Biochemical Pharmacology, Vol. 42, No. 7, pp. 1441–1445, 1991.
Printed in Great Britain.

0006-2952/91 $3.00 + 0.00

INFLUENCE OF ELLAGIC ACID ON ANTIOXIDANT DEFENSE SYSTEM AND LIPID PEROXIDATION IN MICE

SABHIYA MAJID, KRISHAN L. KHANDUJA,* RAJINDER K. GANDHI, SANJAY KAPUR and RATI R. SHARMA

Department of Biophysics, Postgraduate Institute of Medical Education and Research, Chandigarh, 160012, India

(*Received* 24 *January* 1991; *accepted* 31 *May* 1991)

Abstract—Addition of ellagic acid (EA) to liver microsomes of mice resulted in a steady increase in inhibition of NADPH-dependent lipid peroxidation up to 2 mM concentration. The maximum of 70% inhibition of ascorbate-dependent lipid peroxidation was achieved at 1 mM concentration of EA. Feeding of EA significantly increased the levels of reduced glutathione and glutathione reductase in liver and lungs of male and female mice. However, there were no changes in the activities of catalase and superoxide dismutase. On the other hand, microsomes from liver and lungs of EA fed animals showed significantly suppressed NADPH- and ascorbate-dependent lipid peroxidation.

Ellagic acid (EA†) is a naturally occurring plant phenol which is present in grapes, strawberries, raspberries and certain nuts [1, 2]. It has been reported to possess antimutagenic [3] and anti-carcinogenic [4, 5] activities. Involvement of active oxygen species in lipid peroxidation and carcinogenesis is firmly established [6, 7]. Lipid peroxidation has been shown to result in severe damage to cellular membranes, organelles and their associated enzymes in a number of systems [8]. Lipid peroxidation is known to be mediated enzymatically by the microsomal cytochrome P450 system which also activates the chemical carcinogen to bind with cellular critical molecules, like DNA. The chemical carcinogens are also oxidized by prostaglandin synthetase and lipid peroxidation [9, 10]. It is generally accepted that radical formation of the carcinogens, including polycyclic aromatic hydrocarbons, by enzymatic or non-enzymatic processes is of central importance in the carcinogenesis process. The organism can counteract the toxic activity of free radicals and peroxides to some extent owing to the cellular antioxidant defence system [11, 12]. The enzymatic antioxidant defence system influences lipid peroxidation via the activity of the enzymes SOD and catalase [4, 12]. GSH-dependent protection against lipid peroxidation is mediated through the activity of glutathione peroxidases which catalyse the reduction of lipid peroxides to stable lipid alcohols [13] or through other glutathione-dependent mechanisms [14, 15]. The diet may play a critical role in controlling this system and the process of free radical mediated lipid peroxidation. In the present study we report that dietary EA is capable of inhibiting the process of lipid peroxidation and modulating the antioxidant defence system in male and female mice.

MATERIALS AND METHODS

EA, GSH, GR, 5,5′-dithiobis (2-nitrobenzoic acid) were purchased from the Sigma Chemical Co. (St Louis, MO, U.S.A.). All other reagents of analytical grade were purchased locally.

Effect of ellagic acid on lipid peroxidation in vitro. Lipid peroxidation was stimulated non-enzymatically (ascorbate-dependent) and enzymatically (NADPH-dependent) according to the method of Kornburst and Mavis [16] in mouse liver microsomes. The study was carried out in the absence or presence of EA at 0.25, 0.5, 1 and 2 mM concentration.

Effect of feeding of ellagic acid on antioxidant defence system and lipid peroxidation. Male and female mice, 56 in each case, randomized at about 20 g by weight were taken and divided into four groups of 14 animals each. All animals were fed commercial pellet diet and drinking water was supplied *ad lib.* Group I animals served as control and were supplied with plain drinking water. Whereas drinking water of animals of groups II, III and IV was supplemented with EA at a concentration of 3, 6 and 12 μg/mL, respectively. Drinking water was freshly prepared every Monday, Wednesday and Friday. Daily intake of water was found to be about 8 mL/mouse. After 8 weeks of EA feeding the animals were killed after overnight fasting. Liver and lungs were excised. Tissue homogenates were prepared at a concentration of 1 g wet weight/4 mL of KCl/Tris–HCl buffer. GSH was estimated in part of the homogenate according to the method of Moron *et al.* [17]. The rest of the homogenate was centrifuged at 10,000 *g* in a cold centrifuge for 20 min. In 10,000 *g* supernatant the antioxidant enzymes comprising of SOD, catalase, GSH-Px and GR were assayed.

SOD was estimated by the method of Kono [18]. Catalase activity was measured by the method of

* To whom correspondence should be addressed.

† Abbreviations: EA, ellagic acid; GSH, reduced glutathione; GSSG, oxidized glutathione; GST, glutathione-*S*-transferase; SOD, superoxide dismutase; GHS-Px., glutathione peroxidase; GR, glutathione reductase; MDA, malondialdehyde; DTNB, 5,5′-dithiobis (2-nitrobenzoic acid).

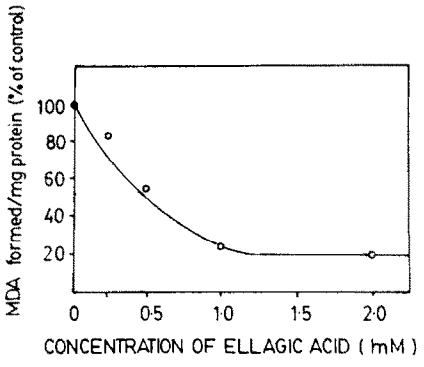

Fig. 1. Effect of EA on NADPH-dependent lipid peroxidation in mouse liver microsomes *in vitro*. EA was added at concentration of 0.25, 0.50, 1.00 and 2.00 mM. Data points represent mean of three replicate determinations.

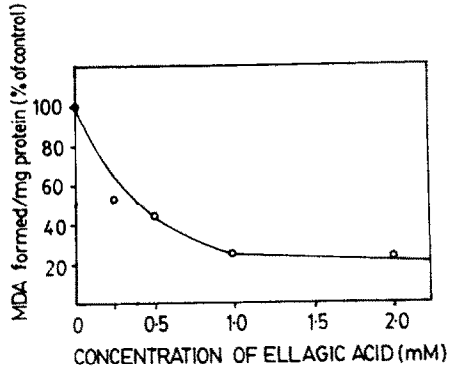

Fig. 2. Effect of EA on ascorbate-dependent lipid peroxidation in mouse liver microsomes *in vitro*. EA was added at concentration of 0.25, 0.50, 1.00 and 2.00 mM. Data points represent mean of three replicate determinations.

Luck [19]. GSH-Px. was measured by the method of Paglia and Valentine [20], and GR was assayed by the method of Dieter Horn [21]. Microsomes were isolated from the 10,000 *g* supernatant by the calcium aggregation method [22]. NADPH- and ascorbate-dependent lipid peroxidation was stimulated according to the method of Kornburst and Mavis [16].

Protein content was estimated by the method of Lowry *et al.* [23].

Determination of statistical significance of differences between groups was done by Student's *t*-test.

RESULTS

In both male and female mice oral feeding of EA at varying doses for 8 weeks had no effect on the body weight, organ weight and protein content per gram tissue.

Figures 1 and 2 show the effect of addition of EA, at 0, 0.25, 0.5, 1 and 2 mM concentration on the *in vitro* NADPH- and ascorbate-dependent lipid peroxidation in hepatic microsomes. EA addition resulted in a concentration dependent inhibition of malondialdehyde formation in both the systems.

In the enzymatically stimulated lipid peroxidation there was a steady increase in inhibition up to 2 mM concentration, and in non-enzymatically stimulated lipid peroxidation maximum inhibition (70%) was achieved at 1 mM concentration of EA. I_{50} values (concentration at which 50% inhibition of MDA formation was achieved) for enzymatically and non-enzymatically stimulated lipid peroxidation were 480 and 400 μM respectively.

Tables 1–4 depict the influence of oral feeding of EA at varying doses on the antioxidant defence system, and lipid peroxidation in liver and lungs of male and female mice. The changes in all the parameters were similar in the two sexes.

There was a significant increase in the GSH levels and GR activity in the liver and lungs of mice treated with EA (Groups II–IV). Though the activity of GSH-Px. in both the organs was increased by increasing the dietary EA level, the difference was not statistically significant. There seems to be no relationship between the feeding of EA and the activity of catalase and SOD in both the organs.

On the other hand, enzymatically and non-enzymatically stimulated lipid peroxidation in the liver and lung microsomes isolated from EA-fed mice were significantly inhibited. Malondialdehyde production was taken as an index of lipid peroxidation. In microsomes separated from the liver of Group II animals fed with EA at the lowest dose (3 μg/mL drinking water) for 8 weeks, malondialdehyde formation was about half of that produced by microsomes isolated from the liver of control animals in the case of male mice. In females too, there was a highly significant inhibition of MDA production in the liver microsomes isolated from EA-fed animals. MDA production in microsomes derived from lungs of both male and female EA-fed mice was also significantly inhibited.

DISCUSSION

Free radical mechanisms in initiation and promotion of carcinogenesis have been widely implicated [24–26]. Therefore, any antioxidant agent that scavenges free radicals directly or that interferes somehow with the generation of free radical mediated events is likely to inhibit the neoplastic process at its various stages of development. A short-term test for the antioxidative properties of a compound is to estimate its effect on NADPH- and ascorbate-dependent lipid peroxidation stimulated in liver microsomes *in vitro*, and we have observed that EA strongly inhibited both NADPH- and ascorbate-dependent lipid peroxidation *in vitro* also. It is known that certain compounds by virtue of their phenolic properties possess antioxidative properties [27–30]. Thus EA, being a plant polyphenol and a strong inhibitor of *in vitro* lipid peroxidation, probably inhibits the various free radical mediated processes. The I_{50} values for NADPH- and ascorbate-dependent lipid peroxidation were 480 and 400 μM, respectively, revealing that EA is a stronger inhibitor

Table 1. Effect of ellagic acid feeding at varying doses on antioxidant defence system and lipid peroxidation in liver of male mice

Parameter*	Ellagic acid (μg/mL drinking water)			
	0	3	6	12
Reduced glutathione	4.23 ± 0.19	6.01 ± 0.23†	7.59 ± 0.11†	8.51 ± 0.18†
Glutathione reductase	6.19 ± 0.89	10.9 ± 1.45†	11.9 ± 1.35†	10.8 ± 0.65†
Glutathione peroxidase	314 ± 46	351 ± 42	314 ± 56	379 ± 60
Catalase	489 ± 23	467 ± 56	489 ± 43	501 ± 29
Superoxide dismutase	0.89 ± 0.10	1.02 ± 0.15	1.0 ± 0.10	0.97 ± 0.20
Enzymatically stimulated lipid peroxidation	15.8 ± 1.77	11.5 ± 1.97†	8.2 ± 1.77†	7.31 ± 0.82†
Non-enzymatically stimulated lipid peroxidation	17.2 ± 2.2	9.16 ± 1.48†	7.52 ± 1.23†	8.02 ± 0.6†

* Units: Glutathione reductase and glutathione peroxidase: nmol NADPH consumed/min/mg protein; catalase and superoxide dismutase: I.U./mg protein; enzymatically and non-enzymatically stimulated lipid Px: nmol MDA formed/mg protein.

P: values in ellagic acid fed group are compared with control groups. † $P < 0.005$.

Table 2. Effect of ellagic acid feeding at varying doses on the antioxidant defence system and lipid peroxidation in liver of female mice

Parameter*	Ellagic acid (μg/mL drinking water)			
	0	3	6	12
Reduced glutathione	4.22 ± 0.40	6.08 ± 0.31‡	7.37 ± 0.53‡	7.87 ± 0.57‡
Glutathione reductase	6.90 ± 0.51	8.70 ± 1.25†	8.98 ± 0.88‡	9.14 ± 1.35‡
Glutathione peroxidase	250 ± 54	289 ± 46	302 ± 50	295 ± 59
Catalase	412 ± 49	415 ± 67	440 ± 62	416 ± 57
Superoxide dismutase	0.69 ± 0.08	0.69 ± 0.09	0.71 ± 0.10	0.70 ± 0.05
Enzymatically stimulated lipid peroxidation	15.3 ± 2.50	8.21 ± 0.83‡	8.69 ± 1.14‡	7.97 ± 1.31‡
Non-enymatically stimulated lipid peroxidation	16.6 ± 1.30	8.4 ± 1.4‡	8.8 ± 1.15‡	8.9 ± 1.38‡

* Units as in Table 1.

P: values in ellagic acid fed group compared with control groups. † $P < 0.01$; ‡ $P < 0.005$.

Table 3. Effect of ellagic acid feeding at varying doses on the antioxidant defence system and lipid peroxidation in lungs of male mice

Parameter*	Ellagic acid (μg/mL drinking water)			
	0	3	6	12
Reduced glutathione	0.27 ± 0.03	0.86 ± 0.11§	0.93 ± 0.08§	1.04 ± 0.04§
Glutathione reductase	4.18 ± 0.35	5.66 ± 0.31§	6.93 ± 0.74§	7.05 ± 0.65§
Glutathione peroxidase	55.7 ± 10.0	55.2 ± 4.20	59.0 ± 8.50	59.6 ± 10.9
Catalase	51.4 ± 4.9	50.0 ± 8.1	53.4 ± 2.9	57.1 ± 3.6
Superoxide dismutase	1.39 ± 0.12	1.41 ± 0.16	1.45 ± 0.18	1.53 ± 0.17
Enzymatically stimulated lipid peroxidation	5.05 ± 0.58	4.30 ± 0.35†	4.08 ± 0.13§	4.31 ± 0.42†
Non-enzymatically stimulated lipid peroxidation	5.28 ± 0.76	4.36 ± 0.51†	3.83 ± 0.74‡	3.04 ± 0.29§

* Units as in Table 1.

P: values in ellagic acid fed group compared with control groups. † $P < 0.05$; ‡ $P < 0.01$; § $P < 0.005$.

Table 4. Effect of feeding ellagic acid at varying doses upon the antioxidant defence system and lipid peroxidation in lungs of female mice

Parameter*	Ellagic acid (μg/mL drinking water)			
	0	3	6	12
Reduced glutathione	0.54 ± 0.05	0.78 ± 0.12§	0.94 ± 0.07§	1.14 ± 0.06§
Glutathione reductase	2.26 ± 0.52	5.74 ± 0.85§	6.82 ± 0.57§	6.78 ± 0.65§
Glutathione peroxidase	44.7 ± 8.5	48.6 ± 7.9	51.2 ± 7.4	49.1 ± 8.8
Catalase	38.6 ± 2.6	38.7 ± 6.1	38.1 ± 4.9	36.4 ± 2.0
Superoxide dismutase	1.03 ± 0.12	1.22 ± 0.23	1.20 ± 0.08‡	1.14 ± 0.16
Enzymatically stimulated lipid peroxidation	5.0 ± 0.49	4.4 ± 0.52†	4.35 ± 0.40+	4.29 ± 0.20‡
Non-enzymatically stimulated lipid peroxidation	5.09 ± 0.41	4.39 ± 0.32‡	4.16 ± 0.28§	3.05 ± 0.18§

* Units as in Table 1.

P: values in ellagic acid fed group are compared with control groups. † $P < 0.05$; ‡ $P < 0.01$; § $P < 0.005$.

of ascorbate-dependent lipid peroxidation than of NADPH-dependent lipid peroxidation. The initial hydrogen abstraction step in the NADPH-dependent lipid peroxidation is carried out by cytochrome P450 reductase and the chain propagatory reactions are free radical mediated. Therefore, the first step in the NADPH-dependent lipid peroxidation would not be effectively inhibited by EA though the peroxyl radical mediated propagation would be effectively inhibited. At the same time, ascorbate-dependent lipid peroxidation being wholly free radical mediated, both initiation and propagation would be strongly inhibited. This would result in a stronger inhibition of ascorbate-dependent lipid peroxidation compared to NADPH-dependent lipid peroxidation, as has been observed in the present study.

The active oxygen species O_2^-, $OH^\cdot$ and $^1O_2$ which are generated as harmful byproducts are known to be causative agents in lipid peroxidation, and are known to cause damage to nucleic acids [31, 32]. $OH^\cdot$ is capable of initiating lipid peroxidation by its ability to abstract hydrogen atoms from allylic positions of unsaturated lipids to yield hydroperoxides [33]. O_2^- which generates $^1O_2$ and $OH^\cdot$ via the Haber–Weiss reaction ($H_2O_2 + O_2^- \xrightarrow[\text{catalyst}]{\text{Fe salt}} O_2 + OH^\cdot + OH^-$) may also be a mediator of lipid peroxidation [31, 34]. The chain auto-oxidative process of lipid peroxidation of unsaturated fatty acids can be inhibited either by scavenging the peroxyl and or alkoxyl free radicals of the unsaturated fatty acids, or by removal of those active oxygen species and free radicals that initiate lipid peroxidation. This may lead to a protection against pathogenesis. EA being a polyphenol can act as a chain breaking antioxidant and may react directly with chain carrying peroxyl radicals ($ROO^\cdot$, where R=H, substituted alkyl etc.), thus terminating the propagation of these free radical mediated reactions. EA feeding did not result in any change in the activities of SOD, catalase and GSH-Px., the enzymes which are directly involved in the elimination of active oxygen species. Whereas, the levels of reduced glutathione were significantly increased by EA feeding in both liver and lungs of mice. Therefore, it is likely that GSH functions both directly and indirectly in protection against oxidative damage *in vivo*. GSH is the most important non-protein thiol present in the animal cells and is a powerful nucleophile. It can participate in a variety of detoxification reactions. An effective defence against oxidative damage (leading to lipid peroxidation) is the glutathione cycle. This includes oxidation of GSH to GSSG during detoxification of peroxides catalysed by GSH-Px. and GST and further reduction of GSSG to GSH by GR. The levels of GR too are significantly elevated on EA feeding. GSH might be involved in direct scavenging of free radicals by virtue of its thiol group. As GSH is present in the aqueous environment, it might not be readily accessible to the lipid environment of the membrane. Thus, it cannot be excluded completely that GSH has a major direct role in the free radical scavenging *in vivo*. GSH would rather function as a reductant for the chromoxyl radical formed during radical scavenging by α-tocopherol, which is the chief biological membrane antioxidant [35].

We also observed that both NADPH- and ascorbate-dependent lipid peroxidation stimulated in microsomes isolated from liver and lungs of EA-fed animals were significantly inhibited; thereby indicating that EA feeding has rendered the microsomes resistant to peroxidative damage. This may be a result of some biochemical changes brought about in the microsomes. It is also likely that EA binds to microsomal proteins, and thus scavenges free radicals. This reasoning is based on the observations that EA binds to calf thymus DNA as well as organ culture DNA *in vitro* [36–38] and thus would be prone to bind to other cellular macromolecules, like proteins, as well. Thus we have reached the conclusion that EA besides acting as an antoxidant, also enhances the GSH-dependent protective mechanisms. Therefore overall consequence of these antioxidant activities might be

beneficial and account for the anticarcinogenic activity of EA.

Acknowledgement—The financial assistance, IRISID 8804900, from the ICMR is duly acknowledged.

REFERENCES

1. Nemark HL, A hypothesis for dietary components as blocking agents of chemical carcinogenesis: plant phenolics and pyrrole pigments. *Can J Physiol Pharmacol* **65**: 461–466, 1987.
2. Doyle B and Griffiths LA, The metabolism of ellagic acid in the rat. *Xenobiotica* **10**: 247–256, 1980.
3. Wood AW, Huang MT, Chang RL, Newmark HL, Lehr RE, Yagi H, Sayer JM, Jerina DM and Conney AH, Inhibition of the mutagenicity of bay region diol epoxides of polycyclic aromatic hydrocarbons by naturally occurring plant phenols: exceptional activity of ellagic acid. *Proc Natl Acad Sci USA* **79**: 5513–5517, 1982.
4. Mukhtar H, Das M, Tito BJ Jr and Bickers DR, Protection against 3-methylcholanthrene induced skin tumorigenesis in BALB/C mice by ellagic acid. *Biochem Biophys Res Commun* **119**: 751–757, 1984.
5. Mukhtar H, Das M and Bickers DR, Inhibition of 3-methylcholanthrene induced skin tumorigenicity in BALB/c mice by chronic oral feeding of trace amounts of ellagic acid in drinking water. *Cancer Res* **46**: 2262–2265, 1986.
6. Cerutti PA, Prooxidant states and tumor promotion. *Science* **227**: 375–381, 1985.
7. Fridovich I, The biology of oxygen radicals. *Science* **201**: 875–880, 1978.
8. Tappel AL, Lipid peroxidation damage to cell components. *Fed Proc* **32**: 1870–1874, 1973.
9. Kadlubar FF, Frederick CB, Weis CC and Zenser TV, Prostaglandin endoperoxide synthetase-mediated metabolism of carcinogenic aromatic amines and their binding to DNA and protein. *Biochem Biophys Res Commun* **108**: 253–258, 1982.
10. Marnett LJ, Polycyclic aromatic hydrocarbon oxidation during prostaglandin biosynthesis. *Life Sci* **29**: 531–546, 1981.
11. Kellog EW and Fridovich I, Liposome oxidation and erythocyte lysis by enzymatically generated superoxide and hydrogen peroxide. *J Biol Chem* **252**: 6721–6728, 1977.
12. Fridovich I, Superoxide dismutase. *Annu Rev Biochem* **44**: 147–159, 1975.
13. Shan X, Aw TY and Jones DP, Glutathione dependent protection against oxidative injury. *Pharmac Ther* **47**: 61–71, 1990.
14. Cammeran R, Kellen J, Kolin A, Malkin A and Farber E, γ-Glutamyltransferase in putative premalignant liver cell populations during hepatocarcinogenesis. *Cancer Res* **38**: 823–829, 1978.
15. Novi AM, Regression of Aflatoxin B_1 induced hepatocellular carcinomas by reduced glutathione. *Science* **212**: 541–542, 1981.
16. Kornburst DJ and Mavis RD, Relative susceptibility of microsomes from lung, heart, liver, kidney, brain and testes to lipid peroxidation correlation with vitamin content. *Lipids* **15**: 315–322, 1980.
17. Moron MS, Depierre JW and Mannervick B, Levels of glutathione, glutathione reductase and glutathione-*S*-transferase activities in rat lung and liver. *Biochim Biophys Acta* **582**: 67–78, 1979.
18. Kono Y, Generation of superoxide radical during auto oxidation of hydroxylamine and an assay for superoxide dismutase. *Arch Biochem Biophys* **186**: 189–195, 1978.
19. Luck H, Catalase. In: *Methods of Enzymatic Analysis* (Ed. Bergmeyer HW), pp. 885–894. Academic Press, New York, 1963.
20. Paglia DE and Valentine WN, Studies on the quantitative and qualitative characterization of erythrocyte glutathione peroxidase. *J Lab Clin Med* **70**: 158–169, 1967.
21. Carlberg I and Mannervik B, Purification and characterization of the flavoenzyme glutathione reductase from rat liver. *J Biol Chem* **250**: 5475–5480, 1975.
22. Litterst CL, Mimnaugh E, Regan G, Reginald L and Gram TE, Drug metabolism by microsomes from extrahepatic organs of rats and rabbits prepared by calcium aggregation. *Life Sci* **17**: 813–818, 1975.
23. Lowry OH, Rosebrough NJ, Farr AL and Randall RJ, Protein measurement with Folin phenol reagent. *J Biol Chem* **193**: 265–275, 1951.
24. Ts'O Pop, Caspary WJ and Lorentzen RJ, In: *Free Radicals in Biology* (Ed. Pryor WA), Vol. 3, p. 251. Academic Press, New York, 1977.
25. Pryor WA, Free radicals in biology: the involvement of free radicals in ageing and carcinogenesis. In: *Medicinal Chemistry* (Ed. Mathieu J), Vol. V, p. 331. Elsevier, Amsterdam, 1977.
26. McBrein DCH and Slater TF (Eds.), *Free Radicals, Lipid Peroxidation and Cancer*. Academic Press, London, 1982.
27. Borek C, Selenium and vitamin E inhibited radiogenic and chemically induced transformation *in vitro* via different mechanisms of action. *Proc Natl Acad Sci USA* **83**: 1490–1494, 1986.
28. Gallagher CH, The effect of antioxidants on poisoning by carbon tetrachloride. *Aus J Exp Biol* **40**: 241–254, 1962.
29. Kim HL and Jones LP, Predictive effects on BHA, ethoxyquin and disulfuran on acute pyrrolizidine alkaloid poisoning in mice. *Res Commun Chem Pathol Pharmacol* **36**: 341–344, 1982.
30. Stohs SJ, Hassan MQ and Murray WJ, Effects of BHT, tocopherol and retinyl acetate on TCDD mediated changes in lipid peroxidation, glutathione peroxidase activity and survival. *Xenobiotica* **14**: 533–537, 1984.
31. Lynch RE and Fridovich I, Permeation of the erythrocyte stroma by superoxide radical. *J Biol Chem* **253**: 1838–1845, 1978.
32. Cadet J and Teoule R, Comparative study of oxidation of nucleic acid components by hydroxyl radicals, singlet oxygen and superoxide anion radicals. *Photochem Photobiol* **28**: 493–508, 1978.
33. Pryor WA and Tang RH, Ethylene formation from methanol. *Biochem Biophys Res Commun* **81**: 498–503, 1978.
34. Beaichamp C and Fridovich I, A mechanism for the production of ethylene from methional. The generation of the hydroxyl radical by xanthine oxidase. *J Biol Chem* **245**: 4641–4646, 1970.
35. Burk RF, Glutathione dependent protection by rat liver microsomal protection against lipid peroxidation. *Biochim Biophys Acta* **751**: 21–28, 1983.
36. Teel RW, Distribution and metabolism of EA in mouse following intraperitoneal administration. *Cancer Lett* **34**: 165–171, 1984.
37. Teel RW, Ellagic acid binding to DNA as a possible mechanism for its antimutagenic and anticarcinogenic action. *Cancer Lett* **30**: 329–336, 1986.
38. Teel RW, Martin RM and Allahyari R, Ellagic acid metabolism and binding to DNA in organ explant cultures in the rat. *Cancer Lett* **36**: 203–211, 1987.