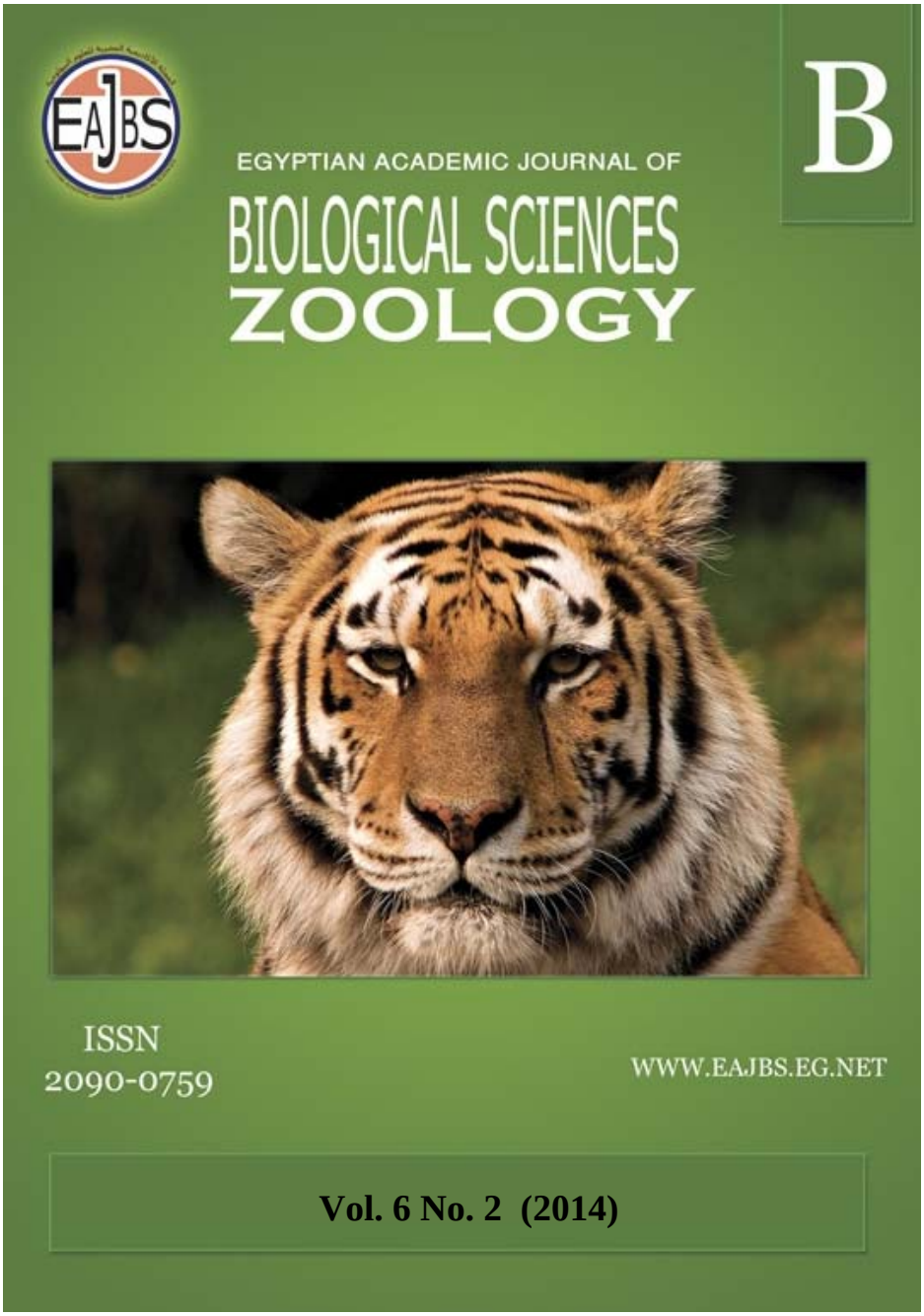


Egyptian Academic Journal of Biological Sciences is the official English language journal of the Egyptian Society of Biological Sciences ,Department of Entomology ,Faculty of Sciences Ain Shams University .

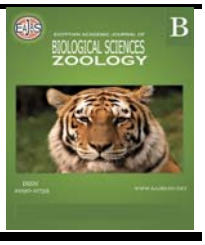
The Journal publishes original research papers and reviews from any zoological discipline or from directly allied fields in ecology, behavioral biology, physiology, biochemistry, development, genetics, systematics, morphology, evolution , and general zoolology

www.eajbs.eg.net

Received : 25/10/2014 Accepted :12/12/2014

Egypt. Acad. J. Biolog. Sci., 6(2): 17- 22 (2014)

Egyptian Academic Journal of Biological Sciences
B. Zoology
ISSN: 2090 – 0759
www.eajbs.eg.net



Effects of interaction between Aflatoxins (AFs) and functional materials FM in the hematological, biochemical parameters and enzyme activity in Rats

Anmar Sael Husain[1], Karkaz M.Thalij[2], Batol Imran Dheeb[3]

1- Biology Department/ College of Science/Tikreet University;
2- Food Science Department/ College of Agriculture/ Tikreet University;
3- Biology Department/ Iraqia University
Corresponding author: batoolomran@yahoo.com

ABSTRACT

The present study investigation the effect of aflatoxin induced the biochemical, hematological and enzyme activity in Rats Animals were fed aflatoxin-contaminated diet 1mg/kg diet for21 days were caused to reduced significantly ($p<0.05$) red blood cells(RBCs), hemoglobin (Hb) and increased significantly the white blood cells(WBCs) also decreased some the biochemical parameters so the liver function enzymes activity measuring were indicated as significantly decreased in alanine aminotransferase (ALT), aspartate aminotransferase (AST)and increased in alkaline phosphatase (ALP). The results of interactions between FMs as gensing GS, Whey protein WP and Butylated Hydroxyl Toluene BHT and AF byCo-treatment with FMs

Keywords: Aflatoxins, functional materials, RBC, Hb whey protein, hydroxyl butylatedtuluen

INTRODUCTION

Aflatoxins are naturally occurring produced by many species of Aspergillus fungus, the most notable ones being *A. flavus* and *A. parasiticus* plus related species, *A. nomius* (Luttfullah and Hussain, 2011). After entering the body, aflatoxinB1 is ingested, once inside the body, it is absorbed by the intestine and carried to the liver (Wild and Turner, 2002). There aflatoxin.B1 may be metabolized by the liver enzymes to changes for a reactive epoxide intermediate or hydroxylated derivatives compounds to become less harmful named aflatoxin M_1(Hudler,1998). Metabolized Aflatoxin, by enzymes in the liver, binds to proteins and causes acute toxicity (aflatoxicosis). Aflatoxin exposure causes acute liver damage and liver cirrhosis, as well as development of tumors or other genetic effects (Wu and Khlangwiset, 2010). On the another hand, the dietary sources, including plants, herbs, spices, vitamins and herbal extracts, play an important role to overcome the reactive metabolites production (Noori, 2012). So, an Antioxidant compound in food, which is main characteristic of an antioxidant, is its ability to catch free radicals and play an important role as a health protecting factor. So the antioxidants compounds were able to reduce the risk for chronic diseases (Percival, 1998).As, Ginseng is widely used medicinal plants which has a wide range of pharmacological and physiological actions (Ko, 1998; Slifman, 1998) Previous studies have shown that ginseng has

Received : 25/10/2014 Accepted :12/12/2014

antioxidant activity (Zhang, 1996; Keum, 2000), and it has protection from toxic substances (Lee, 1984; Mannaa, 2006). Also, whey proteins protect liver effect against oxidative stress-induced cell death in human prostate cells (Bounous, 2000 and Kent, 2003). Also Butylated Hydroxyl Toluene BHT inhibits AFB1 hepato-carcinogenesis

MATERIAL AND METHODS

Aflatoxins Production

Toxins were prepared by developing a strain of mold *Aspergillus parasiticus* NRRL2999 on rice according to shotwell; *et al.* (1966).then aflatoxins were extracted with chloroform according to Matny, (2012) and the AFs content was measured by the use of ELISA.

Experimental Animals

Forty tow Albino.-Sprague Dawley males Rats (110-115 g) at 45 day age have been obtained from the house livestock in the Faculty of Veterinary Medicine, University of Mosel were used to determine the effect of AF, and the prevention role of Gensing extract (GS), Whey protein (WP) and Butylated hydroxyl toluene (BHT) reared at an optimal room temperature ranged between 22–25°C. Animals were fed on locally prepared diet which formulated from natural ingredients suitable for growing maintenance according to (NAS. NRC, 2002). The experimental design consisted of fourteen dietary treatments. JSextract, WP and BHT were added from each ones at 100 and 150 mg/kg diet to the feed each of animal's groups containing AF at 1mg/kg diet.

Preparation of Blood Samples

At the end of the experiment, at 21 days we randomly selected from each treatment (4 rats of each iteration); 5ml of blood were collected with EDTA from Jugular Vein for hematological parameters determination. The measured of experiment as follows which working according to (Theml, *et al*., 2004).and other collected without them that have conducted the process of centrifugation at 3000rpm/15mint., to get the serum that was saved on $20C^0$ until a chemical analysis (Titez, 2005).

Blood Characteristics

The (10^3/ML), as well as, was measuring hemoglobin concentration HGB (g/dL) by used Heamocytometertotal number of red blood cells RBCs (10^6/ML) and the total number of white blood cells WBC was measured in collected blood. Also, all tests in haematology were taken by mathematically according to (Titez,2005).

Biochemical Blood Characteristics

Total protein, albumin, globulin, creatinine were estimated using biochemical kits (Bio labo, France, kits) in the blood serum Alkaline phosphatase (ALP), alanine aminotransferase (ALT), aspartate aminotransferase (AST) were determined by Reflotron using a clinical chemistry analyzer according to the manufacturer′s recommended procedure from Rondex Laboratories Ltd. (Tietz, 200)

RESULTS AND DISCUSSION

The Hematological Parameter

Hematological parameters in rat's animal fed with AF and supplemented with FMs as GS, WP and BHT for 21 day treatments are showed in Fig. (1.1). RBC count, hemoglobin concentration (Hb) and WBCs counts were significant difference

($p<0.05$) reductions in RBC $5.53\times10^3/mm^3$ Hb 9.1 dl/mm and WBC $10.6\times10^3/mm^3$, animals group fed with AF for 21 days compared with the control group which were RBC 7.7, Hb 13.5 and WBC 6.3 respectively, the addition of FMs were not causes significant difference on the levels of above parameters. Animal group fed with a diet contained FMs and AF (1 mg/kg diet) which caused to improve the rat's blood parameters were became close to the control group, which means that FMs effects on moderate the negative effects from AF and may offer the host protection against the negative effects of AF on intestinal absorption. The reason for this decline to the occurrence of lytic anemia because of the dissolution of red blood cells then decrease Hb concentration (Martins, 2004). And Hyperplasia in the bone marrow (Grior,1991 and Huff, *et al.*,1991).

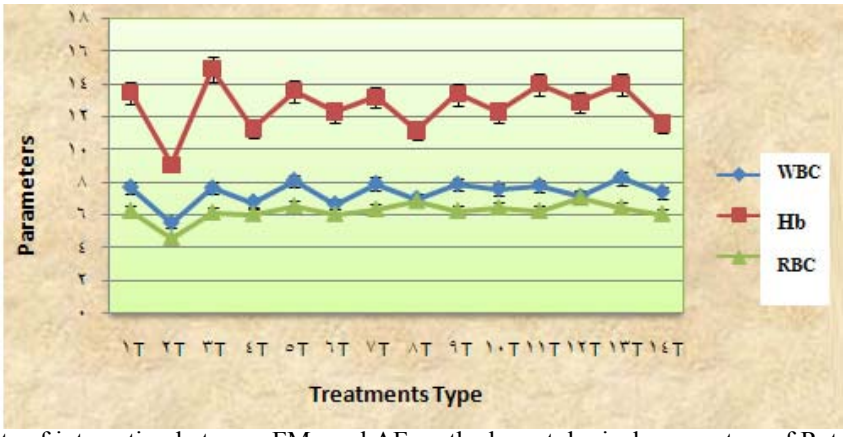


Fig. 1: Effects of interaction between FMs and AF on the hematological parameters of Rat after dietary for 21 days.

T1: control group; T2: 1mgAF/kg;T3:JN (100mg/kg); T4: JN 100mg+1mg AF/kg; T5: JN 150mg/kg; T6: JN 150mg+1mg AF/kg; T7: WP (100mg/kg); T8: WP 100 mg+1mgAF/kg; T9: WP150 mg/kg; T10: WP 150 mg+1mgAF/kg; T11: BHT100mg/kg; T12: BHT 100 mg+1mgAF/kg; T13: BHT 150mg/kg and T4:BHT 150 mg+1mgAF/kg.

Biochemical Parameters

The effect of interactions between AF and FMs in biochemical parameters including total proteins, albumin, globulin and creatinine in rats fed with a diet for 21 days were showed in Fig. 2. The results showed that AF added in diet alone reduced the concentration of total protein, albumin, globulin and the creatinine, significantly different. The addition of FMs alone to the diet of the rats, no significant relationship was found compared with the control group. The addition of FMs with AF to the diet were caused ameliorative the values of these parameters significantly compared with the T2 (AF alone group) and with the T1 (control group). The decrease in total protein concentration which lead to decrease the efficiency of the immune system since the key mechanisms of some immune responses were initial by production of inhibitory factors that effects on defense against the pathogens and other foreign materials, such as antimicrobial peptides and globulins (Buchau and Gallo, 2007). The results indicated that FMs were significantly improved a diet when incorporated with AF which was caused a negative effects, present results agreed with (Verma, *et al*., 2008**)**. Toulah, (2007) reported that the addition of turmeric as animals diet supplement (G4) were significantly decreased total protein and albumin levels, also, Hydrated sodium calcium alumino silicate (HSCAS) has demonstrated an ability to sorb aflatoxins (AFs) with a high affinity. Addition of this compound to feed stuffs contaminated with AFs has shown a protective effect against the development of aflatoxicosis in farm animals were significant improvement in the biochemical parameters. (Abdel-

Wahhab *et al.,* 2002). GS it serve as protective agents in cancer prevention and treatment while treatment rats of the intoxicated with ginseng resulted in significant enhancement in kidney function indicated by the marked decrease in creatinine levels and serum urea (Li, *et al*., 2008).

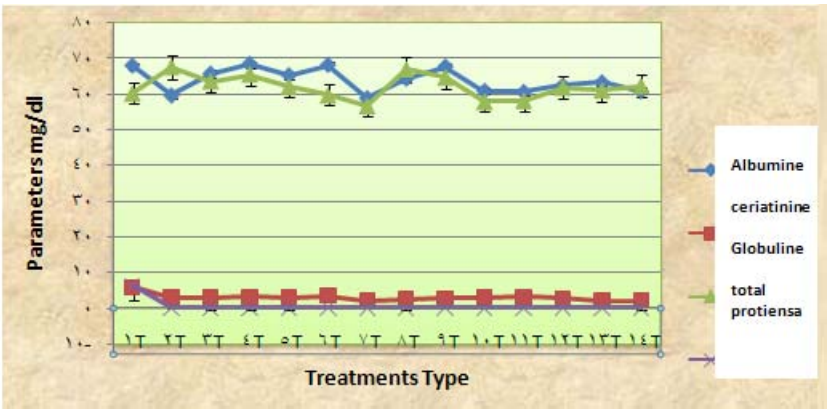


Fig. 2: Effects of interaction between FMs and AF on the biochemical parameters of Rat after dietary for 21 days.

Enzymes Concentrations

The blood enzymes concentrations of serum glutamic-oxaloacetic transaminase (GOT), serum glutamic-oxaloacetic transaminase (ALT) alkaline phosphatase (ALP) after adding FMs and AF for 21 days to the rat diet the results illustrated in Fig. 3. The results indicated that serum alkaline phosphatase was significantly increased ($P \leq 0.05$) in animals treated with AF 1mg/ kg diet and the enzyme concentration reached 857.5 IU/l, while the rats treated with FMs caused to ameliorative the enzymes concentration and became close to the control group of animal rats (286.8 IU/L), for the control group (286.8 IU/L). Serum ALT and AST were decreased significantly in animals when treated with 1mg AF/kg diet comparing with control animals group, GS extract stimulates the production of the reactive glutathione S transferees (one of the phase7II metabolizing enzymes) system found in the cytosol and microsomes catalyzes the conjugation of activated aflatoxins with reduced glutathione, leading to the excretion of aflatoxin (Williams, 2004).

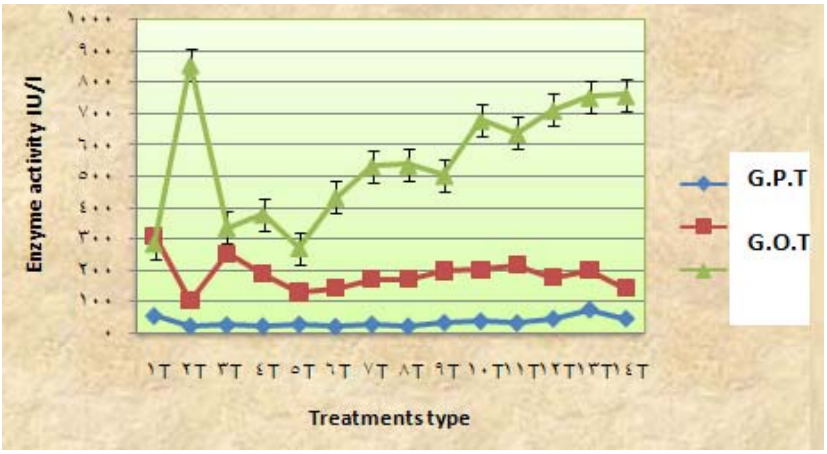


Fig. 3: Effects of interaction between FMs and AF in the enzyme activityof Rats after 21 days treatment.

Moreover, BHT with its potent free radicals cavenging ability was expected to inhibit Fe3+–ascorbate induced damage Since Fe3+ is a Fenton catalyst accelerating

ROS formation, BHT might also be expected to exclude e Fe3+ as a strategy to control ROS. (Khoobchandan, *et al*, 2010).

CONCLUSION

The consumed of food contaminated with aflatoxins from laboratory animals caused in changes the natural and biochemical parameters of the animals blood. Alsothe results were appear that the different concentrations of GS, WP and BHT as a prevention materials against the negative effects of aflatoxins on haematological, and biochemical parameters.

ACKNOWLEDGMENT

I am grateful for all that support me throughout our research.

REFERENCES

Abdel-Wahhab, M. A ., Nada, S. A., Khalil, F. A. (2002). Physiological and toxicological responses in rats fed aflatoxin-contaminated diet with or without sorbent materials. Animal Feed Science and Technology, 97 (3-4):209-219.

Bounous, G. (2000).Why protein conventate (WPC) and glutathione modulation in cancer treatment. Anticancer Res., 20:4785-4792.

Buchau, A. S. and Gallo, R. L. 2007. Innate immunity and antimicrobial defense systems in psoriasis. 295HClin. Dermatol. 25:616-624

Grioir, L. E., Huff, W. E., Kubena, L. F., Harvey, R. B.(1991).The individual and combined toxicity of kojic acid and aflatoxin in broilor chickens. Poult.sci., 70:1351-1356.

Hudler, G .W. (1998). Magical Mushrooms, Mischievous Molds: The Remarkable Story of the Fungus Kingdom and Its Impact on Human Affairs. Princeton University Press. USA.

Huff,W.E., Kubena,L.F.,Harvey,R.B. and Philips,T.D.(1991).Efficacy of hydrated sodium calcium aluminosilicate to reduce individual and combined toxicity of aflatoxin and ochratoxin A.poult,sci.,71:64-67.

Kent,K.D.,Harper,W.J.andBomserJ.A.(2003)Effect of why protein isolate on intracellular glutathione and oxidant-induced cell death in human prostate epithelial cells .Toxicol in vitro.17:27-33.

Keum, Y.S., Park, K.K., Lee, J.M., Chun, K.S., Park, J.H., Lee, S.K., Kwon, H., Surh, Y.J.(2000). Antioxidant and anti-tumor promoting activities of the methanol extracof heat-processed ginseng. Cancer Lett. 150:41–4.

Khoobchandani ,M., B.K. Ojeswi, N., Ganesh, M., Srivastava, S.Gabbanini, R., Matera, R., Iori, L.,Valgimigli(2010). Antimicrobial properties and analytical profile of traditionalEruca sativa seed oil: Comparison with various aerial and root plant extracts. Food Chemismtry 120 :217–224.

Ko, R.J. (1998). Adulterants in Asian patent medicines.N. Engl. J. Med. 39, 847.

Lee, F.C., Park, J.K., Kim, E.K., Ko, J.H., Lee, J.S., Kim, K.Y., (1984). The role of Panax ginseng in detoxification of xenobiotics. In: Proceedings of the Fourth International Ginseng Symposium, pp. 21–26.

Li, Q., Shi, S., Liu, Q., Tang, J., Song, J., Liang, Y. (2008). Anticancer effects of ginsenoside Rg1, cinnamic acid, and tanshinone in osteosarcoma MG-63 cells:

Nuclear matrix down regulation and cytoplasmic trafficking of nucleophosmin. Int. J. Biochem. Cell Biol., 40(9):1918-1929.

Luttfullah, G. and Hussain, A. (2011). Studies on contamination level of aflatoxins in some dried fruits and nutsof Pakistan. Food Cont. 22: 426-429.

Mannaa, F., Abdel-Wahhab, M.A., Ahmed, H.H., Park, M.H. (2006). Protective role of Panax ginseng extract standardized with ginsenoside Rg3 against acrylamideinduced neurotoxicity in rats. J. Appl. Toxicol. 26: 198–206.

Manty, O. N. (2012). Investigation about nut contamination by aflatoxin B1 in local market of Iraq. Education and science, 25(5):171-178. In Arabic.

Martins,M.L. and Martins,H.M.(2000).Aflatoxin M1in raw and temperature-treated milk commercialized in purtuga. Food additives, J. 10:871-879.

National Research Council Recommended (NAS-NRC). (2002). Dietary Allowance. 15th ed. Washington. D.C. National Academy. Press.

Noori, Sh. (2012). An Overview of Oxidative Stress and Antioxidant Defensive System. Open Access Scientific Reports. 1 (8): 1-9.

Percival,M. (1998)Antioxidants clinical nutrition insights Rev., 10/98.

Shotwell, O. L., Hessettin, C. W., Stubbiefield, R. D., Sorenson, W. G.(1966). Production of aflatoxin on rice. Appl Microbiol., 14(3):425-8.

Slifman, N. R., Obermeyer, W. R., Aloi, B. K., Musser, S. M., Correll Jr., W. A., Cichowicz, S. M., Betz, J. M., Love, L. A., 1998. Contamination of botanical dietarysupplements by Digitalis lanata. New Engl. J. Med. 17 339 (12): 806–811.

Tietz, N. ed., (2005). Fundamentals of clinical chemistry. W.B. Saunders, Philadelphia. pp. 723-750.

Theml, H., Diem, H. and Haferlach, T. (2004). Color Atlas of Hematology, Practical Microscopic and Clinical Diagnosis. 2nd revised ed., Stuttgart. New York., pp. 208.

Toulah, F. H. (2007). Effect of aflatoxin on thecoccidial infection in broilers. J. Egypt. Soc. Parasitol., 37(3):785-92.

Tung, H. T., Cook, F. W., Sylla, A., Wyatt, R. D. and Hamilton, P. B. (1975). The anemia causes by aflatoxin. poult. sci., 54:1962-1969.

Verma, R. J., Anita nair, and Neeta mathuria (2008). Vitamine ameliorates Aflatoxin-induced alteration. In the epididymis of Mice. Acta Poloniae Pharmaceutica ñ Drug Research, 65(3):331-337.

Wild, C. P and Turner, P. C. (2002) the toxicology of aflatoxins as a basis for public health decisions. Mutagenesis. 17(6):471-81.

Williams,J., Phillips, T., Jolly, P,. Stiles, J., Jolly, C. and. Aggarwal, D. (2004) Human aflatoxicosis in developing countries: a review of toxicology, exposure, potent ial health consequences, and interventions1,2,3 American Journal of Clinical Nutrition. 80: 11067112.

Wu, F and Khlangwiset, P. (2010). Health economic impacts and cost-effectiveness of aflatoxin reduction strategies 61 in Africa: Case studies in biocontrol and postharvest interventions. Food. Addit. Contam. Part A. Chem. Anal. Control. Expo. Risk Assess., 27(4): 496–509.

Zhang, X (2005). Expression of cytochrome 450 and other biotrnasormation genes in fetal and adult human nasal mucosa. *Drug* Metabolism and Disposition, 33:1423-1428.