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CHEMICAL ANALYSIS OF SHALLOT SUPPLEMENTATION ON ANTIOXIDANT STATUS

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Abstract

The major cause of chronic oxidative stress in humans is exposure to free radicals in cigarette smoke. Cigarette smoke free radicals are considered an important cause of atherosclerosis and cancer (Pryor, 1997; Pittilo, 2000). Thiols are powerful reducing agents that are capable of acting as antioxidants in vivo. Thiols exist in three forms: freethiol and two types of disulfides, namely, homodisulfides and heterodisulfides. Several aminothiols, e.g., cysteine, homocysteine (Hcy), and GSH, and disulfides (e.g., cystine, homocystine, and oxidized glutathione), interact by means of redox and disulfideexchange (Iciek et al., 2004). This dynamic system (with respect to thiol status) is important for normal physiologic function (Morris, 2002). Changes in the redox thiol status lead to the induction of oxidative stress and apoptosis. As both an intracellular and extracellular redox buffer, t-SH plays important roles in the in vivo prevention of atherosclerosis (Ueland et al., 1996). t-SH plays a prominent role in antioxidant reactions, and in catalysis, regulation, and electron-transport reactions, and in reactions that preserve the correct structure of proteins. Mixed disulfides with proteins are formed by reaction of S-thiolation, in which protein thiols conjugate with non-protein thiols (Klatt and Lamas, 2000). This process plays a regulatory and an antioxidant role, since it protects protein −SH groups against irreversible oxidation from −SO2H and −SO3H; moreover, it participates in signal transduction (Padgett and Whorton, 1998). GSH is the most important endogenous antioxidant in humans. It is often accompanying by other endogenous thiols, such as cysteine, cysteinylglycine and even Hcy (in low concentration). These thiols scavenge ROS and are involved in preserving the pro oxidant – antioxidant balance in human tissues. Thai shallot protects human erythrocytes from possible damage from external or internal radicals such as H2O2 or peroxyl radical such as 2,2'-Azobis (2- amidino-propane) dihydrochloride. They reported that thai shallot is able to inhibit lipid peroxidation and glutathione depletion in erythrocytes and suggested that the Thai shallotextracts have protective effect on the GSH deterioration in vitro from protein hydroperoxide (PrOOH) or hydroxyl radical from gamma irradiation. Thai shallot extracts can also protect and scavenge the protein and lipid hydroperoxide (LOOH) formation in vitro study.

Key Words: *Antioxidant, Oxidative stress, Free radicals, In vitro.*

Discussion and Results:

Cigarette smoke, as a pollutant, has been established to include a variety of xenobiotics, some of which are known to be oxidant or free radicals that can directly and indirectly initiate and propagate the process of lipid peroxidation The enhanced susceptibility of erythrocytes of smokers to peroxidation may reflect the lower activities of glucose-6-phosphate

dehydrogenase and glutathione peroxidase. Decreased activity of glucose-6-phosphate dehydrogenase can be caused by extracellular or intracellular lipid hydroperoxides. Differences in glutathione peroxidase activity between smokers and nonsmokers have been reported previously and may be associated with decreased selenium status. Thus, it is believed that smokers encounter a sustained free radical load. It has been shown that cigarette smoking caused an increase in blood and serum malonaldehyde content. Many studies have shown an increased lipid peroxidation due to cigarette smoking. Allium families are reported with high antioxidant potential. In this study to evaluate antioxidant potential of shallot in cigarette smokers, 40 healthy subjects were selected for study. Subjects, who smoke minimum 5 cigarettes per day since last 3 years in the age group between 25-40 years, were included for the study. Subjects were assigned to 2 groups of 20 each (study group and control group). Control group received placebo (rice flour) whereas study group received 500 mg shallot capsule twice a day. Antioxidant activity, lipid peroxidation and total thiol was measured in serum of subjects before and after supplementation.

Total Thiol

As illustrated in Figure 56, total thiol protein of smokers increased significantly after 30 days of shallot supplementation. Subjects were examined for serum total thiol before supplementation and it was found to be 0.146 μmol/ml in study group and 0.157 μmol/ml in placebo group. When shallot capsules were given to study group and placebo to control group, it was observed that total thiol was 0.243 and 0.158 μmol/ml in study and control group respectively. When data were analyzed statistically, a significant difference was observed in study group before and after the study (P= 0.000) but difference was not significant (P= 0.439) in control group which had received placebo capsules. Negative correlation was observed between Number of cigarettes per day and serum total thiol, indicating that when the number of cigarettes smoked per day increases, serum total thiol decreases. It can be due to the increase in oxidation of thiol molecule.

Shallot (*Allium ascalonicum* L.), belonging to the Family Alliaceae, is one of the promising plants which demonstrates significant antioxidant as well as anti-inflammatory properties, useful in the protection of various diseases such as respiratory and nervous diseases. The important activity compounds in Alliaceae family such as onion, garlic or shallot are different but they mainly contain total phenolic compound that have the –OH group. In addition, most

of phenolic compounds such as furostane saponins and high level of quercetin, isorhametin and other glycosides are present in shallot (Fattorusso et al., 2002).

The major cause of chronic oxidative stress in humans is exposure to free radicals in cigarette smoke. Cigarette smoke free radicals are considered an important cause of atherosclerosis and cancer. Thiols are powerful reducing agents that are capable of acting as antioxidants *in vivo*. Thiols exist in three forms: freethiol and two types of disulfides, namely, homodisulfides and heterodisulfides. Several aminothiols, e.g., cysteine, homocysteine (Hcy), and GSH, and disulfides (e.g., cystine, homocystine, and oxidized glutathione), interact by means of redox and disulfide exchange (Iciek et al., 2004). This dynamic system (with respect to thiol status) is important for normal physiologic function (Morris, 2002). Changes in the redox thiol status lead to the induction of oxidative stress and apoptosis. As both an intracellular and extracellular redox buffer, t-SH plays important roles in the *in vivo* prevention of atherosclerosis (Ueland et al., 1996). t-SH plays a prominent role in antioxidant reactions, and in catalysis, regulation, and electron-transport reactions, and in reactions that preserve the correct structure of proteins. Mixed disulfides with proteins are formed by reaction of S-thiolation, in which protein thiols conjugate with non-protein thiols (Klatt and Lamas, 2000). This process plays a regulatory and an antioxidant role, since it protects protein −SH groups against irreversible oxidation from −SO2H and −SO3H; moreover, it participates in signal transduction (Padgett and Whorton, 1998). Maintaining the intracellular thiols, such as GSH, in their reduced form, may allow for

the maintenance of plasma homocysteine and other intracellular thiols in redox states (Moriarty et al., 2003). Since the plasma GSH concentration reflects its levels in various tissues, a reduced plasma concentration of GSH may be a diagnostic indicator of a pathological state (Ashfaq , 2006). GSH is the most important endogenous antioxidant in humans. It is often accompanying

by other endogenous thiols, such as cysteine, cysteinylglycine and even Hcy (in low concentration). These thiols scavenge ROS and are involved in preserving the prooxidant–antioxidant balance in human tissues (Zinellu ,2006). GSH is an abundant tripeptide that protects against oxidative stress and damage in nearlyall cells and tissues (Viña, 1990; 1998). It is the major intracellular antioxidant and functions by scavenging free radicals, detoxifying lipid peroxides via glutathione peroxidase, and conjugating reactive electrophilic toxicants and carcinogens. In addition, GSH is involved in numerous other cellular pathways including

protein and DNA synthesis, DNA repair, and immune surveillance. GSH is oxidized to its disulfide form (GSSG), but is subsequently reduced back to GSH by GSH reductase. An alternative pathway for GSSG metabolism is protein glutathiolation (also referred to as glutathionylation) where thiol–disulfide exchange occurs with cysteine (Cys) residues in proteins to form GSSP. The formation of GSSP within cells can be substantial and reach 200 μM in certain tissues (Kleinman et al., 2003). There is considerable evidence that glutathiolation represents an important redox-sensitive regulator of cellular activities (Sies et al., 1987; Klatt and Lamas, 2000). An induction of GSH synthesis may accompany a smoking-related increase in GSH utilization through oxidation to GSSG and GSSP, resulting in increases in both GSH and GSSP. This would suggest that GSSP: GSH ratios are less sensitive to oxidative stress than GSSP alone. The amount of protein that is glutathiolated in the blood of smokers is reported to be high (ranging from 0.05 to
0.38 mmol/l), 34 to 43% higher than those observed in nonsmokers. A dose–response relationship is reported to be apparent between GSSP levels and tobacco smoke measurements such as cigarettes smoked per day, blood cotinine (an alkaloid found in tobacco and also a metabolite of nicotine) and blood thiocyanate. A similar but smaller increase in plasma GSSP levels was also found in smokers compared with nonsmokers (Muscat , 2004). These findings provide compelling evidence that blood GSSP is an indicator of oxidative stress, and those abundant free radicals in cigarette smoke cause increases in blood GSSP concentrations. As shown in Figure 57 interestingly a negative correlation was observed between total thiol and No. of cigarette smoked.

Serum Lipid Per-oxidation

As explained earlier, smoking also produces xenobiotics and toxic compounds in the body. Many researchers have also reported lower serum vitamin C content in smokers compared with non- smokers. Thus lipid peroxidation increases in smokers. Serum lipid peroxidation was found to be 4.9 and 4.79 at the beginning of the study in study and control group respectively. After 30 days supplementation, estimation was repeated again and found to be 4.3 and 4.71 in study and control group respectively. Statistical analysis showed a significant reduction in the serum lipid peroxidation level of subject after shallot supplementation (P= 0.000), whereas in control, difference was not significant (P= 0.05). A negative correlation was observed between total thiol and lipid peroxidation, which shows that, when oxidative stress is more the total thiol molecule

reduces due to the oxidation (R2 = -0.369). Also a positive correlation (R2 = 0.267) was observed between serum lipid peroxidation and number of cigarettes. Liu and Wei (1999) reported that, the level of total blood glutathione is negatively correlated with the level of plasma lipid peroxides (r=−0.305,P=0.002) and was positively correlated with the smoking index (r=0.307, P=0.019) of all the study subjects.

These results indicate that the activities of glutathione peroxidase and glutathione*S*transferase reduced to a great extent under smoking-mediated oxidative stress in the blood of both young and aging smokers. Moreover, the compensatory generation of total blood glutathione may effectively prevent plasma lipids from peroxidation in young smokers, although the activities of glutathione peroxidase and glutathione*S*-transferase in plasma were decreased. By contrast, total blood glutathione was inadequate for such protection in the aging smokers. We suggest that supplementation of thiol-group-related agents may be considered for the prevention or alleviation of oxidative stress in aging smokers, whose capability and capacity for the disposal of smoking-mediated free radicals and reactive oxygen species are compromised Leelarungrayub et al (2004) evaluated the antioxidant potential of Thai shallot. It is reported that Thai shallot protect human erythrocytes from possible damage from external or internal radicals such as H2O2 or peroxyl radical such as 2,2'-Azobis (2- amidino-propane) dihydrochloride. They reported that thai shallot is able to inhibit lipid peroxidation and glutathione depletion in erythrocytes and suggested that the Thai shallot extracts have protective effect on the GSH deterioration *in vitro* from protein hydroperoxide (PrOOH) or hydroxyl radical from gamma irradiation (Leelarungrauyub et al., 2004). Thai shallot extracts can also protect and scavenge the protein and lipid hydroperoxide (LOOH) formation *in vitro* study (Leelarungrayub et al., 2004).

Total Antioxidant by FRAP assay

It has been reported that the smokers, because of their increased oxidative stress, had 12% lower FRAP than did the nonsmokers. These data suggest that smokers have less plasma antioxidant potential, which would be consistent with their greater plasma isoprostane concentrations (Bruno et al., 2005). Plasma uric acid is the greatest predictor of FRAP and accounts for 60% of the total predicted FRAP, whereas ascorbic acid contributes to 15% of the value (Benzie and Strain, 1996). Results revealed that significant increase was there in experimental group whereas changes in control group were not significant. Slightly positive correlation was observed between serum total antioxidant capacity and total thiol molecule. A

negative correlation was seen between serum total antioxidant capacity and No. of cigarettes smoked per day, whereas correlation between serum total antioxidant capacity and lipid peroxidation level and between TAC and total thiol molecule was found to be positive**.**

REFERENCES:

Ansari P (2012). "The effect of ginger on diabetic nephropathy, plasma antioxidant capacity and lipid peroxidation in rats." Food Chemistry 101(1): 148153.

Jadon D (2011). "Antibacterial activity of Allium cepa (Onions) and Zingiber officinale (Ginger) on Staphylococcus aureus and Pseudomonas aeruginosa isolated from high vaginal swab." The Internet Journal of Tropical Medicine 3(2): 110-115.

Bhatt M (2011). "Cancer chemopreventive activity of resveratrol." Annals of the New York Academy of Sciences 957(1): 210-229.

Gaffar A (2010). "Flavonoid, hesperidine, total phenolic contents and antioxidant activities from Citrus species." African Journal of Biotechnology 9(3): 326-330.

Jayaprakasha C (2010). "Antioxidant activity of grape seed (Vitis vinifera) extracts on peroxidation models in vitro." Food Chemistry 73(3): 285290.