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**Short communication**

# Effect of aflatoxin B1 on DNA structural characteristic

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**The present investigation is an attempt to detection the effect of aflatoxin B1 on structural characteristic of DNA in different tissue, include liver, kidney, spleen and blood, *in vivo* by treated weight albino rate to AFB1 for 2 weeks as oral daily dose 1 ml/ kg. DNA fragmentation test was used to detect changes in DNA and micronuclei test in bone marrow cell. the results show that AFB1 causes lyses in DNA in all tissue and blood in variant level compare with negative control, liver show greater level than other organs 9660 pb, spleen was 9133 pb, kidney was 6166 pb and lower level in blood was 1366 pb. So it cause increased in micronuclei formation in bone marrow cell.**

**Key words**: *DNA fragmentation test, Aflatoxin B1, DNA lyses level.*

Aflatoxines are one of mycotoxin that product by fungi like *Asperagillus* spp. Aflatoxine B1 (AB1) is one of among potent genotoxic agent (8) it causes chromosomal aberration and DNA breakage in plant and animal cell and induced lethal mutation in mice and significant decrease in the fertility by genetic damage or mutation in germ cell (3,18) other researchers reported that the large dose of aflatoxin may be lethal but sub lethal cases chronic toxicity and low chronic dose causes neoplasma, liver cancer, inhibit DNA synthesis, RNA synthesis, and proteine synthesis (4,11). So AF causes hepatocarcinogeneses in human and mutation in p53 tumor suppression gene (1). Furthermore studies in rat, mice, and fish improved the aflatoxin activation protooncogenes and transversion mutation G→A and G→T in codon 12 in Ki-ras gene (10).

## Materials and methods

- ***Aflatoxin*:** aflatoxin extraction from *Asperagillus terreus* according to (7), detection AFB1 by thin layer chromatography (TLC) by use methanol: chloroform (97: 3) (v/v) (16).
- ***Experimental design***: using female rats (250±50 g body weight, age 14±2 weaks) animals divided in to 2 groups for DNA studying, first treatment by tap water as negative control, second treatment by aflatoxine B1 in daily dose 1ml/kg for 15 days. For micronuclei animal divided in to 3 grouped, first treatment by tap water as negative control , second treatment by aflatoxine B1 in daily dose 1ml/kg for 15 days, third treated by cyclophosphomide drug as positive control in 10 mg\kg for 15 day .
- ***DNA assay*** : using DNA fragmentation test , DNA extraction from liver , kidney, spleen and whole blood by Genomic DNA purification kit (Promega USA ) . The purity and concentration according to (13) then electrophoresis of DNA according to (12) with some modification.

- ***Micronuclei assay:*** Micronuclei assay was performed according to (9) with some modification , slide colored by Giemza stain then exam in microscope (Meiji, Japan) to detection micronuclei in 1000 cell.

**Results and Discussion**

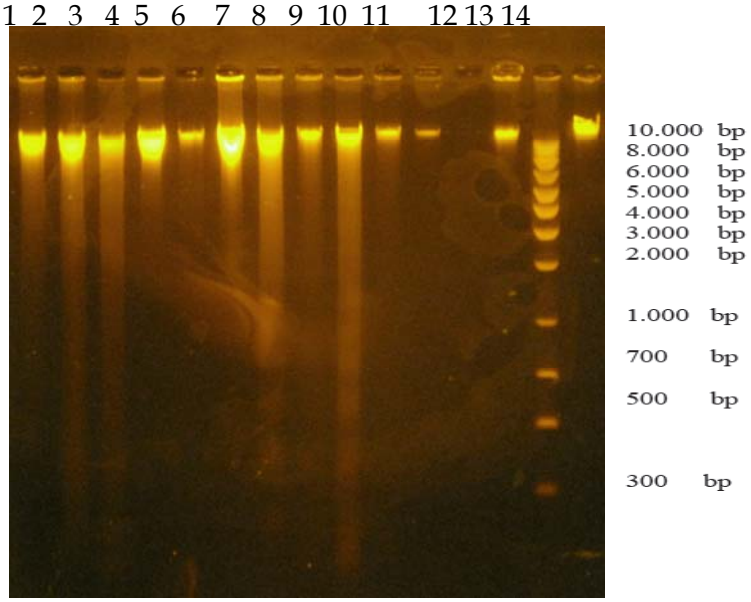


**Figure (1) Electrophoresis of DNA extraction from different tissues of animal treated by aflatoxine B1. Lane 1, 2, 3 DNA extract from liver. Lane 4, 5, 6 DNA extract from kidney. Lane 7, 8, 9 DNA extract from spleen. Lane 10, 11, 12 DNA extract from blood. Lane 13 DNA marker. Lane 14 DNA extract from negative control.**

**Table (1) DNA lyses level of different tissues explained in figure (1)**

| Lane number | Approximately Molecular size of DNA (bp) | Lyses level | Tissue type | Average of lyses level bp |
|---|---|---|---|---|
| 1 | 1000 | 9000 | liver | ≈ 9660 |
| 2 | < 300 | ≈ 9700 | liver | |
| 3 | < 300 | ≈ 9700 | liver | |
| 4 | 500 | 9500 | kidney | ≈ 6166 |
| 5 | < 5000 | ≈ 5000 | kidney | |
| 6 | < 6000 | ≈ 4000 | kidney | |
| 7 | 300 | 9700 | spleen | ≈ 9133 |
| 8 | 2000 | 8000 | spleen | |
| 9 | < 300 | ≈ 9700 | Spleen | |
| 10 | 6000 | 4000 | Blood | ≈ 1366 |
| 11 | 10000 | No lyses | Blood | |
| 12 | ≈ 10000 | < 100 | Blood | |
| 13 | DNA marker | No lyses | | _ |
| 14 | > 10000 | No lyses | Negative control | _ |

**Table 2. Macronuclei value in bone marrow cells.**

| Treatment | Macronuclei |
|---|---|
| Negative control | 10.00±7.50 |
| AFB1 | 64.66±16.02 |
| CP | 100.0±21.0* |

M±Se * significant ($p<0.05$)

The result show that aflatoxine B1 cusses DNA damage in liver , kidney , spleen and blood in different level show in figure (1) , the reason of this damage because AFB1- DNA adduct that form 8,9-epoxide and 8,9 – dihydro-8-( $N^7$-guanyl)-9-hydroxy- AFB1 by covalent bond between C8 of AFB1 – 8,9-epoxides and N7 of G base in DNA, or by pre covalent intercalation complex between double strand DNA and highly electrophelic unstable AFB1 – exo -8,9 peroxide isomers (15) . in the other hand oxidative stress caused by rats exposure to AFB1 can induced DNA lyses by forming (8-oxo dG) in liver DNA (14) . The level of DNA lyses have been showed in table (1). Liver was affected more than other tissue, the lyses level was 9660 pb, other researchers improved that rats exposed to single dose of AFB1 showed 10- folded greater level of DNA adduct in liver than in kidney , similar result have been reported in fish (6) , so the metabolic activation of AF by cytochrome CYP1A2 in liver it was more sensitive to toxic and mutagenic effect of AFB1 (5), kidney showed lower level of DNA lyses than liver and spleen, it had 6166 pb Bailey *et al*. (1988) reported that DNA adduct in kidney was low level than other organs in rainbow trout and salmon (2) blood DNA showed lower lyses level it was 1366 pb, the lower level of blood than other organs and tissue because blood cleaned by kidney and detoxificated by liver . micronuclei was increased in bone marrow cell of rats this may because of genotoxity of AFB1 it most potent genotoxic agent, is mutagenic in many model systems and produces chromosomal aberrations, micronuclei, sister-chromatid exchange, unscheduled DNA synthesis, and chromosomal strand breaks as well as forms adducts in rodent and human cells (17) .

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