Anti-oxidation action of Curcumin in two forms of bed rest: Oxidative stress Serum and salivary markers

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Abstract

Microgravity is associated with an increase of peroxidative damage. The effect is more pronounced after long-duration space flight and can even last for several weeks after landing. The extensive research is going on preventation of peroxidative damge due to microgravity. It have been evidence that curcumin (diferuloylmethane), a yellow pigment in curry powder, exhibits antioxidant, anti-inflammatory, and proapoptotic activities. To determine preventive effect of curcumin on peroxidative damage due to two bed rest conditions. 20 healthy male volunteers equally divided into two groups (10 with curcumin and 10 without curcumin) were studied in condition before , during, and just bed rest conditions -6° head-down-tilt (HDT) bed rest and bed rest position (BD) for 10 days. We measured the salivary and serum oxidative markers such as Malonaldehyde, 8-hydroxydeoxyguanosine, vitamin C and E just before HDT & BD, during HDT & BD experiment, and in recovery in with curcumin and without curcumin group. The value of serum and salivary Vitamin C & E showed statistically significant decrease in both bed rest conditions condition as compared to before and in recovery stage , however levels were decreased less in curcumin groups as compared to without curcumin groups (Table-1&2, $P<0.05$) .MDA and 8-OHdG levels showed significant increase in simulating microgravity and Zero gravity condition as compared to before and in recovery stage , however levels were low in curcumin grops as compared to without curcumin groups (Table-1& 2, $P<0.05$). Serum and salivary correlation analysis revealed strong and highly significant correlation for MDA, Vitamin C & E and 8 dihydro-2 deoxyguanosine (8-OHdG) in before , during and recovery in both bed rest conditions. Since, saliva can be easily collected, non-invasive and measurement of salivary markers levels may prove to useful in space research. Hence, curcumin prevent peroxidative damage in both bed rest condition. Further study is required on antioxidation action of curcumin in space microgravity condition.

Key Words- Curcumin, Serum, Saliva, oxidative stress, two bed rest position, space microgravity.

Introduction

Current projects of missions to Mars, resulting in 2 years of microgravity conditions, demand the critical needs for the development of the optimal nutritional programs and physical countermeasures to prevent body mass and functional alterations. On long duration space flights such as mars mission, astronauts undergo many physiological changes such as loss of bone mass, muscle strength, and cardiovascular fitness, as a result of reduced metabolic activities and lower cellular and tissue oxygen demand 1-12. There is a balance in the body between oxidant production and antioxidant defence, with the balance shifted slightly in favour of oxidants 1-3. Mainly products of this "leakage" are the two ROS: superoxide radical (O2_) and H2O2 2. Other ROS include free radicals such as nitric oxide and compounds such as ozone and HOCl. ROS can attack and damage cellular constituents such as DNA, proteins, and membrane lipids. Oxidative damage from free radicals to DNA and lipids has been implicated in the etiology of a wide variety of chronic diseases and acute pathologic states 2-8. The

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chronic diseases range from oral disease such as periodontitis and oral cancer to cardiovascular disease and neurodegenerative disease including Alzheimer and Parkinson diseases.9-13 It has been observed that there is increased lipid peroxidation in human erythrocyte membranes and reductions in some blood antioxidants after long-duration space flight 13-15 It has been observed that there was urinary excretion of 8-iso-prostaglandin F2- and 8-oxo-7,8 dihydro-2 deoxyguanosine (8-OHdG) in six subjects during and after long-duration space flight (90 to 180 d) 16-17. Isoprostane 8-isoprostaglandin F2- and 8-OHdG are markers for oxidative damage to lipids and DNA, respectively 16-17 .Most rodent studies showed increased production of lipid peroxidation products postflight and decreased antioxidant enzyme activity post-flight 18. It has been found space flight to simultaneously down regulate antioxidant defence capacity and elicit an oxidative stress in the liver. There was an approximately 50% increase in liver malondialdehyde concentration with space flight 19. Vitamin E is the primary chain-breaking antioxidant in cell membranes 9,20,21. The protective role of vitamin C seems to lie in its ability to reduce the oxidized form of vitamin E, thereby making it reusable by the cell 9,23.

Curcumin (diferuloylmethane), a dietary pigment responsible for the yellow color of turmeric, is used as a traditional medicine, well documented in Ayurveda for the treatment of numerous inflammatory conditions. Extensive research within the past half-decade has confirmed that curcumin mediates anti-inflammatory effects through the downregulation of transcription factor nuclear factor-?B (NF-?B) , tumor necrosis factor (TNF) interleukin-6 , interleukin-8 , adhesion molecules , inducible nitric oxide synthase (iNOS) , matrix metalloproteinase-9 (MMP-9) , cyclooxygenase-2(COX-2), 5-lipoxygenase (5-LOX), and glutathione reversed the inhibition 24-34 . It has been reported that Curcumin act as antioxidant agents 30-34. Curcumin have shown that agent can be administered safely at oral doses of up to 8 g/d .There was no dose-limiting toxicity; dosing was limited by the number of pills that patients could or would swallow daily 35-36. Hence, this study was planned the effect of curcumin on serum and salivary markers of oxidative stress due to two forms of bed rest.

Materials and methods

The subjects of this investigation were 20 male volunteers aged (18-22 years, mean weight of 72.5 +_ 3.2 kg and mean height of 174.9+_ 3.4 cm) participated in an 8-hour 6° HDT bed-rest exposure (18-21 years, mean weight of 71.8 +_ 2.3 kg and mean height of 174.8+_ 3.3 cm) and bed rest position (18-24 years, mean weight of 73.6 +_ 3.4 kg and mean height of 175.1+_ 4.1 cm), who had not participated in systemic endurance training for 10 days prior to study and each subject was given a detailed explanation of the experimental protocol and provided written and verbal consent. Each subject completed a medical and dental history questionnaire to determine the status of systemic diseases, smoking, alcoholic and drugs history as well as clinical examination for systemic diseases, chronic diseases and oral & dental diseases. Patients were excluded from study who had systemic diseases, chronic diseases, oral & dental disease, smoking, alcoholic and drugs history. Five volunteers of each HDT and BD were selected and gave a curcumin once a day and others five volunteers of each HDT and BD did not gave anything.

Curcumin- 1 g caplet form Curcumin (900 mg curcumin, 80 mg desmethoxycurcumin, and 20 mg bisdesmethoxycurcumin) from Sabinsa was obtained .

Blood and saliva samples were taken just before HDT, throughout the time course of the HDT &BD experiment, and during recovery. Subjects were asked to awake at 6 A.M. on the day of the study and to remain seated or in standing position until arrival at research centre .Baseline control measurements were obtained during the hour before HDT &BD. At -9 A.M. the subjects were transferred supine to a gurney and tilted to 6' HDT& BD, where they remained for the next 8 h. At -5 P.M. till 10 days, after 10 days the subjects returned to a chair and stayed in seated position for the 4-h recovery period. Blood and saliva samples were prepared at the same time

Whole unstimulated saliva was collected over a five-min period from subjects with directions to allow saliva to pool at the bottom of the mouth

and drain into a collection tube, when necessary. Unstimulated whole saliva produced in a 5-min period (about 3 mL) was collected, allowed to drain into a plastic container, and centrifuged at 3,000 ×g, in 4°C for 5 min to remove bacterial and cellular debris. Saliva samples were stored at -80°C until analysis. Blood samples were collected into Vacutainer tubes. The blood was centrifuged at 1,700 g for 10 min and the plasma was separated. Plasma was stored at -80°C until analysis. Serum and salivary levels were assessed for MDA using thiobarbituric acid (TBA) method of Buege and Aust 37 . Concentrations of both vitamins were measured using liquid chromatography 38 .Quantitative measurement of the oxidative DNA adduct 8-OHdG was performed according to the method described by Toyokuni et al.39 Briefly, the saliva samples were centrifuged at 10,000g for 10 minutes and the supernatant was used to determine 8- OHdG levels with a competitive ELISA kit (Japan Institute for the Control of Aging, Shizuoka, Japan). The determination range was 0.5-200 ng/mL. Serum 8-OHdG levels were measured in duplicate by a competitive ELISA kit (OXIS, Portland, OR, USA) according to the manufacturer's instructions. The sensitivity of the method was 1 ng/ml. All data were statistically analyzed using SPSS statistical package (SPSS, version13, Chicago,IL,USA). Data are expressed as mean ± standard deviation. Differences between pre, during and after microgravity simulation were analyzed for significant, using one-way ANOVA test. Correlation assessment was performed using the Spearman correlation analysis. Statistical significance was defined as $p< 0.05$.

Results

The value of serum and salivary Vitamin C &E showed statistically significant decrease in simulating microgravity & zero gravity condition as compared to before and in recovery stage in with and without Curcumin groups , also lower in recovery stage as compared to before two bed rest conditions (Table-1&2, $P<0.05$) , however decrease in curcumin group was low as compared to without curcumin group. MDA and 8-OH dG levels showed statistically significant increase in both condition as compared to before and in recovery stage , also relatively higher in without curcumin group as compared with curcumin group (Table-1&2, $P<0.05$). Serum and salivary correlation analysis revealed strong and highly significant correlation for MDA, Vitamin C& E and 8 dihydro-2 deoxyguanosine (8-OHdG) in both bed rest condition in both groups ($r=0.86, r=0.67, r=0.76$, $P<0.001$) & ($r=0.67, r=0.66, r=0.64$, $P<0.001$) respectively

Discussion

In the present study, serum and salivary Vitamin C & E were significantly lowered in both conditions in both groups (Table-1&2, $P<0.05$), which support the previous studies 40-42.Decreased antioxidant defence may be one of the reasons for increased levels of ROS and subsequent tissue damage in two bed rest conditions. MDA levels in both rest conditions environment were significantly elevated in both groups contrast to before and in recovery stage. This indicates that increased lipid peroxidation due to 'free radical'-mediated injury occurs in both rest conditions. Increased lipid peroxidation can occur if the rate of production of reactive oxygen species is higher or the antioxidant level is low which concur with previous studies 40-44 . The 8-OHdG levels were increased in both conditions as in previous studies 28, 30-33. Different aspects of oxidative stress are measured by 8-OHG namely DNA damage and cell membrane damage, respectively 44-48. The increased 8-OHG, MDA levels and decreased Vitamin C and E levels were low in curcumin groups as compared without curcumin group as in previous studies 34 . Several reports suggest that curcumin can induce ROS 47,48 . There are also reports which suggest that curcumin quenches ROS production and thus acts as an antioxidant 49. Other reports suggest that curcumin quenches ROS production at low concentrations and induces ROS production at high concentrations 50. It may due to curcumin, like vitamin C, acts as both a pro-oxidant and an antioxidant. Whereas the pro-oxidant mechanism mediates apoptotic effects, the antioxidant mechanism mediates NF-?B-suppressive effects.

Hence, in both rest conditions have not only systemic but also oral antioxidant levels were reduced. Antioxidant defence (vitamin E and C)

Table-1 Salivary and serum MDA, Vitamin C& E and 8 dihydro-2 deoxyguanosine (8-OH dG) concentration in the plasma and saliva of 20 Normal healthy subject in before HDT without Curcumin (A), throughout the time course of the HDT experiment (B), during recovery (C) and before HDT with Curcumin (AA), throughout the time course of the HDT experiment (BB), during recovery (CC).

Markers	Serum and saliva	A	AA €	B	BB £,€	C	CC £
MDA	Salivary (µmol/L)	0.24±0.06 a	0.22±0.13	0.34±0.12*,a	0.25±0.14	0.25±0.13*	0.24±0.23
	Serum (µmol/L)	1.14 ±0.37 a	1.06 ±0.89	1.36 ±0.36*,a	1.01 ±0.68	1.18 ±0.24*	1.01 ±0.75
Vitamin C	Salivary (µg /L)	1.01±0.32 a	1.56±0.66	0.82±0.21*,a	1.23±0.67	0.97±0.24*	1.29±0.68
	Serum (µg/L)	8.23±1.23 a	8.96±2.46	7.56±1.89*,a	8.82±2.33	8.05±1.95*	8.88±2.86
Vitamin E	Salivary (µg/L)	0.43±0.12 a	0.56±0.46	0.31±0.14*,a	0.48±0.45	0.41±0.16*	0.54±0.29
	Serum (µg/L)	8.01±1.12 a	8.46±2.32	7.32±1.21*,a	8.23±3.34	7.90±1.12*	8.94±3.32
8-OH dG	Salivary (ng/ml)	0.32±0.04 a	0.22±0.13	0.45±0.07*,a	0.24±0.11	0.38±0.08*	0.22±0.12
	Serum (ng/ml)	2.12 ± 1.24 a	1.45 ± 1.11	2.79 ± 1.23*,a	1.89 ± 1.36	2.32 ± 1.26*	1.77 ± 1.12

*p < 0.05, as compared to after condition (C)
ap < 0.05, as compared to Before condition (A).
£p < 0.05, as compared to after condition (CC)
•p < 0.05, as compared to Before condition (AA).

Table-2 Salivary and serum MDA, Vitamin C& E and 8 dihydro-2 deoxyguanosine (8-OHdG) concentration in the plasma and saliva of 20 Normal healthy subject in before BD without Curcumin (A), throughout the time course of the BD experiment (B), during recovery (C) and before HDT with Curcumin (AA), throughout the time course of the HDT experiment (BB), during recovery (CC).

Markers	Serum and saliva	A	AA €	B	BB £,€	C	CC £
MDA	Salivary (µmol/L)	0.25±0.08 a	0.11±0.03	0.37±0.14*,a	0.15±0.13	0.28±0.19*	0.12±0.16
	Serum (µmol/L)	1.25 ±0.45 a	0.78 ±0.65	1.35 ±0.41*,a	0.76±0.65	1.23 ±0.78*	0.78 ±0.67
Vitamin C	Salivary (µg /L)	1.02±0.45 a	1.23±0.67	0.85±0.47*,a	1.08±0.76	.94±0.38*	1.09±0.69
	Serum (µg/L)	7.48±1.54 a	8.32±2.01	7.06±1.02*,a	8.00±2.01	7.41±1.84*	8.03±2.09
Vitamin E	Salivary (µg/L)	0.48±0.14 a	0.59±0.23	0.35±0.15*,a	0.51±0.23	0.47±0.19*	0.53±0.22
	Serum (µg/L)	8.11±1.08 a	8.88±2.13	7.54±1.09*,a	8.23±3.19	8.09±1.01*	8.56±2.56
8-OH dG	Salivary (ng/ml)	0.35±0.06 a	0.23±0.08	0.41±0.05*,a	0.26±0.08	0.34±0.02*	0.22±0.05
	Serum (ng/ml)	2.15 ± 1.26 a	1.89 ± 1.43	2.89 ± 1.25*,a	1.98 ± 1.13	2.14 ± 1.26*	1.78 ± 1.23

is compromised and oxidative stress was increased in both rest conditions. Curcumin act as antioxidant in both rest conditions. Hence, better formulations of curcumin may provide more antioxidant effect. Further study is required on the effect of curcumin as antioxidant agent in space microgravity & zero gravity condition.

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References

1. Freed, L.E., Vunjak-Novakovic, G. Spaceflight bioreactor studies of cells and tissues. Adv. Space Biol. Med, 2002; 8: 177-195.
2. Halliwell B. Anti-oxidants and human disease: a general introduction. Nutr Rev 1997;55:S44-S52.
3. Ames BN. Endogenous oxidative DNA damage, aging, and cancer. Free Radic Res Commun, 1989; 7: 121-25.
4. Loft S, Poulsen HE. Estimation of oxidative DNA damage in man from urinary excretion of repair products. Acta Biochim Pol, 1998; 45: 133-44.
5. Morrow JD, Roberts LJ. The isoprostanes: unique bioactive products of lipid peroxidation. Prog Lipid Res, 1997; 36: 1-21.
6. Rokach J, Khanapure SP, Hwang SW, et al. The isoprostanes: a perspective. Prostaglandins, 1997; 54: 823-51
7. National Research Council, Food and Nutrition Board. Dietary reference intakes for vitamin C, vitamin E, selenium and carotenoids. Washing ton, DC: National Academy Press, 2000.
8. Maxwell SR. Prospects for the use of anti-oxi dant therapies. Drugs, 1995; 49: 345.
   Rai B, Kharb S, Jain R, Anand SC. Salivary vita mins E and C in oral cancer. Redox Rep, 2007; 12(3): 163-4.
9. Rai B, Jain R, Anand S C, Kharb S. 8-hydroxydeoxyguanosine levels: Periodontitis in smoker and non-smoker: a pilot study. J Pak Dent Assoc, 2006; 15(2): 89-90.
10. Rai B, Kharb S, Anand S C. Effect of scaling and root planning on salivary 8-hydroxydeoxyguanosine levels : periodontitis. J Pak Dent Assoc, 2007; 16(4): 189-91.
11. Rai B, Kharb S, Jain R, Anand S.C. Salivary Lipid Peroxidation Product Malonaldehyde in Various Dental Diseases. World J Med Sci, 2006; 1(2): 100-101.
12. Markin AA, Zhuravleva OA. Lipid peroxidation and anti-oxidant defense system in rats after a 14-day space flight in the "Space-2044" spacecraft. Aviakosm Ekolog Med, 1993; 27: 47-50.
13. Markin AA, Popova IA, Vetrova EG, Zhuravleva OA, Balashov OI. Lipid peroxidation and activ ity of diagnostically significant enzymes in cos monauts after flights of various durations. Aviakosm Ekolog Med, 1997; 31: 14-8.
14. Markin AA, Zhuravleva OA. Lipid peroxidation and indicators of anti-oxidant defence system in plasma and blood serum of rats during 14-day spaceflight on-board orbital laboratory "Spacelab-2." Aviakosm Ekolog Med, 1998; 32: 53-5.
15. Loft S, Poulsen HE. Markers of oxidative dam age to DNA: anti-oxidants and molecular dam age. Methods Enzymol, 1999; 300: 166-84.
16. Awad JA, Roberts LJ II, Burk RF, Morrow JD. Isoprostanes-prostaglandin-like compounds formed in vivo independently of cyclooxygenase: use as clinical indicators of oxidant damage. Gastroenterol Clin North Am, 1996; 25(2): 409-27.
17. Hollander J, Gore M, Fiebig R, et al. Spaceflight down regulates anti-oxidant defense systems in rat liver. Nutrition, 1998; 24(2): 385-90.
18. Lee MD, Tuttle R, Girten B. Effect of spaceflight on oxidative and anti-oxidant enzyme activity in rat diaphragm and intercostal muscles. J Gravit Physiol, 1995; 2: 68-72.
19. Sen CK. Oxidants and anti-oxidants in exercise. J Appl Physiol, 1995; 79(3): 675-86.
20. Burton GW, Joyce A, Ingold KU. First proof that vitamin E is major lipidsoluble,chain-breaking anti-oxidant in human blood plasma. Lancet, 7; 2(8293): 327.
21. Janero DR, Hreniuk D, Sharif HM. Hydrogen per oxide-induced oxidative stress to the mammalian heart-muscle cell (cardiomyocyte): lethal peroxidative membrane injury. J Cell Physiol, 1991; 149(3): 347-64.
22. Packer JE, Slater TF, Willson RL. Direct observa tion of a free radical interaction between vitamin E and vitamin C. Nature, 1979; 278(5706): 737-8.
23. Singh, S.; Aggarwal, B. B. Activation of transcrip tion factor NF-kappa B is suppressed by curcumin (diferuloylmethane) [corrected]. J. Biol. Chem, 1995; 270: 24995-25000.
24. Aggarwal, S.; Ichikawa, H.; Takada, Y.; Sandur, S. K.; Shishodia, S.; Aggarwal, B. B. Curcumin (diferuloylmethane) down-regulates expression of cell proliferation and antiapoptotic and meta static gene products throughsuppression of IkappaBalpha kinase and Akt activation. Mol. Pharmacol, 2006; 69: 195-206.
25. Jang, M. K.; Sohn, D. H.; Ryu, J. H. A curcuminoid and sesquiterpenes as inhibitors of macrophage

TNF-alpha release from Curcuma zedoaria. Planta Med, 2001; 67: 550-552.
26. Gaddipati, J. P.; Sundar, S. V.; Calemine, J.; Seth, P.; Sidhu, G. S.; Maheshwari, R. K. Differential regulation of cytokines and transcription factors in liver by curcumin following hemorrhage/re suscitation. Shock, 2003; 19: 150-156.
27. Biswas, S. K.; McClure, D.; Jimenez, L. A.; Megson, I. L.; Rahman, I. Curcumin induces glu tathione biosynthesis and inhibits NF-kappaB ac tivation and interleukin-8 release in alveolar epi thelial cells: mechanism of free radical scaveng ing activity. Antioxid. Redox Signaling, 2005; 7: 32-41.
28. Aggarwal, B. B.; Shishodia, S.; Takada, Y.; ` Banerjee, S.; Newman, R. A.; Bueso-Ramos, C. E.; Price, J. E. Curcumin suppresses the paclitaxelinduced nuclear factor-kappaB pathway in breast cancer cells and inhibits lung metastasis of human breast cancer in nude mice. Clin. Can cer Res, 2005; 11: 7490-7498.
29. Chan, M. M.; Huang, H. I.; Fenton, M. R.; Fong, D. In vivo inhibition of nitric oxide synthase gene expression by curcumin, a cancer preventive natu ral product with anti-inflammatory properties. Biochem. Pharmacol, 1998; 55: 1955-1962.
30. Liacini, A.; Sylvester, J.; Li, W. Q.; Zafarullah, M. Inhibition of interleukin-1-stimulated MAP ki nases, activating protein-1 (AP-1) and nuclear fac tor kappa B (NF-kappa B) transcription factors down-regulates matrix metalloproteinase gene expression in articular chondrocytes.Matrix Biol, 2002; 21: 251-262.
31. Zhang, F.; Altorki, N. K.; Mestre, J. R.; Subbaramaiah, K.; Dannenberg, A. J. Curcumin inhibits cyclooxygenase-2 transcription in bile acid- and phorbol ester-treated human gas trointestinal epithelial cells. Carcinogenesis, 1999; 20: 445-451.
32. Huang, M. T.; Lysz, T.; Ferraro, T.; Abidi, T. F.; Laskin, J. D.; Conney, A. H. Inhibitory effects of curcumin on in vitro lipoxygenase and cyclooxygenase activities in mouse epidermis. Cancer Res, 1991; 51: 813-819.
33. Sandur SK, Ichikawa H, Pandey MK, Kunnumakkara AB, Sung B, Sethi G, Aggarwal BB. Role of pro-oxidants and antioxidants in the anti-inflammatory and apoptotic effects of curcumin (diferuloylmethane). Free Radical Bi ology & Medicine, 2007; 43: 568-580.
34. Cheng AL, Hsu CH, LinJK, et al. Phase I clinical trial of curcumin, a chemopreventive agent, in pa tients with high-risk or pre-malignant lesions. An ticancer Res, 2001; 21: 2895-90.
35.. Sharma RA, McLelland HR, Hill KA, et al.Pharmacodynamic and pharmacokinetic study of oral Curcuma extract in patientswith colorectal cancer Clin Cancer Res, 2001; 7: 1894-900.
36. Buege JA.; Aust SD. Microsomal lipid peroxidation. Meth Enzymol, 1978;.51: 302-310.
37. Nierenberg DW, Lester DC. Determination of vi tamins A and E in serum and plasma using a sim plified plasma clarification method and high-per formance liquid chromatography. J Chromatogr, 1985; 345: 275-84.
38. Toyokuni S, Tanaka T, Hattori Y, et al. Quantita tive immunohistochemical determination of 8-hy droxy-20-deoxyguanosine by a monoclonal anti body N45.1: its application to ferric nitrilotriacetate-induced renal carcinogenesis model. Lab Invest, 1997; 76: 365-374.
39. Yang TB, Zhong P, Qu LN, Yuan YH. Space flight and peroxidative damage. Space Med Med Eng (Beijing), 2003; 16(6): 455-8.
40. Stein TP. Space flight and oxidative stress. Nutri tion, 2002; 18(10): 867-71.
41 .Maillet A, Beaufrere B, Di Nardo P, Elia M, Pichard C. Weightlessness as an accelerated model of nutritional disturbances. Curr Opin Clin Nutr Metab Care, 2001; 4(4): 301-6.
42. Chan AC. Partners in defense, vitamin E and C. Can J PhysioPharmacol, 1993; 71: 725-31.
43. Stein TP, Leskiw MJ, Schluter MD, Donaldson MR, Larina I. Protein kinetics during and after long term space flight on MIR. Am J Physiol Endocrinol Metab, 1999; 276: E1014.
44. Nomura J, Arase Y, Sugaya S, Moriya T, Chen Z, Takahashi S, Kita K, Kikuno K, Nomura F, Suzuki N. Modification of urinary secretion of 8-hydroxy-2'-deoxyguanosine and serum ACTH concentra tion following repetitive parabolic flights. J Gravit Physiol, 2001; 8(1): P125-6.
45. Nomura J, Arase Y, Chen Z, Sugita T, Sugaya S, Takahashi S, Kita K, Suzuki N. Search for mol ecules that are biological indicators of gravity stress in the human body. J Gravit Physiol, 2000; 7(2): 65-6.
46. Atsumi, T.; Fujisawa, S.; Tonosaki, K. Relationship between intracellular ROS production and mem brane mobility in curcumin- and tetrahydrocurcumin- treated human gingival fi broblasts and human submandibular gland car cinoma cells. Oral Dis, 2005; 11: 236-242.
47. Strasser, E.M.;Wessner, B.;Manhart, N.; Roth, E. The relationship between the anti-inflammatory effects of curcumin and cellular glutathione con tent in myelomonocytic cells. Biochem. Pharmacol, 2005; 70: 552-559.
48. Das, K. C.; Das, C. K. Curcumin (diferuloylmethane), a singlet oxygen ((1) O(2)) quencher. Biochem. Biophys. Res. Commun. 2002; 295: 62-66.
49. Mishra, S.; Kapoor, N.; Mubarak Ali, A.; Pardhasaradhi, B. V.; Kumari, A. L.; Khar, A.; Misra, K. Differential apoptotic and redox regu latory activities of curcumin and its derivatives. Free Radic. Biol. Med, 2005; 38: 1353-1360.