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The Molecular Pathogenesis of Aflatoxin with Hepatitis B Virus-Infection in Hepatocellular Carcinoma

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1. Introduction

Hepatocellular carcinoma (HCC) is the fifth most common cancer worldwide and represents the third cause of mortality among deaths from cancer (Semela & Heim, 2011; EI-Serag & Rudolph, 2007; Yuen et al, 2009; Lodato et al, 2006; Hainaut & Boyle, 2008). There are upwards of 600, 000 new cases each year in Asia and sub-Saharan Africa, where populations suffer both from a high prevalence of hepatitis B virus (HBV) infection and largely uncontrolled aflatoxin exposure in food, and more than 200, 000 HCC-related deaths annually in the People's Republic of China alone (Kew, 2002; Wang et al, 2002). Aflatoxins may play a causative role in 5%-28% of all global HCC cases (Liu & Wu, 2010).

Aflatoxin is a kind of mycotoxins produced by the mold Aspergillus flavus, which can be found in legumes, corns, soybeans, rice, milk, and cheese throughout their lives. Aflatoxin B1 (AFB1) is regarded as a class I carcinogen by the World Health Organization (Henry et al, 2001; López et al, 2002). Aflatoxin B1, the most commonly occurring and potent of the aflatoxins is associated with a specific AGG to AGT transversion mutation at codon 249 of the p53 gene in human HCC, providing mechanistic support to a causal link between exposure and disease (Goldman & Shields, 2003; Sugimura, 2000; Wild & Montesano, 2009). Prospective epidemiological studies have shown a more than multiplicative interaction between HBV and aflatoxins in terms of HCC risk. In this chapter, the available evidence for the mechanism of aflatoxin with HBV-infection in HCC will be reviewed.

2. Mechanisms of aflatoxins in HCC

Aflatoxins, first described in the early 1960s, are the most toxic and carcinogenic compounds among the known mycotoxins. They are produced by several Aspergillus species and consist of at least 16 structurally related furanocoumarins, of which AFB1, AFB2, AFG1, and AFG2 are the four most abundant aflatoxins. The designation B and G came from the Blue or Green fluorescence emitted by these compounds under long-wave ultraviolet light, respectively (McLean & Dutton, 1995). Human exposure to aflatoxins is mainly through consumption of contaminated staples, such as maize and pea-nuts. Contaminations are often the result of inappropriate storage of grain, which leads to the infestation by aflatoxin-

producing fungi (Williams et al, 2004). Liver is the primary target organ for acute and chronic injury. Ingestion of high doses of aflatoxin over a short time period can result in acute aflatoxicosis, with symptoms such as hemorrhagic necrosis of the liver, bile duct proliferation, edema and lethargy (Williams et al, 2004). Chronic exposure to low or moderate doses of aflatoxin may lead to the development of HCC.

AFB1 is a potent human carcinogen (IARC, 1993), which occurs in the low or sub micrograms per kilogram range and is regulated by legislation in the European Union (EU) at 2 µg/kg in foods for direct human consumption (European Community, 2006). Several studies have evaluated an association between the risk for HCC and exposure to AFB1. A prospective case-control study from China mainland showed that individuals with the presence of urinary aflatoxin biomarkers had a significantly increased risk of HCC after adjusting for HBV surface antigen (HBsAg) seropositivity and cigarette smoking (Qian et al, 1994). These data were further supported by a community-based cohort study from Taiwan which found that elevated AFB1 exposure measured by detectable AFB1-albumin adducts was an independent risk factor for HCC after adjustment for other important confounding factors (odds risk : 5.5, 95% confidence interval : 1.2-24.5) (Chen et al, 1996a).

2.1 Metabolism of AFB1

The cytochrome P450 (CYP) enzymes are a superfamily of hemeproteins that are important in the oxidative, peroxidative and reductive metabolism of endogenous compounds and participate in the chemical carcinogenesis process (Gonzalez & Lee, 1996). AFB1 is activated by CYP enzymes, mainly CYP 1A2 and 3A4, to form four major metabolites: AFM1, AFQ1, AFB1-endo-8, 9-epoxide and AFB1-exo-8, 9-epoxide. AFM1 and AFQ1 are polarmolecules; AFB1-endo-8, 9-epoxide reacts poorly with DNA (McLean & Dutton, 1995; Guengerich et al, 1998; Iyer et al, 1994). The overall contribution of these enzymes to AFB1 metabolisms in vitro depends on the affinity of the enzyme but in vivo it also depends on expression levels in human liver where CYP 3A4 is predominant. Expression of CYP 1A1/2 and 3A4 in liver tissues of hepatocellular carcinoma cases and controls was detected and their relationship to HBV and AFB1-DNA adducts was also investigated (Pfeifer et al, 1993). For CYP 3A4, in contrast to control tissues, there was a significant association with AFB1-DNA adducts in tumor and adjacent non-tumor tissues in patients with HCC. Human beings show large interindividual variations in metabolism activities that lead to different susceptibilities to the genotoxic actions of carcinogens (Zhang et al, 2000).

A model using human liver epithelial cell lines stably expressing cytochrome P450 cDNA revealed that CYP 1A2 and CYP 3A4 both contribute to the formation of AFB1-induced p53 mutation whereas CYP 2A6 does not appear to play a significant role (Macé et al, 1997). In an in vitro study, inhibition of CYP 1A2 and CYP 3A4 by oltipraz, a drug which has been reported to inhibit AFB1 activation in human hepatocytes, was shown (Langouët et al, 1995).

AFB1 can also be metabolized by NADPH-dependent reductase into a carcinogenic metabolite aflatoxicol (AFL) (Salhab & Edwards, 1977; Wong & Hsieh, 1976). AFL acts as a reservoir of AFB1, prolonging its lifetime in body, as it can be reconverted to AFB1, which then can be further metabolized. Formation of AFL does not decrease the toxicity of AFB1 because it can also bind to DNA and is as potent carcinogen like AFB1 (Bailey et al, 1994).

Glutathione S-transferase (GST) is a family of conjugation enzymes involved in the metabolism of exogenous and endogenous lipophilic compounds for their excretion and detoxification. For AFB1, the detoxification pathway is via GST-mediated conjugation with

reduced glutathione (GSH) to form AFB1 exo- and endo-epoxide GSH conjugates (Guengerich et al, 1998; Raney et al, 1992; Johnson WW, 1997a). Members of the GST family, such as GST-μ (GSTM1) and GST-θ (GSTT1), are important candidates for involvement in susceptibility to AFB-associated HCC, because they may regulate an individual's ability to metabolize the ultimate carcinogen of aflatoxin, the exo-epoxide (Johnson WW, 1997a). Epidemiological studies have suggested that genetic polymorphisms in AFB1 metabolizing enzymes are factors in individual susceptibility to aflatoxin-related HCC (McGlynn et al, 1995; Chen et al, 1996b). GSTM1 genotype can be categorized into two classes: the homozygous deletion genotype (GSTM1null genotype) and genotypes with one or two alleles present (GSTM1 non-null genotype); GSTT1 can also be deleted (Pemble et al, 1994; Rebbeck et al, 1999). Carriers of GSTM1 and GSTT1 homozygous null genotypes lack of the corresponding enzyme activities (Pemble et al, 1994). Chen et al (1996b) documented a biological gradient between serum AFB1-albumin adduct levels and HCC risk among chronic HBsAg carriers who had null GSTM1 and GSTT1 genotypes but not among those who had non-nullgenotypes in a Taiwan population. Wild et al (2000) reported an association between the GSTM1 null genotype and AFB1-albumin adducts in a Gambian population, although the association was restricted to people who were not infected with HBV. The effect of aflatoxin exposure on HCC risk was also more pronounced among chronic HBsAg carriers with the GSTT1 null genotype than those who were non-null (Sun et al, 2001). Based on the above studies conducted in different places and others not reviewed, whether or not there are interactions among AFB1, HBV infection and GSTs genotypes in the development of HCC is still controversial.

2.2 Damage to DNA

The main genotoxic product of AFB1 metabolites is AFB1-exo-8, 9-epoxide, an unstable metabolite which reacts with DNA, and forms adducts with a yield of 98% (Johnson & Guengerich, 1997). This reactivity is 1000-fold greater than that of its endo-isomer (Iyer et al, 1994). AFB1-exo-epoxide can intercalate between the bases in DNA, and can form covalent bonds with DNA by electrophilic attack on the N7 position in guanines, leading to the formation of the adduct 8, 9-dihy-dro-8-(N7-guanyl)-9-hydroxyaflatoxin B1 (AFB1-N7-Gua). This adduct is fairly unstable due to a positive charge on the imidazole ring. As a result, AFB1-N7-Gua adducts undergo processing reactions that include either depurination to form an apurinic (AP) site, or opening of the imidazole ring to form a more stable, AFB1–formamidopyrimidine (AFB1–FAPY) adduct (Smela et al, 2001). It has been proposed that AFB1 can also cause DNA damage through production of reactive oxygen species (Petitjean et al, 2007). AFB1-DNA bulky adducts, including AFB1-N7-Gua and AFB1–FAPY, are repaired mainly by Nucleotide Excision Repair (NER) (Bedard & Massey, 2006) but all AFB1-DNA adducts are not repaired with the same efficiency. The AP site is repaired by Base Excision Repair (BER) (Bedard & Massey, 2006). AFB1-induced DNA lesions that escape DNA repair can result in mutations (Smela et al, 2001). AFB1-N7 -Gua, AFB1-FAPY and AP sites are all candidate precursors of mutations, predominantly induce G-T transversions.

As this is the pathway of its toxicological effects, the two most extensively studied biomarkers are the urinary aflatoxin N-7-guanine adduct, which results from AFB1 reaction with DNA, and aflatoxin-lysine adduct, which is obtained by digestion of the aflatoxin–albumin adduct occurring in serum (Shephard, 2009). Early experimental studies around 1980 demonstrated that the major aflatoxin-nucleic acid adduct, AFB1-N7-Gua, was excreted

exclusively in the urine of exposed rats (Bennett et al, 1981; Egner et al, 2006). The serum aflatoxin-albumin adduct was also examined as a biomarker of exposure because the longer half-life of albumin would be expected to integrate exposures over longer time periods, i.e., months instead of days. Studies in experimental models found that the formation of aflatoxin-DNA adducts in liver, urinary excretion of the aflatoxin-nucleic acid adduct, and formation of the serum albumin adduct were highly correlated events (Groopman, 1994).

2.3 AFB1 inducing p53 mutaion (G-T transversions)

AFB1 covalently binds to guanine and cytosine residues of DNA both in vivo and in vitro and forms AFB1-DNA adducts, it also forms RNA and protein adducts impairing DNA, RNA and ultimately protein synthesis (Santella et al, 1998; Meneghini & Schumacher, 1977; Amstad & Cerutti, 1983). The presence of AFB1-DNA adducts can contribute to genetic alterations involved in the development of HCC. In 1977, Lin et al (1977) reported that adduct formation by metabolically activated reactive intermediates with hepatocyte DNA could lead to mutations in the host genome. The p53 tumor suppressor gene is the most frequently mutated gene in human cancers. Two groups found at the same time, that mutations of the p53 gene on chromosome 17 are frequent in HCC and a point mutation at the third position of codon 249 resulting in a G:C to T:A transversion was common in HCC tissues which were collected in China and Africa (Hsu et al, 1991; Bressac et al, 1991).

This mutation is present in up to 50% of patients with HCC who are indigenous to geographic regions with high exposure to AFB1(Bressac et al, 1991; Coursaget et al, 1993; Hsu et al, 1991; Ming et al, 2002). In contrast, it is absent in patients with HCC from regions with low exposure to AFB1 as well as in cancers other than HCC(Aguilar et al, 1994; Challen et al, 1992). The mutant allele is defined as R249S and the mutant protein as p.R249S. R249S accounts for more than 90% of TP53 mutations found in HCC cases from regions with high aflatoxin exposure levels, including Qidong in China and several sub-Saharan African countries. This percentage drops to around 30–40% in Taiwan and Hong Kong, where aflatoxin exposure levels are considered to be moderate. Among HCCs from low exposure regions, such as the United States and Japan, R249S only accounts for less than 6% of TP53 mutations(Petitjean et al, 2007).

This hot spot mutation in HCC from regions with high levels of dietary aflatoxins links this genetic change to exposure to aflatoxins. In recent years, the p53 codon 249 mutation has also been detected in plasma or serum DNA of HCC patients(Kirk et al, 2000; Jackson et al, 2001; Kuang et al, 2005). This mutated DNA may serve as a biomarker of exposure to AFB1 and for detection of early HCC(Jackson et al, 2001).

2.4 P53 mutation in HCC

The crystal structure of the wild-type p53 core domain in complex with DNA reveals that the arginine at position 249 provides four essential bridges to other residues in the L2 and L3 loops of the DNA-binding surface, but does not make direct contacts with DNA(Cho et al, 1994). Replacement of the arginine by a serine thermodynamically destabilizes the core domain and impairs its DNA-binding capacity(Bullock et al, 2000). Nuclear Magnetic Resonance (NMR) showed that the general structure of p.R249S core domain was similar to that of wild-type p53, but that the mutant demonstrated local structural distortion around position 249 in the L3 loop, with increased flexibility of the beta-sandwich scaffold(Friedler et al, 2004). The presence of a serine at position 249 induces a reorientation of M243. In the wild-type core domain, M243 is exposed to solvent when the protein is not bound to DNA

but is buried within at the interface between p53 oligomers when complexed with target DNA. In T-p53C-R249S, M243 displaces M246 from its buried location within a hydrophobic pocket of the zinc-binding region, leading to the formation of a short alpha-helix and a local conformational change that displaces the DNA-contact residue R248. This effect may explain the loss of the DNA-binding capacity of p.R249S.

p.R249S has lost DNA-binding and transactivation capacities towards most, if not all, promoters that contain p53 consensus binding sequence. In yeast assays, its residual activity towards p53-dependent promoters is of less than 20% of that of wild-type p53, similar to most other ''hotspot" p53 mutants. At biological level, Ponchel and colleagues(1994) have reported an increase in colony formation but not of tumorigenicity in nude mice upon transfection of R249S in Hep3B (p53-null hepatoblastoma cells). Using non-immortalized human epithelial cells, Schleger and colleagues (1999) found that p.R249S expression increased the size but not the number of colonies in clonogenicity assays, but did not prolong the lifespan of the cells. Two studies have addressed the effects of transgenic expression of the murine homologous p.R246S in mouse liver. Yin and colleagues (1998) have reported enhanced cell cycle activity in the liver due to an increased entry into G1 phase. Thus, none of the above mechanisms provide a convincing functional explanation for the apparent selection of p.R249S in aflatoxin-induced HCC.

Tumor-derived p53 mutant proteins contribute to carcinogenesis through three overlapping mechanisms (Gouas et al, 2010): loss of wild-type p53 trans-activation function (loss of function); capacity to inhibit the activity of wild-type p53 (dominant-negative effect) and possible 'gain-of-function' effects, by which mutant proteins have acquired new, pro-oncogenic properties (Brosh & Rotter, 2009; Oren & Rotter, 2010). Several mechanisms underlying such gain-of-function effects have been described, including: transactivation of gene enhancing proliferation, inhibiting apoptosis or chemoresistance or increasing invasiveness, inflammation and angiogenesis (Brosh et al., 2009); interaction with various proteins, in particular TAp63 and TAp73, the products of two genes related to TP53 that exert differentiation and growth suppressive effects during development and morphogenesis (Bergamaschi et al, 2003; Gaiddon et al, 2001; Marin et al, 2000; Strano et al, 2002). It is not clear whether p.R249S exerts such gain-of-function effects which needs further studies.

2.5 Others changes

Moreover, it has been recently demonstrated that AFB1-albumin adducts in patients with HCC correlate significantly with the presence of plasma DNA hypermethylation and mutations in the p16 and p53 tumor suppressor genes (Zhang et al, 2006). AFB1-induced HCC in Fischer 344 rats showed activating mutations in codon 12 of K-ras but in human HCC, the incidence of point mutation of K-ras and N-ras oncogenes was low (Tsuda et al, 1989). In an in vitro study, AFB1 interfered with the molecular mechanisms of cell cycle regulation (Ricordy et al, 2002). Gursoy-Yuzuqullu (2011) showed that genotoxic doses of AFB1 induce an incomplete and inefficient checkpoint response in human cells. This defective response may contribute to the mutagenic and carcinogenic potencies of aflatoxins. AFB1 also induced mitotic recombination (Stettler & Sengstag, 2001) and minisatellite rearrangements (Kaplanski et al, 1997). Mitotic recombination and genetic instability may therefore be alternative mechanisms by which aflatoxin contributes to genetic alterations in HCC (Wild & Turner 2002).

Long and colleagues(2009) report that there were interactions between the genetic polymorphism of XPD codon 751 and AFB1-exposure years, and imply that this

polymorphism may have functional significance in HCC induced by AFB1.While XPD protein, encoded by XPD gene, is a DNA-dependent ATPase/helicase that is associated with the TFIIH transcription-factor complex, and plays a role in NER pathway. During NER, XPD participates in the opening of the DNA helix to allow the excision of the DNA fragment containing the damaged base (Benhamou & Sarasin, 2002; Manuguerra et al, 2006). These results suggest that the genotypes of XPD with codon 751 Gln alleles may increase the risk of AFB1-related HCC and the NER pathway may play an important role in the mechanism of action of this genotoxin.

3. Interaction between AFB and HBV in HCC

The marked worldwide heterogeneity of HCC incidence in HBV-endemic regions might be related, at least in part, to aflatoxin exposure (Yu &Yuan 2004; Yuen et al, 2009; Lodato et al, 2006). It should be stressed that areas with high exposure to AFB1 are also characterized by a high prevalence of HBV infection. AFB1 is independent of the risk conferred by HBV, however concomitant exposure to both HBV and AFB1 markedly increases the risk of HCC. Ross et al (1992) formerly demonstrated a synergistic interaction between HBV and AFB1 in the development of liver cancer. Subsequently, Sun et al (1999) followed a cohort of Chinese men with chronic HBV for 10 years and found that the relative risk of HCC was significantly increased in subjects with detectable AFM1 levels. Aflatoxin exposure in association with HBV infection induces a 60-fold increase in risk of HCC(RR: 59.4, 95% CI: 16.6-212.0), while aflatoxin alone increases the risk fourfold.(Yu &Yuan 2004; Ming et al, 2002; Turner et al, 2002). In a case-control study in The Gambia, the codon 249ser mutation was examined in the plasma of HCC cases, cirrhosis patients and controls. The presence of both the codon 249ser mutation and HBV infection was associated with an OR = 399 (95% CI: 48.6–3270) (Kirk et al, 2004; Kirk et al, 2005). HBV transgenic mice overexpressing HBsAg in the liver showed more HCC than non-transgenic littermates when exposed to AFB1 (Sell & Heim, 1991).

Aflatoxin exposures multiplicatively increase the risk of liver cancer in people chronically infected with hepatitis B virus (HBV), which illustrates the deleterious impact that even low toxin levels in the diet can have on human health (Qian et al, 1994; Ross et al, 1992; Wang LY et al, 1996). Ghebranious and Sell (1998) have investigated the induction of HCC in p.R246S transgenic mice: they found that the presence of p.R246S enhanced aflatoxin-induced formation of liver tumor in conjunction with a HBsAg transgene.

Recently, a large-scale quantitative analysis of TP53 mutations in the serum of healthy individuals from The Gambia, a country with high aflatoxin exposure levels and endemic for HBV infection, has been performed. The study revealed the presence of R249S DNA in the serum or plasma of apparently healthy subjects, with a seasonal variation that recapitulated the known variation in exposure to aflatoxin. Moreover, the presence and mean concentrations of R249S DNA in the plasma or serum were significantly associated with HBV carriage, with 44% of carriers showing detectable levels of the mutation compared to only 24% of non-carriers. These results are consistent with the hypothesis that HBV infection may specifically contribute to the acquisition and/or the retention of R249S mutation in hepatocytes (Gouas et al, 2009).

However, the biology underlying this statistical interaction is not fully understood. Several lines of evidence suggest that HBV infection may enhance the mutagenic effects of AFB1. First, the presence of the virus may interfere with AFB1 metabolism. The expression of

CYP450 enzymes that metabolize AFB1 is increased in HBsAg-transgenic mice (Kirby et al, 1994). Moreover, Gambian children and adolescents chronically infected with HBV show higher concentrations of AFB1 adducts than uninfected individuals (Kew, 2003). Second, viral replication and chronic inflammation due to infection induces oxidative stress in hepatocytes, which may contribute to the mutation load at codon 249 (Hussain et al, 1994) .Third, HBV may also promote these mutations through indirect mechanisms, e.g. by inducing chronic inflammation which, in turn, increases hepatocyte turnover rate and the risk of acquisition of a mutation such as R249S. Another possible mechanism of interference between HBV and mutagenesis at codon 249 may occur at the level of cellular DNA repair. AFB1–DNA adducts, in particular, AFB1-N7-Gua and AFB1-FAPY, are repaired through the NER pathway. The viral antigen HBx can decrease NER efficiency, probably through physical interaction with wild-type p53 and/or with the components of the NER machinery, such as XPB and XPD DNA helicases (Jia et al, 1999). The inhibition of NER by HBx may therefore lead to the persistence of AFB1-DNA adducts, which increases the risk for mutations. However, this mechanism does not account for the specific retention of R249S, unless some form of sequence-specificity in preferential repair of DNA at other positions than the third base of codon 249 is concerned. The high rate of R249S mutation may also be the consequence of the fact that this mutant provides some kind of selective advantage to infected liver cells. This may lead to the clonal expansion of p.R249S-expressing hepatocytes and may increase the probability for further accumulation of other cancer-prone mutations. This property may be due to a special capacity of p.R249S to form complexes with selected viral antigens. It has been reported that the HBx protein physically associates with p53 and apparently blocks its normal function in vitro and in vivo (Feitelson et al, 1993; Wang XW et al, 1994). In agreement with this view, in mice carrying HBx transgene, the X protein may bind to p53 and induce its retention into the cytoplasm, thus functionally inactivating its function (Ueda et al, 1995; Lin Y et al, 1997). Whether this interaction is of relevance for mutant p53, and in particular for p.R249S, is not clear. Of note, those HBx transgenic mice have a higher levels of G: C–T:A transversions induced by AFB1 than AFB1-exposed wild-type mice (Madden et al, 2002).

There maybe exists other mechanisms, including the predisposition of HBV-infected hepatocytes to aflatoxin induced DNA damage; an increase in susceptibility to chronic HBV infection in aflatoxin-exposed individuals; and oxidative stress exacerbated by co-exposure to aflatoxins and chronic hepatitis infection. Some epidemiological studies have clearly shown that two factors of importance in determining the risk of HCC in the natural history of HBV infection are the age at primary infection and the presence of serum HBeAg or HBV DNA, biomarkers of active viral replication, in patients with chronic active hepatitis (Yang et al, 2002). In addition, hepatitis B virus X (HBx) protein affects various cellular functions relevant to cancer development, namely p53 and oxidative DNA damage (Hussain et al, 2007). This correlation may be indicative of AFB1 exposure itself inducing oxidative stress, HBV could predispose hepatocytes to the carcinogenic action of aflatoxins. For example, human liver epithelial cells, expressing wild-type p53 and transfected with HBx gene were more sensitive to the cytotoxic action of AFB1-8, 9-epoxide than were the parent cells (Sohn et al, 2000). The HBx expressing cells were also more prone to apoptosis and to induction of mutations at codon 249 of the p53 gene. Aflatoxin exposure may alter the effects of the hepatitis virus infection, perhaps affecting susceptibility to infection or viral replication. HepG2 cells transfected with re-circularised HBV and treated with AFB1 showed a 2-3-fold increase in HBsAg at 96 hours post-treatment (Banerjee et al, 2000). DNA damage can also

increase viral DNA integration into the host genome (Dandri et al, 2002) and it is possible that AFB1 could exert this effect directly, or indirectly via the oxidative stress mentioned above. In addition, the recent study in Guangxi, China (Xu et al, 2010) found additive effects of HBV BCP mutations and high serum AFB1-lysine adduct level on the risk of developing HCC.

A recent study provided the first evidence that cirrhosis may play a contributory role in the pathogenesis of AFB1-induced HCC (Jiang et al, 2010). Kuniholm (2008) and co-workers used an ultrasound-based method to diagnose the presence of cirrhosis in 97 black Africans. A score of at least 7 out of a possible 11 points on the ultrasound-based scale was the criterion for the diagnosis of cirrhosis. This method has 77.8% sensitivity and 92.5% specificity in comparison with liver biopsy in identifying cirrhosis in HBV-infected patients (Lin DY et al, 1993; Hung et al, 2003). Three hundred and ninety seven individuals with no evidence of liver disease and a normal serum AFB1 concentration served as controls. Long-term exposure to AFB1 was assessed in the patients with cirrhosis and the controls on the basis of two observations: A history of lifetime groundnut (peanut) intake or the finding in the serum of a genetic marker of heavy exposure to AFB1, the 249 ser p53 mutation. An increased relative risk of cirrhosis development of 2.8 (95% confidence interval 1.1-7.7) was calculated using a history of life-time dietary intake of groundnuts as the criterion for significant exposure, and of 3.8 (95% confdence interval 1.5-9.6) using the finding of the 249 ser p53 mutation in serum as the criterion, allowing for the possible confounding effect of HBV and HCV infection in each instance.

4. Conclusion

Further understanding of the interaction of HBV infection, genetic variation and exposure to environmental chemical carcinogens will help to elucidate mechanisms of human hepatocarcinogenesis and develop more effective strategies for HCC prevention. At present, simple, low-technology, and inexpensive practices can result in a striking decrease in exposure to AFB1.

5. References

Aguilar F, Harris CC, Sun T, et al. (1994) Geographic variation of p53 mutational profle in nonmalignant human liver. Science, 1994 May 27; 264(5163): 1317-1319

Amstad P, Cerutti P.(1983) DNA binding of aflatoxin B1 by co-oxygenation in mouse embryo fibroblasts C3H/10T1/2. Biochem Biophys Res Commun, 1983 May 16; 112(3): 1034-1040

Bailey GS, Loveland PM, Pereira C, et al.(1994) Quantitative carcinogenesis and dosimetry in rainbow trout for aflatoxin B1 and aflatoxicol, two aflatoxins that form the same DNA adduct. Mutat Res.1994 Aug;313(1):25-38

Banerjee R., Caruccio L., Zhang Y.J, et al. (2000) Effects of carcinogen-induced transcription factors on the activation of hepatitis B virus expression in human hepatoblastoma HepG2 cells and its implication on hepatocellular carcinomas, .Hepatology 2000 Aug 32(2):367–374.

Bedard L.L., Massey T.E. (2006) Aflatoxin B1-induced DNA damage and its repair. Cancer Lett. 2006 Sep 28; 241(2):174–183.

Benhamou S, Sarasin A.(2002) ERCC2/XPD gene polymorphisms and cancer risk. Mutagenesis 2002 Nov; 17(6):463-469.

Bennett RA, Essigmann JM, Wogan GN. (1981). Excretion of an aflatoxin-guanine adduct in the urine of aflatoxin B1-treated rats. Cancer Res. 1981 Feb;41(2):650–654

Bergamaschi D, .Gasco M, Hiller L, et al. (2003) p53 polymorphism influences response in cancer chemotherapy via modulation of p73-dependent apoptosis. Cancer Cell, 2003 Apr;3(4):387–402.

Bressac B, Kew M, Wands J, et al.(1991) Selective G to T mutations of p53 gene in hepatocellular carcinoma from southern Africa. Nature. 1991 Apr 4; 350(6317): 429-431

Brosh R, Rotter V.(2009) When mutants gain new powers: news from the mutant p53 field. Nat. Rev. Cancer.2009 Oct;9(10), :701–713.

Bullock A.N, Henckel J., Fersht A.R.(2000) Quantitative analysis of residual folding and DNA binding in mutant p53 core domain: definition of mutant states for rescue in cancer therapy, Oncogene.2000 Mar 2; 19 (10):1245–1256.

Challen C, Lunec J, Warren W, et al.(1992) Analysis of the p53 tumor-suppressor gene in hepatocellular carcinomas from Britain. Hepatology 1992 Dec; 16(6): 1362-1366

Chen CJ, Wang LY, Lu SN, et al.(1996a) Elevated aflatoxin exposure and increased risk of hepatocellular carcinoma. Hepatology. 1996 Jul; 24(1): 38-42.

Chen CJ, Yu MW, Liaw YF, et al.(1996b). Chronic hepatitis B carriers with null genotypes of glutathione S-transferase M1 and T1 polymorphisms who are exposed to aflatoxin are at increased risk of hepatocellular carcinoma. Am J Hum Genet. 1996 Jul; 59(1): 128-134

Cho Y, Gorina S, Jeffrey P.D., et al.(1994) Crystal structure of a p53 tumor suppressor-DNA complex: understanding tumorigenic mutations. Science. 1994 Jul 15;265 (5170) :346–355.

Coursaget P, Depril N, Chabaud M, et al.(1993). High prevalence of mutations at codon 249 of the p53 gene in hepatocellular carcinomas from Senegal. Br J Cancer. 1993 Jun; 67(6): 1395-1397

Dandri M, Burda M.R, Bürkle A, et al.(2002) Increase in de novo HBV DNA integrations in response to oxidative DNA damage or inhibition of poly(ADP-ribosyl)ation. Hepatology.2002 Jan;35 (1): 217–223.

Egner PA, Groopman JD, Wang JS, et al.(2006). Quantification of aflatoxin-B1-N7-Guanine in human urine by high-performance liquid chromatography and isotope dilution tandem mass spectrometry. Chem Res Toxicol. 2006 Sep;19(9):1191–1195

EI-Serag HB, Rudolph KL.(2007).Hepatocellular carcinoma: epidemiology and molecular carcinogenesis. Gastroenterology. 2007 Jun; 132(7): 2557–2576.

European Commission Regulation (EC) No 1881/2006 of 19 December 2006. Off J Eur Union L364/5

Feitelson M.A., Zhu M., . Duan L.X, et al.(1993).Hepatitis B x antigen and p53 are associated in vitro and in liver tissues from patients with primary hepatocellular carcinoma. Oncogene.1993 May;8(5): 1109–1117.

Friedler A., DeDecker B.S, Freund S.M., et al.(2004) Structural distortion of p53 by the mutation R249S and its rescue by a designed peptide: implications for mutant conformation. J Mol Biol.2004 Feb 6; 336 (1): 187–196

Gaiddon C., Lokshin M., Ahn J., et al. (2001) A subset of tumor-derived mutant forms of p53 down-regulate p63 and p73 through a direct interaction with the p53 core domain. Mol Cell Biol. 2001 Mar; 21(5):1874–1887.

Ghebranious N., Sell S.(1998).The mouse equivalent of the human p53ser249 mutation p53ser246 enhances aflatoxin hepatocarcinogenesis in hepatitis B surface antigen transgenic and p53 heterozygous null mice. Hepatology.1998 Apr; 27 (4): 967–973.

Goldman R, Shields PG. (2003).Food mutagens. J Nutr. 2003 Mar;133 Suppl 3:965S–973S.

Gonzalez FJ, Lee YH.(1996) Constitutive expression of hepatic cytochrome P450 genes. FASEB J. 1996 Aug; 10(10): 1112-1117

Gouas D, Shi H, Hainaut P. (2009).The aflatoxin-induced TP53 mutation at codon 249 (R249S): biomarker of exposure, early detection and target for therapy. Cancer Lett. 2009 Dec 1;286(1):29-37

Gouas DA, Shi H, Hautefeuille AH, et al.(2010) Effects of the TP53 p.R249S mutant on proliferation and clonogenic properties in human hepatocellular carcinoma cell lines: interaction with hepatitis B virus X protein. Carcinogenesis.2010 Aug;31(8):1475-1482

Gursoy-Yuzuqullu O, Yuzugullu H, Yilmaz M, et al.(2011) Aflatoxin genotoxicity is associated with a defective DNA damage response by passing p53 activation. Liver Int.2011 Apr; 31(4):561-571

Groopman JD.1994. Molecular dosimetry methods for assessing human aflatoxin exposures. In The Toxicology of Aflatoxins: Human Health, Veterinary, and Agricultural Significance, ed. DL Eaton, JD Groopman, pp. 259–79. San Diego: Academic

Guengerich FP, Johnson WW, Shimada T, et al. (1998) Activation and detoxication of aflatoxin B1. Mutat Res.1998 Jun 18;402 (1-2):121–128.

Hainaut P, Boyle P. (2008).Curbing the liver cancer epidemic in Africa. Lancet. 2008 Feb 2; 371(9610): 367–368.

Henry S.H., Whitaker T., Rabbani I., et al. (2001) Aflatoxin M1: Joint ExpeCommittee on Food Additives (JECFA). Hamilton, Ontar Canada

Hsu IC, Metcalf RA, Sun T, et al.(1991) Mutational hotspot in the p53 gene in human hepatocellular carcinomas. Nature. 1991 Apr 4; 350(6317): 427-428

Hung CH, Lu SN, Wang JH, et al.(2003) Correlation between ultrasonographic and pathologic diagnoses of hepatitis B and C virus-related cirrhosis. J Gastroenterol. 2003; 38(2): 153-157

Hussain SP, Aguilar F, Amstad P, et al, Oxy-radical induced mutagenesis of hotspot codons 248 and 249 of the human p53 gene, Oncogene.1994 Aug; 9 (8): 2277–2281.

Hussain SP, Schwank J, Staib F, et al.(2007) TP53 mutations and hepatocellular carcinoma: insights into the etiology and pathogenesis of liver cancer. Oncogene.2007 Apr 2; 26 (15): 2166–2176

International Agency for Research on Cancer (1993) WHO IARC monographs on the evaluation of carcinogenic risks to humans. Some naturally occurring substances: food items and constituents, heterocyclic aromatic amines and mycotoxins.aflatoxins. IARC, Lyon 6:245–395

Iyer RS, Coles BF, Raney KD, et al.(1994), DNA adduction by the potent carcinogen aflatoxin B1: mechanistic studies. J Am Chem Soc.1994, 116 (5) :1603–1609.

Jackson PE, Qian GS, Friesen MD, et al.(2001) Specifc p53 mutations detected in plasma and tumors of hepatocellular carcinoma patients by electrospray ionization mass spectrometry. Cancer Res. 2001 Jan 1; 61(1): 33-35

Jia L, Wang XW, Harris CC. (1999) Hepatitis B virus X protein inhibits nucleotide excision repair. Int J Cancer. 1999 Mar 15;80 (6) :875–879

Jiang W, Wang XW, Unger T, et al.(2010). Cooperation of tumor-derived HBx mutants and p53-249(ser) mutant in regulating cell proliferation, anchorage-independent growth

and aneuploidy in a telomerase-immortalized normal human hepatocyte-derived cell line. Int J Cancer. 2010 Sep1;127(5):1011-1020

Johnson WW, Ueng YF, Widersten M, et al.(1997) Conjugation of highly reactive aflatoxin B1 exo-8, 9-epoxide catalyzed by rat and human glutathione transferases: estimation of kinetic parameters. Biochemistry. 1997 Mar 18; 36(11): 3056-3060

Johnson WW, Guengerich FP. (1997) Reaction of aflatoxin B1 exo-8, 9-epoxide with DNA: kinetic analysis of covalent binding and DNA-induced hydrolysis. Proc Natl Acad Sci USA 1997 Jun 10;94 (12) :6121–6125.

Kaplanski C, Chisari FV, Wild CP.(1997) Minisatellite rearrangements are increased in liver tumours induced by transplacental afatoxin B1 treatment of hepatitis B virus transgenic mice, but not in spontaneously arising tumours. Carcinogenesis 1997 Apr; 18(4): 633-639

Kew MC. (2002) Epidemiology of hepatocellular carcinoma. Toxicology 2002 Dec 27;(181-182):35–38

Kew M.C. (2003) Synergistic interaction between aflatoxin B1 and hepatitis B virus in hepatocarcinogenesis. Liver Int. 2003 Dec;23 (6): 405–409.

Kirby GM, Chemin I, Montesano R, et al.(1994) Induction of specific cytochrome P450s involved in aflatoxin B1 metabolism in hepatitis B virus transgenic mice. Mol Carcinog.1994 Oct;11 (2) :74–80.

Kirk GD, Camus-Randon AM, Mendy M, et al. (2000) Ser-249 p53 mutations in plasma DNA of patients with hepatocellular carcinoma from The Gambia. J Natl Cancer Inst. 2000 Jan 19; 92(2): 148-153

Kirk GD, Lesi OA, Mendy M, et al.(2004) The Gambia liver cancer study: infection with hepatitis B and C and the risk of hepatocellular carcinoma in West Africa. Hepatology. 2004 Jan; 39 (1): 211–219.

Kirk GD, Lesi OA, Mendy M, et al.(2005) 249(ser)TP53 mutation in plasma DNA, hepatitis B viral infection, and risk of hepatocellular carcinoma. Oncogene. 2005 Sep 1; 24 (38): 5858–5867.

Kuang SY, Lekawanvijit S, Maneekarn N, et al.(2005) Hepatitis B 1762T/1764A mutations, hepatitis C infection, and codon 249 p53 mutations in hepatocellular carcinomas from Thailand. Cancer Epidemiol Biomarkers Prev. 2005 Feb; 14(2): 380-384

Kuniholm MH, Lesi OA, Mendy M, et al. (2008) Aflatoxin exposure and viral hepatitis in the etiology of liver cirrhosis in the Gambia, West Africa. Environ Health Perspect. 2008 Nov; 116(11): 1553-1557

Langouët S, Coles B, Morel F, et al. (1995) Inhibition of CYP1A2 and CYP3A4 by oltipraz results in reduction of aflatoxin B1 metabolism in human hepatocytes in primary culture. Cancer Res. 1995 Dec 1; 55(23): 5574-5579

Lin DY, Sheen IS, Chiu CT, et al.(1993) Ultrasonographic changes of early liver cirrhosis in chronic hepatitis B: a longitudinal study. J Clin Ultrasound. 1993 Jun; 21(5): 303-308

Lin JK, Miller JA, Miller EC. (1977) 2, 3-Dihydro-2-(guan-7-yl)-3-hydroxy-aflatoxin B1, a major acid hydrolysis product of aflatoxin B1-DNA or -ribosomal RNA adducts formed in hepatic microsome-mediated reactions and in rat liver in vivo. Cancer Res. 1977 Dec; 37(12): 4430-4438

Lin Y, Nomura T, Yamashita T, et al.(1997) The transactivation and p53-interacting functions of hepatitis B virus X protein are mutually interfering but distinct. Cancer Res. 1997 Nov 15;57 (22): 5137–5142.

Liu Y, Wu F.(2010) Global burden of aflatoxin-induced hepatocellular carcinoma: a risk assessment. Environ Health Perspect. 2010 Jun; 118(6):818–824.

Lodato F, Mazzella G, Festi D, et al.(2006).Hepatocellular carcinoma prevention: a worldwide emergence between the opulence of developed countries and the economic constraints of developing nations. World J. Gastroenterol. 2006 Dec 7; 12(45): 7239–7249.

Long XD, Ma Y, Zhou YF, et al. (2009) XPD codon 312 and 751 polymorphisms, and AFB1 exposure, and hepatocellular carcinoma risk. BMC Cancer.2009 Nov 17;9:400

López C, Ramos L, Bulacio L, et al. (2002) Aflatoxin B1 content in patients with hepatic diseases. Medicina.2002;62(4):313–316.

Macé K, Aguilar F, Wang JS, et al.(1997) Aflatoxin B1-induced DNA adduct formation and p53 mutations in CYP450-expressing human liver cell lines. Carcinogenesis. 1997 Jul; 18(7): 1291-1297

Madden CR, Finegold MJ, Slagle BL. (2002) Altered DNA mutation spectrum in aflatoxin b1-treated transgenic mice that express the hepatitis B virus x protein. J Virol. 2002 Nov;76 (22): 11770–11774.

Manuguerra M, Saletta F, Karagas MR, et al.(2006) XRCC3 and XPD/ERCC2 single nucleotide polymorphisms and the risk of cancer: a HuGE review.Am J Epidemio. 2006 Aug 15; 164(4):297-302

Marin MC, Jost CA, Brooks LA, et al. (2000) A common polymorphism acts as an intragenic modifier of mutant p53 behaviour. Nat Genet. 2000 May; 25(1): 47–54.

McGlynn KA, Rosvold EA, Lustbader ED, et al. (1995). Susceptibility to hepatocellular carcinoma is associated with gentic variation in the enzymatic detoxifcation of afatoxin B1. Proc Natl Acad Sci USA, 1995 Mar 14; 92(6): 2384-2387

McLean M, Dutton MF.(1995) Cellular interactions and metabolism of aflatoxin: an update. Pharmacol. Ther.1995 Feb; 65 (2): 163–192.

Meneghini R, Schumacher RI. (1977) Aflatoxin B1, a selective inhibitor of DNA synthesis in mammalian cells. Chem Biol Interact. 1977 Sep;18(3): 267-276

Ming L, Thorgeirsson SS, Gail MH, et al.(2002) Dominant role of hepatitis B virus and cofactor role aflatoxin in hepatocarcinogenesis in Qidong, China. Hepatology. 2002 Nov; 36(5): 1214–1220.

Oren M, Rotter V. (2010) Mutant p53 gain-of-function in Cancer. Cold Spring Harb Perspect Biol.2010 Feb; 2(2):a001107

Pemble S, Schiroeder KR, Spencer SR, et al.(1994) Human glutathione S-transferase theta (GSTT1): cDNA cloning and the characterization of a genetic polymorphism. Biochem J. 1994 May 15; 300(pt1): 271-276

Petitjean A, Mathe E, Kato S, et al.(2007) Impact of mutant p53 functional properties on TP53 mutation patterns and tumor phenotype: lessons from recent developments in the IARC TP53 database. Hum Mutat. 2007 Jun;28 (6):622–629.

Pfeifer AM, Cole KE, Smoot DT, et al.(1993) Simian virus 40 large tumor antigen-immortalized normal human liver epithelial cells express hepatocyte characteristics and metabolize chemical carcinogens. Proc Natl Acad Sci USA 1993 Jun 1; 90(11): 5123-5127

Ponchel F, Puisieux A, Tabone E, et al.(1994) Hepatocarcinoma-specific mutant p53-249ser induces mitotic activity but has no effect on transforming growth factor beta 1-mediated apoptosis. Cancer Res. 1994 Apr 15;54 (8):2064–2068.

Qian GS, Ross RK, Yu MC, et al.(1994). A follow-up study of urinary markers of aflatoxin exposure and liver cancer risk in Shanghai, People's Republic of China. Cancer Epidemiol Biomarkers Prev. 1994 Jan-Feb; 3(1): 3-10

Raney KD, Meyer DJ, Ketterer B, et al.(1992) Glutathione conjugation of aflatoxin B1 exo- and endo-epoxide by rat and human glutathione S-transferases. Chem Res Toxicol. 1992 Jul-Aug;5(4): 470-478

Rebbeck TR, Walker AH, Jaffe JM, et al.(1999) Glutathione S-transferase-mu (GSTM1) and -theta (GSTT1) genotypes in the etiology of prostate cancer. Cancer Epidemiol Biomarkers Prev. 1999 Apr; 8(4 Pt 1): 283-287

Ricordy R, Gensabella G, Cacci E, et al.(2002) Impairment of cell cycle progression by aflatoxin B1 in human cell lines. Mutagenesis 2002 May; 17(3): 241-249

Ross RK, Yuan JM, Yu MC, et al. (1992) Original article. Urinary aflatoxin biomarkers and risk of hepatocellular carcinoma. Lancet.1992 Apr 18; 339(8799):943–946

Salhab AS, Edwards GS.(1977) Production of aflatoxicol from aflatoxin B1 by postmitochondrial liver fractions.J Toxicol Environ Health.1977 Jan;2(3):583-587

Santella RM, Chen CJ, Zhang YJ, et al. (1998) Biological markers of aflatoxin B1 in hepatocellular cancer in Taiwan. In: Mendelsohn ML, Mohr LC and Peeters JP, editors. Biomarkers: medical and workplace applications. Washington, DC: Joseph Henry Press, 1998: 355-364

Schleger C, Becker R, Oesch F, et al.(1999) The human p53 gene mutated at position 249 per se is not sufficient to immortalize human liver cells. Hepatology. 1999 Mar;29 (3): 834–838

Sell S, Hunt JM., Dunsford HA, et al.(1991) Synergy between hepatitis B virus expression and chemical hepatocarcinogens in transgenic mice. Cancer Res. 1991 Feb 15;51 (4) :1278–1285.

Semela D, Heim M. (2011).Hepatocellular carcinoma. Ther Umsch. 2011 Apr;68(4):213-217.

Shephard GS.(2009) Aflatoxin analysis at the beginning of the twenty-first century.Anal Bioanal Chem.2009 Nov;395(5):1215-1224

Smela ME, Currier SS, Bailey EA, et al.(2001) The chemistry and biology of aflatoxin B(1): from mutational spectrometry to carcinogenesis. Carcinogenesis 2001 Apr;22 (4): 535–545

Sohn S, Jaitovitch-Groisman I, .Benlimame N, et al.(2000) Retroviral expression of the hepatitis B virus x gene promotes liver cell susceptibility to carcinogen-induced site specific mutagenesis. Mutat Res. 2000 Jun 30; 460 (1): 17–28.

Stettler PM, Sengstag C.(2001) Liver carcinogen aflatoxin B1 as an inducer of mitotic recombination in a human cell line. Mol Carcinog 2001 Jul; 31(3): 125-138

Strano S, Fontemaggi G, Costanzo A, et al. (2002) Physical interaction with human tumor-derived p53 mutants inhibits p63 activities. J Biol Chem.2002 May 24;277(21):18817–18826

Sugimura T.(2000) Nutrition and dietary carcinogens. Carcinogenesis.2000 Mar;21(3):387-395.

Sun CA, Wang LY, Chen CJ, et al.(2001) Genetic polymorphisms of glutathione S-transferases M1 and T1 associated with susceptibility to aflatoxin-related hepatocarcinogenesis among chronic hepatitis B carriers: a nested case-control study in Taiwan. Carcinogenesis. 2001 Aug; 22(8): 1289-1294

Sun Z, Lu P, Gail MH, et al.(1999) Increased risk of hepatocellular carcinoma in male hepatitis B surface antigen carriers with chronic hepatitis who have detectable urinary aflatoxin metabolite M1. Hepatology.1999 Aug;30(2):379-383

Tsuda H, Hirohashi S, Shimosato Y, et al.(1989) Low incidence of point mutation of c-Ki-ras and N-ras oncogenes in human hepatocellular carcinoma. Jpn J Cancer Res. 1989 Mar; 80(3): 196-199

Turner PC, Sylla A, Diallo MS, et al.(2002) The role of aflatoxins and hepatitis viruses in the etiopathogenesis of hepatocellular carcinoma: a basis for primary prevention in Guinea-Conakry, West Africa. J Gastroenterol Hepatol. 2002 Dec; 17 Suppl: S441–S448.

Ueda H, Ullrich SJ, Gangemi JD, et al.(1995) Functional inactivation but not structural mutation of p53 causes liver cancer. Nat. Genet. 1995 Jan;9 (1): 41–47.

Wang LY, Hatch M, Chen CJ, et al. (1996). Aflatoxin exposure and risk of hepatocellular carcinoma in Taiwan. Int J Cancer. 1996 Sep 4; 67(5):620–625

Wang XW, Forrester K, Yeh H, et al.(1994) Hepatitis B virus X protein inhibits p53 sequence-specific DNA binding, transcriptional activity, and association with transcription factor ERCC3. Proc Natl Acad Sci USA.1994 Mar 15; 91 (6): 2230–2234.

Wang XW, Hussain SP, Huo TI, et al.(2002) Molecular pathogenesis of human hepatocellular carcinoma. Toxicology. 2002 Dec 27;181-182:43–47

Wild CP, Yin F, Turner PC, et al.(2000) Environmental and genetic determinants of aflatoxin-albumin adducts in the Gambia. Int J Cancer. 2000 Apr 1; 86(1): 1-7

Wild CP, Turner PC.(2002) The toxicology of aflatoxins as a basis for public health decisions. Mutagenesis 2002 Nov; 17(6): 471-481.

Wild CP, Montesano R.(2009) A model of interaction: Aflatoxins and hepatitis viruses in liver cancer aetiology and prevention. Cancer Lett. 2009 Dec 1;286(1):22-28

Williams JH, Phillips TD, Jolly PE, et al. (2004) Human aflatoxicosis in developing countries: a review of toxicology, exposure, potential health consequences, and interventions. Am J Clin Nutr. 2004 Nov;80 (5): 1106–1122

Wong JJ, Hsieh DP.(1976) Mutagenicity of aflatoxins related to their metabolism and carcinogenic potential.Proc Natl Acad Sci USA.1976 Jul;73(7):2241-2244

Xu L, Qian G, Tang L, et al. (2010) Variations of hepatitis B virus and serum aflatoxin-lysine adduct on high risk of hepatocellular carcinoma in Southern Guangxi, China.J Hepatol.2010 Oct;53(4):671-676

Yang HI, Lu SN, Liaw YF, et al.(2002) Hepatitis B e antigen and the risk of hepatocellular carcinoma. N Engl J Med. 2002 Jul 18:347 (2002) 168–174.

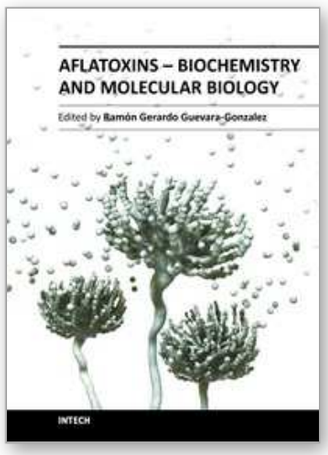
Yin L, Ghebranious N, Chakraborty S, et al.(1998) Control of mouse hepatocyte proliferation and ploidy by p53 and p53ser246 mutation in vivo. Hepatology.1998 Jan; 27 (1) :73–80.

Yu MC, Yuan JM. (2004) Environmental factors and risk for hepatocellular carcinoma. Gastroenterology 2004 Nov; 127(5 Suppl 1): S72–S78

Yuen MF, Hou JL, Chutaputti A, et al.(2009). Hepatocellular carcinoma in Asia Pacific region. J Gastroenterol Hepatol.2009 Mar; 24(3): 346–353.

Zhang YJ, Chen SY, Tsai WY, et al. (2000) Expression of cytochrome P450 1A1/2 and 3A4 in liver tissues of hepatocellular carcinoma cases and controls from Taiwan and their relationship to hepatitis B virus and aflatoxin B1- and 4-aminobiphenyl-DNA adducts. Biomarkers. 2000 July 1;5(4):295-306

Zhang YJ, Rossner P Jr, Chen Y, et al. (2006)Aflatoxin B1 and polycyclic aromatic hydrocarbon adducts, p53 mutations and p16 methylation in liver tissue and plasma of hepatocellular carcinoma patients. Int J Cancer 2006 Sep1; 119(5): 985-991



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Aflatoxins â€“ Biochemistry and Molecular Biology is a book that has been thought to present the most significant advances in these disciplines focused on the knowledge of such toxins. All authors, who supported the excellent work showed in every chapter of this book, are placed at the frontier of knowledge on this subject, thus, this book will be obligated reference to issue upon its publication. Finally, this book has been published in an attempt to present a written forum for researchers and teachers interested in the subject, having a current picture in this field of research about these interesting and intriguing toxins.

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