Research Article

Effect of Fish Oil on Antioxidant Defense in Rats Exposed to Cigarette Smoke

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ABSTRACT

We studied the effect of fish oil in 30 and 90 days exposure of cigarette smoke in rats. The activities of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and the levels of reduced glutathione (GSH) were significantly decreased in cigarette smoke exposed group when compared to control. These changes were prevented by fish oil co-treatment in group IV rats. No significant alterations were noticed in fish oil *Per se* group. These results suggest that the supplementation of fish oil has an antioxidative property in rats exposed to cigarette smoke. The protective effect of fish oil can be extrapolated to passive smokers.

Keywords: Passive smoking, Fish oil, Antioxidant

INTRODUCTION

Cigarette smoking is one of the major risk factors for both cardiovascular diseases and lung cancer[1]. Cigarette smokers are at increased risk of both myocardial infarction (MI) and sudden death[2]. The likely mechanism for the association observed between cigarette smoking and Coronary Heart Disease (CHD) is related to increased thrombogenesis and decreased oxygen carrying capacity in cigarette smokers[3].

The risk of developing lung cancer is quantitatively related to cigarette smoke exposure and it is casually associated with cancer of larynx, oral cavity, oesophagus, pancreas and stomach in both men and women[4]. The incidence of sudden death is higher in smokers than in nonsmokers. Individuals who stop smoking have a lower incidence of both MI and CHD than those who continue to smoke[5].

Several reactive oxygen intermediates are generated in biological processes involved in the cellular respiration and respiratory burst of phagocytic cells. Exogenous insult of free radicals to respiratory tract may derive from polluting environmental agents, cigarette smoke, drugs, toxic compounds and hyperoxia[6]. Oxidizing radicals cause damage to proteins, lipids, carbohydrates, enzymes, nucleic acids and other biological constituents.

They are counterbalanced by different defence mechanisms present in the body, whose action may be enhanced by exogenous antioxidant supply. Oxidants are involved in the pathogenesis and progression of atherosclerotic heart disease[7] and chronic obstructive pulmonary disease (COPD)[8].

Fish oil containing n-3 polyunsaturated fatty acids (PUFA) have anti-thrombotic activity attributable to antagonism of platelet aggregation and possibly to profibrinolytic changes in the coagulation system[9,10]. Dietary fish oil modifies several risk factors of atherosclerosis. It is hypolipidemic[11] and lowers blood pressure in hypertensive patients[12]. Fish oil is an antidote for the cardiovascular risks of smoking[13] and fish consumption limits damage to the lungs caused by cigarette smoking[14]. The objective of the present study is to prevent the deleterious effects of free radicals present in the cigarette smoke by fish oil treatment.

MATERIALS AND METHODS

Male albino rats of Wistar strain weighing about 120-150 g were obtained from King Institute of Preventive Medicine, Chennai, for the study. The rats were maintained on a commercial food (M/s. Hindustan Lever Foods, Bangalore) and water *ad libitum*.

The experimental rats were divided into eight groups with six animals in each. Group I - rats were not exposed to cigarette smoke served as control, Group II - rats were exposed to cigarette smoke (Charminar brand, nicotine content 2.5 mg/cigarette)[15].

Group II - rats were exposed to cigarette smoke from a lighted cigarette was drawn from the bottom by slow suction. The animals were exposed to cigarette smoke for 30 and 90 days twice a day and duration each time being increased by 25 minutes on first day, 30 minutes on second day, 1 hour on third day and 2 hours on fourth day. On fifth day onwards the duration remained 2 hours. Group III - control rats administered with fish oil (Menhaden oil, Sigma) orally for 30 and 90 days at the dosage of 0.5 ml/kg. b.wt. / day and group IV - fish oil co-treated rats. At the end of 30 and 90 days rats were sacrificed after overnight fasting, blood and tissues (heart and lungs) were collected.

The following parameters, SOD[16], CAT[17], GPx[18] and levels of reduced GSH[19] were carried out in hemolysate and tissues of heart and lungs by standard procedures. The data of 6 animals in each group were statistically compared by using students 't'-test.

Table 1: Activities of superoxide dismutase and catalase in hemolysate and tissues. (Values are expressed as mean ± S.D. for 6 animals in each group)

{PRIVATE}	Superoxide dismutase and catalase after							
	30 days in Group				90 days in Group			
	I	II	III	IV	I	II	III	IV
Superoxide dismutase								
Hemolysate	3.80 ± 0.52	3.14 ± 0.24**	3.58 ± 0.46NS	3.72 ± 0.26***	3.78 ± 0.40	1.93 ± 0.09***	3.54 ± 0.36NS	3.49 ± 0.59***
Heart	6.40 ± 0.55	5.3 ± 0.45**	5.94 ± 0.58NS	6.37 ± 0.49***	6.47 ± 0.58	3.60 ± 0.37***	6.31 ± 0.58NS	6.24 ± 0.54***
Lungs	3.73 ± 0.21	3.32 ± 0.31**	3.69 ± 0.27NS	3.27 ± 0.24**	3.78 ± 0.34	2.54 ± 0.29***	3.59 ± 0.41NS	3.50 ± 0.32***
Catalase[b]								
Hemolysate	1.42 ± 0.15	1.26 ± 0.08**	1.54 ± 0.21NS	1.39 ± 0.09**	1.34 ± 0.16	0.60 ± 0.08***	1.40 ± 0.15NS	1.32 ± 0.13***
Heart	7.62 ± 0.43	6.84 ± 0.59**	7.46 ± 0.60NS	7.58 ± 0.50**	7.52 ± 0.61	5.24 ± 0.30***	7.41 ± 0.54NS	7.50 ± 0.62***
Lungs	6.89 ± 0.43	5.78 ± 0.62**	6.75 ± 0.54NS	6.72 ± 0.60**	7.05 ± 0.65	5.02 ± 0.49***	0.679 ± 0.51NS	6.68 ± 0.50***

a - 50% inhibition of epinephrine autooxidation; b - µmoles of H_2O_2 decomposed/min/mg Hb or gm tissue

Statistical comparison of group II and group III vs group I and group IV vs group II. Group I - Control, Group II - Cigarette smoke exposed, Group III - Fish oil *Per se*, Group IV - Smoke - Fish oil co-treated, NS - Non significant, ** p<0.01, *** - p<0.001

Table 2: Activities of glutathione peroxidase and levels of reduced glutathione in hemolysate and tissues. (Values are expressed as mean ± S.D for 6 animals in each group)

{PRIVATE}	Glutathione peroxidase and reduced glutathione after							
	30 days in Group				90 days in Group			
	I	II	III	IV	I	II	III	IV
Glutathione peroxidase[a]								
Hemolysate	7.21 ± 0.78	5.81 ± 0.66**	7.63 ± 0.68NS	7.11 ± 0.56***	7.40 ± 0.75	3.91 ± 0.45***	7.03 ± 0.69NS	6.89 ± 0.57***
Heart	15.32 ± 1.56	12.78 ± 1.08**	15.04 ± 1.59NS	14.86 ± 1.38**	15.24 ± 1.51	9.05 ± 1.05***	15.29 ± 1.59NS	15.01 ± 1.50***
Lungs	2.64 ± 0.18	2.35 ± 0.17**	2.57 ± 0.28NS	2.62 ± 0.19**	2.60 ± 0.28	1.33 ± 0.19***	2.59 ± 0.29NS	2.54 ± 0.30***
Reduced glutathione[b]								
Hemolysate	54.27 ± 2.62	50.57 ± 2.1**	53.9 ± 2.4NS	54.01 ± 2.2**	51.26 ± 1.19	42.14 ± 2.64***	52.86 ± 2.89NS	51.08 ± 2.06***
Heart	33.40 ± 3.01	28.40 ± 2.64**	30.42 ± 2.96NS	32.90 ± 3.14**	35.50 ± 3.05	16.19 ± 1.69***	32.26 ± 2.94NS	28.29 ± 2.89***
Lungs	36.47 ± 3.12	30.72 ± 2.96**	31.96 ± 3.18NS	35.49 ± 2.98**	36.05 ± 3.17	20.19 ± 2.54***	33.18 ± 3.04NS	34.35 ± 3.09***

a - µg of GSH Utilised/min/mg Hb or gm tissue; b - µ moles of GSH/mg protein or gm tissue

Statistical comparison of group II and group III vs group I and group IV vs group II. Group I - Control, Group II - Cigarette smoke exposed, Group III - Fish oil *Per se*, Group IV - Smoke - Fish oil co-treated, NS - Non significant, ** p<0.01, *** - p<0.001

RESULTS

The activities of SOD and CAT were significantly decreased in rats exposed to cigarette smoke for 30 (p<0.01) and 90 (p<0.001) days. Co-treatment with fish oil prevented these changes. Fish oil per se group had no significant effect (Table 1).

GPx activity and levels of reduced GSH were significantly decreased in rats exposed to cigarette smoke for 30 (p<0.01) and 90 (p<0.001) days. Co-treatment with fish oil prevented these changes. Fish oil per se group had no significant effect (Table 2).

DISCUSSION

The heart and lungs are particularly subjected to oxidant damage which has been implicated in the pathogenesis of numerous heart and lung diseases[20]. Cigarette smoke is potentially capable of generating high free radical load in the body[21]. Free radicals are highly toxic to the cells which should be detoxified. This detoxification is done by antioxidants and antioxidant enzymes present in the cells which in turn causes depletion of these enzymes. Reduced levels of antioxidants and antioxidant enzymes were found in cigarette smokers[22] and experimental animals[23,24]. Our present study also coincide well with above findings. Near normal activities of SOD, CAT, GPx and the levels of antioxidant GSH were found in fish oil co-treated group and this might be due to the components present in the fish oil. Fish oil modifies the composition of membrane phospholipids[25-27] and increases both n-3/n-6 ratio and the double bond index[28]. COPD mainly caused by mainstream and side stream cigarette smoke exposure[29, 30] and this may be less likely to develop in those with a greater intake of omega-3 fatty acids[31]. Demoz reported that hypolipidemic doses of purified eicosapentaenoic acid enhances the hepatic antioxidant defenses and reduced the lipid peroxide levels in mice[32]. Our present results were good agreement with above datas. From our observation, it is evident that the administration of fish oil prevents oxidant damage by cigarette smoke in experimental rats; the protective effect of fish oil can be extrapolated to passive smokers to counteract free radical damage by tobacco smoke.

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