Antimutagenic Profile of Antioxidant Vitamins in Drosophila Mulation Test

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Objective To assess the antimutagenicity of antioxidant vitamins (vitamins A, C, and E) as expressed by their efficacy to lower aflatoxin-induced mutations. **Methods** The Muller-5 method for mutation detection was used to assay the frequency of X-chromosome linked recessive lethal mutations (XRLMs) in Drosophila. Larvae were exposed to dietary concentration of aflatoxins and/or the human therapeutic doses of any of the three antioxidant vitamins. Absence of normal eyed males among M_2 progeny gave an indication of mutation induction. **Results** Aflatoxin supplimentation significantly increased the incidence of XRLMs in Drosophila. Mutation frequency was registered when aflatoxin-treated larvae were concomitantly fed with any of the three antioxidant vitamins. **Conclusion** Aflatoxin exposure can enhance the frequency of gene mutation in Drosophila which is significantly lowered by each of the three antioxidant vitamins. The degree of amelioration produced by them is almost identical. This mitigation is based on the scavenging/trapping by antioxidant vitamins of DNA-reactive products (metabolites and radicals) emanating from aflatoxin metabolism.

Key words: Antimutagenicity; Antioxidant vitamins

REFERENCES

- Berna Vetro I, Solti L, Szabo E, *et al.* (1997). Sensitive Enzyme linked immunosorbent assay for screening aflatoxin B₁ in food and feed stuff. *Cer Res Commun* 25, 291-292.
- Sinha S P, Prasad V (1990). Effect of dietary concentration of crude aflatoxin on meiotic chromosomes, sperm morphology and sperm count in mice, *Mus musculus*. *Proc Ind Natl Sci Acad* B 56, 269-279.
- Bose S, Sinha S P (1991). Aflatoxin-induced structural chromosomal changes and mitotic disruption in mouse bone marrow. *Mutat Res* 261, 15-19.
- 4. Pohland A E (1993). Mycotoxins in review. *Fd Contam* **10**, 17-28.
- Jelinek C E, Pohland A E, Wood G E (1989). Worldwide occurrence of mycotoxins in foods and feeds-an updated assessment. Assoc Off Anal Chem 72, 223-230.
- Kumari D, Sinha S P (1990). Combined effect of aflatoxin and vitamin A on clastogeny in mice chromosome. *Cytologia* 99, 387-390.
- Sinha S P, Dharmshila K (1994). Vitamin A ameliorates the genotoxicity in mice of aflatoxin B₁ containing *Aspergillus flavus* infested food. *Cytobios* **79**, 85-95.
- Sinha S P, Bose S (1992). Effect of dietary concentration of aflatoxin B₁ and vitamin C on meiotic chromosomes, sperm head morpholoty and sperm count in mice, *Mus musculus*. *Cytologia* 57, 75-80.
- Sinha S P, Prasad V (1989). Vitamin E minimizes aflatoxin-induced clastogenicity in mice. *The Nucleus* 32,

142-145.

- Nair A, Verma R J (2000). Effect of aflatoxins on testis of mouse and amelioration by vitamin E. Ind J Toxicol 7, 109-116.
- Elsas L J, Mc Cormick D B (1986). Genetic defects in vitamin utilization, Part-I: General aspects and fat-soluble vitamins. *Vit Horm* 43, 103-137.
- Bilgrami K S (1984). Mycotoxins in food. J Indian Bot Soc 63, 109-120.
- Strickberger M W (1985). *Genetics*, 3rd ed., pp. 457-459. Macmillan Publishing Company, New York.
- Herzbero P A (1983). Principles of Statistics. pp. 227-228. Wiley, New York.
- 15. Benasutti M, Ejadi S, Whitlow M D, *et al.* (1988). Mapping the binding site of aflatoxin B₁ in DNA : systematic analysis of the reactivity of aflatoxin B₁ with guanine in different DNA sequences. *Biochem* 27, 427-481.
- 16.Refolo L M, Benetta C B, Humayun M Z (1987). Mechanisms of frame shift mutagenesis by aflatoxin B₁, 2-3 dichloride. J *Mol Biol* **193**, 609-615.
- 17. Stark A A, Malca-Mor L, Hermann Y, et al. (1988). DNA strand scission and apurinic sites induced by photoactivated aflatoxins. *Cancer Res* 48, 3070-3076.
- 18. Yu F L, Bender W, Geronimo I H (1990). Base and sequence specificities of aflatoxin B₁ binding to single and double stranded DNA. *Carcinogen* **11**, 475-478.
- Shen H M, Shi C Y, Lee H P, et al. (1994). Aflatoxin B₁ induced lipid peroxidatin in rat liver. *Toxicol Appl Pharmacol* 127, 145-150.

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^{20.} Shen H M, Ong C H, Lee B L, et al. (1995). Aflatoxin B1

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induced 8-hydroxydeoxyguanosine formation in rat hepatic DNA. *Carcinogen* **16**, 419-422.

- 21.Hartman P E, Shankel D M (1990). Antimutagens and carcinogens: A survey of putative interceptor molecules. *Environ Mol Mutagen* 15, 145-182.
- 22. Salvadori D M F, Ribeiro L R, Natrajan A T (1993). The anticlastogenicity of β -carotene evaluated on human hepatoma cells. *Mutat Res* **303**, 151-156.
- 23.Sahu K, Das R K (1994). Reduction of clastogenic effect of clofazimine, an antileprosy drug by vitamin A and vitamin C in bone marrow cells of mice. *Fd Chem Toxicol* **32**, 911-915.
- 24. Sinha A K, Khan P K (1996). Anticlastogenicity of vitamin C in vivo against the cytogenetic toxicity of muriate of potash in murine bone marrow cells. In vivo 10, 111-112.
- Kalinina L M, Sardarly G M, Alekperov L (1979). Antimutagenic effect of alpha-tocopherol on the frequency of gene mutation in Salmonella. *Genetika* 15, 1880-1883.
- 26.Busk L, Ahlborg U G (1982). Retinoids as inhibitor of ortho-amino-azotoluene induced mutagenesis in Salmonella/ liver microsome test. *Mutat Res* 104, 225-231.
- 27.Norkus E P, Kuenzing W, Conney A H (1983). Studies on the mutagenic activity of ascorbic acid *in vitro* and *in vivo*. *Mutat Res* 117, 183-191.
- De Flora S, Ramel C (1988). Mechanism of inhibitors of mutagenesis and carcinogenesis: Classification and overview. *Mutat Res* 202, 285-306.
- 29. Radi R (1993). Biological antioxidant defences. *Toxicol Indus Hlth* **9**, 53-62.
- Sies H (1993). Strategies of antioxidant defence. *Eur J Biochem* 215, 213-219.
- 31.Mulholland C W, Strain J J (1983). Total radical trapping antioxidant potential (TRAP) of plasma-effect of supplementation of young healthy volunteers with large doses of α-tocopherol and ascorbic acid. Int J Vit Nutr Res 63, 27-30.
- 32.Sapper H, Kang S, Paul H, et al. (1982). The reversibility of vitamin C redox system: electrochemical reasons and biochemical aspects. Z Naturf 37, 942-946.
- 33.Rose R C (1990). Ascorbic acid metabolism in protection against free radicals: a radiation model. *Biochem Biophys Res*

Commun 169, 430-436.

- 34.Bhattacharya R K, Francis A R, Shetty T K (1987). Modifying role of dietary factors on the mutagenicity of aflatoxin B₁: In vitro effects of vitamin. *Mutat Res* 188, 121-128.
- 35.Surh Y J, Park K K, Millar J A (1994). Inhibitory effect of vitamin C on the mutagenicity and covalent DNA binding of the electrophilic and carcinogenic metabolite, 6-sulfoozymenthyl benzo (a) pyrene. *Carcinogen* 15, 917-920.
- 36.Rees S, Slater T F (1987). Ascorbic acid and lipid peroxidation: The cross-over effect. Acta Biochim Biophys Hung 22, 241-246.
- Krinsky N I (1989). Antioxidant functions of carotenoids. Free Radic Biol Med 6, 209-216.
- Busk L, Ahlborg U G (1980). Retinol (Vitamin A) as an inhibitor of mutagenicity of aflatoxin B₁. *Toxicol Lett* 6, 243-249.
- 39. Firozi P F, Aboobaker V S, Bhattacharya R K (1987). Action of vitamin A on DNA adduct formation by aflatoxin B₁ in a microsome catalysed reaction. *Cancer Lett* 34, 213-220.
- 40.Bhattacharya R K, Prabhu A L, Aboobaker V S (1989). *In vivo* effect of dietry factors on the molecular action of aflatoxin-B₁: Role of vitamins on the catalytic activity of liver fractions. *Cancer Lett* 44, 83-88.
- 41.Odin A P (1997). Vitamins as antimutagens: Advantages and some possible mechanism of antimutagenic action. *Mutat Res* 386, 39-67.
- 42. Van Acker S A B E, Koymans L M H, Best A (1993). Molecular pharmacology of Vitamin E: Structural aspects of antioxidant activity. *Free Radic Biol Med* 15, 311-328.
- 43.Sato K, Niki E, Shimasaki H (1990). Free radical mediated chain oxidation of low density lipoprotein and its synergistic inhibition by vitamin E and vitamin C. *Arch Biochem Biophys* 279, 402-405.
- 44.Chattopadhyay M K (2003). Antioxidants. Curr Sci 86, 366-367.
- 45.Kenyon A, Andress L (1980). Does Vitamin C induce X-linked recessive lethal mutation in *Drosophila melanogaster? Genet* 94, 552-553.

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