause of cancer death (1–3). In the People's Republic of China, HCC is the second leading cause of cancer mortality, and in one high-risk region, Fusui County in Guangxi Zhuang Autonomous Region, the annual incidence rate is >50 cases per 100,000 people (4, 5). HCC usually strikes people at earlier ages in high-risk areas where the median age of onset of this malignancy is between 35 and 50 years. Previous epidemiological studies have found that chronic infection with HBV and dietary aflatoxin exposure are two major etiological risk factors for HCC in China (6–10). The synergistic interaction of HBV infection, aflatoxin exposure, and HCC has been observed in populations in Fusui County (11), Shanghai (12, 13), and other regions (14, 15). To date, there is still a low prevalence of hepatitis C virus infection in these areas of the People's Republic of China (16, 17).

Primary prevention, such as vaccination for HBV in infants and food safety procedures to control aflatoxin contamination, offer strategies for lowering HCC rates in the world; however, positive outcomes will require many years. An immediate challenge in cancer prevention and control is to manage those who are already at high risk, such as individuals who are HBsAg carriers and have chronic aflatoxin exposure. The purpose of the following study was to characterize HCC incidence and mortality, status of dietary aflatoxin exposure, and HBV infection in the high-risk region of Fusui County for use in the design of future chemopreventive intervention studies (18, 19).

## Materials and Methods

**Study Population.** Zhuqing Village, located 45 km southwest of Fusui County capital, is a rural farming community of ~3300 residents and is one of the three townships with the highest incidence and mortality of HCC in Fusui County. Data for analyses of incidence and mortality of malignant tumors were collected from village clinics and were confirmed by the Office of Malignant Tumor Reporting operated by the Fusui Liver Cancer Institute. The incidence and mortality were standardized according to the National Population Proportion of China in 1990.

**Procedure for Molecular Biomarkers Study.** This collaborative study by the Johns Hopkins University, Guangxi Cancer Institute, Shanghai Cancer Institute, and Fusui Liver Cancer Institute used methods and consent forms approved by the Institutional Review Boards for human studies at Johns Hop-

Received 7/19/00; revised 10/24/00; accepted 11/17/00.

The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked *advertisement* in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

[1] This work was supported financially by Program Project Grant ES06052 from the National Institute for Environmental Health Sciences, NIH.

[2] To whom requests for reprints should be addressed, at The Institute of Environmental and Human Health, Texas Tech University System, Box 41163, Lubbock, TX 79409-1163. Phone: (806) 885-0320; Fax: (806) 885-4577; E-mail: js.wang@ttu.edu.

[3] The abbreviations used are: HCC, hepatocellular carcinoma; HBV, hepatitis B virus; HBsAg, hepatitis B surface antigen; HBs, hepatitis B surface protein; HBc, hepatitis B core protein; HBe, hepatitis B virus envelope; $AFB_1$, aflatoxin $B_1$; $AFM_1$, aflatoxin $M_1$.

kins University and at the Guangxi Cancer Institute. The consent form in bilingual (Chinese and English) format was developed, approved, and explained in detail at village meetings among residents and investigators prior to recruitment. To be eligible, participants had to meet the following criteria: adults 25–60 years of age in good general health with no history of chronic illness, no personal history of cancer, no use of prescribed medications, no pregnancy or lactation for women residents of different households, agree to stay in the village for the 1-week study period, and able to provide necessary informed consent. In April 1999, weighed portions of each meal from the participants and 24-h urine in three cycles (morning, noon, and evening) were collected from participants for 7 consecutive days. The urine volume was measured and tested with chemsticks for renal function, and a 200-ml urine aliquot per day was stored at −20°C. In addition, 10 ml of blood were drawn into Vacutainers at the beginning and 5 ml at the end of the 7-day study from each participant. Serum samples were immediately separated by centrifugation at the village clinic and stored at −20°C until analysis.

**Measurement of HBV Seromarkers and Liver Function.** All serum samples were tested for HBsAg and anti-HBs by RIA using the AUSRIA II kit (Abbott Laboratories, North Chicago, IL). A test for the presence of anti-HBc and HBe antigen/anti-HBe was then performed using a commercially available Corab kit purchased from Abbott Laboratories. Liver function tests (aspartate aminotransferase, alanine aminotransferase, and $\alpha$-fetoprotein) were performed in the clinical laboratory of Guangxi Tumor Hospital, which is affiliated with Guangxi Cancer Institute and Guangxi Medical University, according to the clinical diagnostic procedures.

**Measurement of $AFB_1$ in Food, Serum Albumin Biomarkers, and Urinary Biomarker.** Food analysis for $AFB_1$ was performed using a previously published immunoaffinity method (20). Urinary aflatoxin biomarkers and serum aflatoxin-albumin adducts were analyzed according to previously published methods (19, 21).

**Statistical Analysis.** All analytical data are expressed as mean ± SE, and levels of serum aflatoxin-albumin adducts were compared between the beginning and end of the study and statistically analyzed by Student's *t* test. Raw data of cases and deaths were adjusted to standardized incidence and mortality using the National Population Proportion of China in 1990. Time trends analyses were performed according to the method described by Parkin *et al.* (2).

## Results

Over the past 27 years, Zhuqing Village reported 110 cases and 105 deaths from cancer. Of this total, 70 cases were HCC, resulting in 68 deaths and a standardized HCC incidence of 83.3 cases per 100,000 residents per year and a mortality of 80.9 cases per 100,000 residents per year. Time trend analysis demonstrated no significant change in incidence and mortality over this period. HCC is the leading malignancy in this village, constituting nearly 64% of total cancers, followed by stomach cancer (13.6%). The ratio of HCC in males *versus* females was ~4.8:1 (58 cases *versus* 12 cases). More strikingly, the age distribution of HCC cases revealed that most cases occurred in early middle age. The median of age of HCC onset was 34 years in males and 41 years in females.

To characterize $AFB_1$ and HBV exposure in this village, 15 males and 14 females from different households were recruited. Two of the study subjects dropped out during the 1-week period. In total, 56 blood samples, 90 raw food samples, 279 cooked food samples, which were aggregated by subject, and 567 urine samples were collected. Additionally, a consecutive 7-day dietary survey for each participant household was obtained. The daily diet of residents consisted mainly of corn and rice with side dishes including vegetables and, occasionally, pork. Almost all of the residents of the village stored ground corn for their daily food. Ground corn either in corn rice or corn porridge was consumed by all study participants, and the average daily corn consumption was 575 g for male subjects and 322 g for female subjects. Rice was consumed only at dinner; the average daily rice intake was 185 g in men and 117 g in women. Locally produced peanut oil was the sole source for cooking oil; daily intake was ~18 g per participant.

*Table 1* Detection of $AFB_1$ in food of study participants of Zhuqing Village, Fusui, Guangxi, China

| Sample | *n* | $AFB_1$ | | |
|---|---|---|---|---|
| | | Positive | % Positive | Mean ± SE (range), ppb |
| Corn | 30 | 23 | 76.7 | 23.7 ± 6.6 (0.4–128.1) |
| Peanut oil | 30 | 20 | 66.7 | 7.8 ± 3.2 (0.1–52.5) |
| Rice | 30 | 7 | 23.3 | 1.1 ± 0.3 (0.3–2.0) |

The results of the $AFB_1$ analysis in mixed food samples obtained from study participants are listed in Table 1. Corn was the major source of dietary aflatoxin exposure. Twenty-three of the 30 (76.7%) mixed ground corn samples had detectable levels of $AFB_1$ with an average of 23.7 ± 6.6 ppb (range, 0.4–128.1 ppb). Nine of the 30 ground corn samples had $AFB_1$ levels >20 ppb, and 5 of these were >50 ppb. Peanut oil was a second major source of dietary aflatoxin exposure. Twenty of 30 peanut oil samples had detectable levels of $AFB_1$ with an average of 7.8 ppb (range, 0.1–52.5 ppb). Four of these 30 samples contained >10 ppb. Rice, when compared with corn and peanut oil, was a minor source of dietary aflatoxin exposure. Although 7 of 30 (23.3%) samples had detectable levels of $AFB_1$, the average level was 1.1 ppb (range, 0.3–2.0 ppb).

Serum samples collected from study participants were analyzed for levels of aflatoxin-albumin adduct, and all 56 serum samples were positive. The average level of aflatoxin-albumin adducts for the 29 serum samples collected at the beginning of the study was 1.24 ± 0.31 pmol/mg of albumin (range, 0.81–2.41 pmol/mg of albumin). The mean level of aflatoxin-albumin adducts for 27 serum samples collected at the end of the study was 1.21 ± 0.19 pmol/mg of albumin (range, 0.92–1.67 pmol/mg of albumin). Thus, there was no statistically significant change for the adduct levels across these two time points.

Pooled urine samples collected from 27 study participants at day 7 were analyzed for urinary aflatoxin biomarkers, and the results are shown in Table 2. $AFM_1$ was detectable in 24 of 27 (88.9%) samples with an average level of 192.3 ± 65.2 ng/24-h urine (range, 0.9–1258 ng/24-h urine). AFB-mercapturic acid was also found in 24 of 27 (88.9%) samples with an average level of 103.6 ± 25.3 ng/24-h urine (range, 6.6–494.9 ng/24-h urine). AFB-$N^7$-guanine was detectable in 11 of 27 (40.7%) samples with an average level of 407.3 ± 158.7 ng/24-h urine (range, 64.9–1789.8 ng/24-h urine). Aflatoxin $P_1$ was detectable in 8 of 27 (29.6%) samples with an average level of 664.9 ± 425.4 ng/24-h urine (range, 77.3–3569.7 ng/24-h urine), and aflatoxin $Q_1$ was detectable in 7 of 27 (25.9%) samples with an average level of 92.2 ± 8.3 ng/24-h urine (range, 77.3–137.5 ng/24-h urine).

The results of serum HBV markers analyses for the 29

*Table 2* Detection of aflatoxin urinary biomarkers in study participants of Zhuqing Village, Fusui, Guangxi, China

| AF[a] biomarker | No. of samples | Positive | % Positive | AF, mean ± SE, ng/24-h urine (range) |
|---|---|---|---|---|
| $AFM_1$ | 27 | 24 | 88.9 | 192.3 ± 65.2 (0.9–1258.0) |
| AFB-NAC | 27 | 24 | 88.9 | 103.6 ± 25.3 (6.6–494.9) |
| AFB-$N^7$-Gua | 27 | 11 | 40.7 | 407.3 ± 158.7 (64.9–1789.8) |
| $AFQ_1$ | 27 | 7 | 25.9 | 92.2 ± 8.3 (77.3–137.5) |
| $AFP_1$ | 27 | 8 | 29.6 | 664.9 ± 425.4 (80.4–3569.7) |

[a] AF, aflatoxin; AFB-NAC, $AFB_1$-mercapturic acid; AFB-$N^7$-Gua, $AFB_1$-$N^7$-guanine; $AFQ_1$, aflatoxin $Q_1$; $AFP_1$, aflatoxin $P_1$.

*Table 3* Detection of serum HBV markers in study participants of Zhuqing Village, Fusui, Guangxi, China

| HBV marker | No. of samples | Positive | % |
|---|---|---|---|
| HBsAg | 29 | 4 | 13.8 |
| HBsAb[a] | 29 | 14 | 48.3 |
| HBcAb | 29 | 24 | 82.8 |
| HBeAg | 29 | 2 | 6.9 |
| HBeAb | 29 | 24 | 82.8 |

[a] HBsAb, anti-HBsAg; HBcAb, anti-HBc; HBeAg, HBe antigen; and HBeAb, anti-HBe antigen.

people recruited to the study are shown in Table 3. All participants had normal values for alanine aminotransferase and aspartate aminotransferase and were negative for $\alpha$-fetoprotein, although 4 of 29 (13.8%) were HBsAg positive. Three of these four people were men, constituting 20% (3 of 15) of the male participants in the study. Two of these male participants were positive for HBe antigen. Higher percentages (48.3–82.8%) of anti-HBs, anti-HBc, and anti-HBe marker were detected, further demonstrating a high rate of HBV infection in this population.

## Discussion

Over the past three decades, 64% of malignant tumor cases in Zhuqing Village, Fusui County were HCC, and exposure to aflatoxin and HBV was also prevalent. In this study, the rate of HBsAg positivity was found to be comparable to previous studies in the same region, in which ~12–15% of the general population was HBsAg positive. More than 90% of subsequent HCC cases tested HBsAg positive, and incidence of HCC among HBsAg carriers was close to 1% per year (11, 16, 22). Studies by Yeh and Shen (8) in the early 1980s in Fusui described a strong interaction between HBV infection and dietary aflatoxin exposure. Furthermore, a nested case-control study in Shanghai (12, 13) revealed a statistically significant synergistic increase in the relative risk of HCC with exposure to aflatoxin and HBV. This synergistic interaction between HBV and $AFB_1$ was further confirmed by several studies that used similar biomarkers for aflatoxins and HBV in other populations (14, 15).

Data from the present study showed that dietary $AFB_1$ exposure is still predominant in this population as demonstrated by the high positive rate and level of aflatoxin contamination in corn and peanut oil samples (Table 1), which were consumed daily by the study participants. The absolute $AFB_1$ daily intake, as calculated by the average contamination in the diet multiplied by the amount of food consumed, was ~14 $\mu$g/day in men and 8 $\mu$g/day in women. These levels are less than that reported for the region 15 years earlier (23), which may reflect a switch from almost 100% corn to some portions of rice as the primary dietary item within the past decade. However, this determination may underestimate the real amount of aflatoxin exposure because the heterogeneity of aflatoxin contamination of foodstuffs can lead to inter- and intraindividual variations in the population.

Human urine and serum specimens collected from the Fusui area have been used in the development and validation of molecular dosimetry and biomarkers for aflatoxin exposure. Groopman *et al.* (23) analyzed urine samples collected from a study group of 42 people in Fusui County in 1984 and found associations between urinary excretion of $AFB_1$-$N^7$-guanine adduct and $AFM_1$ and dietary $AFB_1$ intake. Gan *et al.* (24) examined formation of aflatoxin-albumin adducts in serum samples from the same group. A significant correlation ($r$ = 0.69) of aflatoxin-albumin adduct level with $AFB_1$ intake was observed. When serum aflatoxin-albumin adduct data were compared with urine AFB-$N^7$-guanine adduct level, a significant correlation was found ($r$ = 0.73). The results of the present study are consistent with these previous findings.

Finally, a recent study showed that 57% (20 of 35) of HCC cases from Fusui and neighboring areas of Guangxi Region had a G→T transversion at codon 249 of the *p53* tumor suppressor gene (25), a frequency of *p53* mutations comparable to other parts of the world with high levels of aflatoxin contamination (26, 27). Thus, the high-risk region around Fusui County is a strong candidate for implementation of primary and chemopreventive interventions.

## Acknowledgments

We are grateful to members of the study group from Fusui Liver Cancer Institute and Zhuqing Village Clinic for their help in this study and to the administrative support of Guangxi Cancer Institute, Fusui Liver Cancer Institute, Zhuqing Village, Qujiu Township, and Fusui County. We also appreciate the full cooperation of the study participants in Zhuqing Village.

## References

1. IARC Working Group on the Evaluation of Carcinogenic Risks to Humans. Some naturally occurring substances: food items and constituents, heterocyclic aromatic amines and mycotoxins. IARC Monogr. Eval. Carcinog. Risks Hum., *56:* 245–395, 1993.

2. Parkin, D., Pisani, P., and Ferlay, J. Estimates of the worldwide incidence of 18 major cancers in 1985. Int. J. Cancer, *54:* 594–606, 1993.

3. Bosch, F. X. Global epidemiology of hepatocellular carcinoma. *In:* K. Okuda and E. Tabor (eds.), Liver Cancer, pp. 13–28. New York: Churchill Livingstone, 1997.

4. Li, N-D., Lu, F-Z., Zhang, S-W., Mo, R., and Sun, X-D. Trends and prediction in malignant tumors mortality in past 20 years in China. Chin. J Oncol., *19:* 3–9, 1997.

5. Tang, Z-Y., Wu, M-C., and Xia, S-S. Primary Liver Cancer. Berlin: Springer-Verlag, 1989.

6. Harris, C. C., and Sun, T-T. Multifactorial etiology of human liver cancer. Carcinogenesis (Lond.) *5:* 697–701, 1984.

7. Wogan, G. N. Aflatoxins as risk factors for hepatocellular carcinoma in humans. Cancer Res., *52* (Suppl.)*:* 2114s–2118s, 1992.

8. Yeh, F-S., and Shen, K-N. Epidemiology and early diagnosis of primary liver cancer in China. Adv. Cancer Res., *47:* 297–329, 1986.

9. Groopman, J. D., Wang, J-S., and Scholl, P. Molecular biomarkers for aflatoxins: from adducts to gene mutations to human liver cancer. Can. J. Physiol. Pharmacol., *74:* 203–209, 1996.

10. IARC Working Group on the Evaluation of Carcinogenic Risks to Humans. Hepatitis viruses. IARC Monogr. Eval. Carcinog. Risks Hum., *59:* 45–221, 1994.

11. Yeh, F-S., Yu, M. C., Mo, C-C., Luo, S., Tong, M. J., and Henderson, B. E. Hepatitis B virus, aflatoxins, and hepatocellular carcinoma in southern Guangxi, China. Cancer Res., *49:* 2506–2509, 1989.

12. Ross, R., Yuan, J-M., Yu, M. C., Wogan, G. N., Qian, G-S., Tu, J-T., Groopman, J. D., Gao, Y-T., and Henderson, B. E. Urinary aflatoxin biomarkers and risk of hepatocellular carcinoma. Lancet, *339:* 943–946, 1992.

13. Qian, G-S., Yu, M. C., Ross, R., Yuan, J-M., Gao, Y-T., Wogan, G. N., and Groopman, J. D. A follow-up study of urinary markers of aflatoxin exposure and liver cancer risk in Shanghai, P.R.C. Cancer Epidemiol. Biomark. Prev., *3:* 3–11, 1994.

14. Lunn, R. M., Zhang, Y-J., Wang, L-Y., Chen, C-J., Lee, P-H., Lee, C-S., Tsai, W-Y., and Santella, R. M. p53 mutations, chronic hepatitis B virus infection, and aflatoxin exposure in hepatocellular carcinoma in Taiwan. Cancer Res., *57:* 3471–3477, 1997.

15. Yu, M-W., Lien, J-P., Chiu, Y. H., Santella, R. M., Liaw, Y. F., and Chen, C. J. Effect of aflatoxin metabolism and DNA adduct formation on hepatocellular carcinoma among chronic hepatitis B carriers in Taiwan. J. Hepatol., *27:* 320–330, 1997.

16. Yuan, J-M., Govindarajan, S., Henderson, B. E., and Yu, M. C. Low prevalence of hepatitis C infection in hepatocellular carcinoma (HCC) cases and population controls in Guangxi, a hyperendemic region for HCC in the People's Republic of China. Br. J. Cancer, *74:* 491–493, 1996.

17. London, W. T., Evans, A. A., McGlynn, K., Buetow, K., An, P., Gao, L-L., Lustbader, E., Ross, E., Chen, G-C., and Shen, F-M. Viral, host and environmental risk factors for hepatocellular carcinoma: a prospective study in Haimen City, China. Intervirology, *38:* 155–161, 1995.

18. Kensler, T. W., He, X., Otieno, M., Egner, P. A., Jacobson, L. P., Chen, B., Wang, J-S., Zhu, Y-R., Zhang, B-C., Wang, J-B., Wu, Y., Zhang, Q-N., Qian, G-S., Kuang, S-Y., Fang, X., Li, Y-F., Yu, L-Y., Prochaska, H. J., Davidson, N. E., Gordon, G. B., Gorman, M. B., Zarba, A., Enger, C., Munoz, A., Helzlsouer, K. J., and Groopman, J. D. Oltipraz chemoprevention trial in Qidong, People's Republic of China: modulation of serum aflatoxin albumin adduct biomarkers. Cancer Epidemiol. Biomark. Prev., *7:* 127–134, 1998.

19. Wang, J-S., Shen, X., He, X., Zhou, Y-R., Zhang, B-C., Wang, J-B., Qian, G-S., Kuang, S-Y., Zarba, A., Egner, P. A., Jacobson, L. P., Munoz, A., Helzlsouer, K. J., Groopman, J. D., and Kensler, T. W. Protective alterations in phase 1 and 2 metabolism of aflatoxin B1 by Oltipraz in residents of Qidong, People's Republic of China. J. Natl. Cancer Inst. (Bethesda), *91:* 347–354, 1999.

20. Trucksess, M. W., and Wood, G. E. Recent methods of analysis for aflatoxins in foods and feeds. *In:* D. L. Eaton and J. D. Groopman (eds), The Toxicology of Aflatoxins: Human Health, Veterinary, and Agricultural Significance, pp. 409–431. San Diego: Academic Press, Inc., 1994.

21. Wang, J-S., Qian, G-S., Zarba, A., He, X., Zhu, Y-R., Zhang, B-C., Jacobson, L., Gange, S. J., Munoz, A., Kensler, T. W., and Groopman, J. D. Temporal patterns of aflatoxin-albumin adducts in hepatitis B surface antigen positive and negative residents of Daxin, Qidong County, People's Republic of China. Cancer Epidemiol. Biomark. Prev., *5:* 253–261, 1996.

22. Yeh, F-S. Primary liver cancer in Guangxi. *In:* Z-Y. Tang, M-C. Wu and S-S. Xia (eds.), Primary Liver Cancer, pp. 223–236. Berlin: Springer-Verlag, 1989.

23. Groopman, J. D., Zhu, J-Q., Donahue, P. R., Pikul, A., Zhang, L-S., Chen, J-S., and Wogan, G. N. Molecular dosimetry of urinary aflatoxin DNA adducts in people living in Guangxi Autonomous Region, People's Republic of China. Cancer Res., *52:* 45–51, 1992.

24. Gan, L-S., Skipper, P. L., Peng, X-C., Groopman, J. D., Chen, J-S., Wogan, G. N., and Tannenbaum, S. R. Serum albumin adducts in the molecular epidemiology of aflatoxin carcinogenesis: correlation with aflatoxin B1 intake and urinary excretion of aflatoxin M1. Carcinogenesis (Lond.), *9:* 1323–1325, 1988.

25. Deng, Z., Ma, Y., Pan, L., and Peng, H. A molecular epidemiologic marker of hepatocellular carcinoma from aflatoxin B1 contaminated area in the southwest of Guangxi. Chin. J. Cancer Res., *9:* 167–169, 1997.

26. Hsu, I. C., Metcalf, R. A., Sun, T., Wesh, J. A., Wang, N. J., and Harris, C. C. Mutational hotspot in the p53 gene in human hepatocellular carcinomas. Nature (Lond.), *350:* 427–428, 1991.

27. Bressac, B., Kew, M., Wands, J., and Ozturk, M. Selective G to T mutations of p53 gene in hepatocellular carcinoma from Southern Africa. Nature (Lond.), *350:* 429–431, 1991.

Cancer Epidemiology, Biomarkers & Prevention

# Hepatocellular Carcinoma and Aflatoxin Exposure in Zhuqing Village, Fusui County, People's Republic of China

Jia-Sheng Wang, Tiren Huang, Jianjia Su, et al.

*Cancer Epidemiol Biomarkers Prev* 2001;10:143-146.