

Effect of cannabis smoking on Vitamin C and E levels of male cannabis smokers in Nnewi, Nigeria

I.P. Ezeugwunne¹, J.E. Okwara², S. Onogwu³, E.C. Ogbodo^{4*}, I.J. Onuora⁵, R.A. Analike⁶, A.K. Amah⁷, C.N. Obi-Ezeani⁸, D.C. Ejiofor⁹, C. Nwanegwo¹⁰

¹Lecturer, ^{2,9,10}Senior Lecturer, ^{3,4}Medical Laboratory Scientist, ^{5,6}Chief Medical Laboratory Scientist, ⁷Associate Professor, ⁸Senior Medical Laboratory Scientist, ¹Dept. of Human Biochemistry, ^{2,5,6,8}Dept. of Chemical Pathology, ^{3,4}Dept. of Medical Laboratory Science, ^{7,9,10}Dept. of Human Physiology, ¹⁻⁶Nnamdi Azikiwe University, Nnewi, Nigeria, ^{7,9,10}Imo State University, Owerri, Nigeria, ⁸Chukwuemeka Odumegwu Ojukwu University, Awka, Nigeria

*Corresponding Author: E.C. Ogbodo

Email: augustinee442@gmail.com

Abstract

Smoking has been known to cause a wide range of deleterious effects on human health but the impact of cannabis use on the antioxidant vitamins are not well known. In this study, we evaluated the effect of cannabis smoking on antioxidant vitamin C and E levels of male cannabis smokers in Nnewi, Nigeria. A total of hundred (100) male participants aged between 18 and 45 years (50 cannabis smokers and 50 non-smokers) were recruited for the present study. Also, the anthropometric indices (height, weight and body mass index (BMI) of the participants were measured while other relevant data such the feeding habit, life style and age of the subjects were obtained using a structured questionnaire. Afterwards, 5 ml of venous blood sample was collected from each participant into plain containers and used for laboratory analysis by using standard methods. There was a significant decrease in the vitamin C and E levels in cannabis smokers than in non-smokers ($p=0.000$), whereas, the weight and BMI of both groups were similar ($p>0.05$). This study has shown that cannabis smoking depletes the antioxidant vitamins C and E status in users. Thus, antioxidant supplementation may be necessary in the diet of cannabis smokers as a way of boosting their antioxidant status.

Keywords: Smoking, Cannabis sativus, Marijuana, Oxidative stress, Antioxidant, Vitamin C, Vitamin E.

Introduction

Smoking is the act of inhaling and exhaling the fumes of burning plant material. A variety of plant materials are smoked including marijuana (*Cannabis sativa*) and hashish, but the act is most commonly associated with tobacco as smoked in a cigarette, cigar or pipe.¹ Cannabis is consumed by various routes with the most common route being smoking, followed by vaporization and then by the oral route. Cannabis products may be taken by ingesting edibles, sublingual or rectal administration via transdermal delivery, eye drops and aerosols.² *Cannabis sativa* is a dioecious plant of the Cannabaceae family and it is widely distributed all over the world.³ *Cannabis sativa* has been an important source of food, fiber and medicine for thousands of years in the Old World.⁴⁻⁷ Cannabis is the most commonly cultivated, trafficked, and abused illicit drug worldwide; according to the World Health Organization (WHO), marijuana consumption has an annual prevalence rate of approximately 147 million individuals or nearly 2.5% of the global population.⁸ The fiber from the stalk is still used in the modern production of durable fabrics and special papers in some countries; e.g. as canvas, linen, tea bags, paper money, cigarette papers and other strong, thin papers that are used to make thick books. Both the ripened seed of hemp and seed meal are excellent sources of dietary oil, fiber and protein.⁸

Vitamins are a class of essential nutrients in the body; thus, they play important roles in human health. The chemicals are involved in many physiological functions and both their lack and excess can put health at risk.⁹ Vitamins are involved in the processes of normal metabolism and cell regulation, and they are necessary for growth and

development; thus, they are chemicals that we all need to stay healthy.¹⁰ Although about 4000 antioxidants have been identified, the best known are vitamin E,¹¹ vitamin C¹² and the carotenoids.¹³ The antioxidant effect is due to donation of an electron to a rampaging reactive oxygen species (ROS), which destroys it and its capacity to cause damage. Antioxidant properties of tocopherols have been known and exploited for some time. Traditional supplementation of tocopherols has primarily focused on its form. Many plants however, including hemp, tend to have significantly higher levels of gamma-tocopherol. Although both exhibit antioxidant activity, their differing metabolic paths confer other specific activities to their respective isomeric forms.¹⁴ The biological activity of alpha-tocopherol tends to be significantly higher than gamma-tocopherol as a result of its greater affinity to be secreted by the liver into very-low density lipoproteins.¹⁵ Gamma-Tocopherol has been shown to have significant antioxidant effects in vitro even at concentrations less than 50 ppm.¹⁶ Within hemp seed oil, gamma-tocopherol is present in significantly higher quantities than alpha-tocopherol. They both however, play an important role as antioxidants in their respective physiological systems. The additional bioactive properties they possess add to their benefits as components of the seed oil.¹⁴ The principal cannabinoids in the cannabis plant include Δ 9-tetrahydrocannabinol (THC), Cannabidiol (CBD) and cannabinal (CBN).¹⁷ THC is the primary psychoactive compound, with CBD, a non-psychoactive compound, ranking second.¹⁸ In the ambit of non-psychoactive compounds, CBD represents the most valuable one from the pharmaceutical point of view, since it has been found to

possess a high antioxidant and anti-inflammatory activity, together with antibiotic, neuroprotective, anxiolytic, and anticonvulsant properties.³ The THC content is highest in the flowering tops, declining in the leaves, lower leaves, stems and seeds of the plant.¹⁹ Cannabidiol (CBD) at lower doses has physiological effects that promote and maintain health, including antioxidative, anti-inflammatory, and neuroprotection effects. For instance, CBD is more effective than vitamin C and E as a neuroprotective antioxidant and can ameliorate skin conditions such as acne.²⁰ Direct measurement of oxidative stress reveals that cannabinoids prevent cell death by antioxidation. The antioxidative property of cannabinoids is confirmed by their ability to antagonize oxidative stress and consequent cell death induced by the powerful oxidant, retinoid anhydroretinol. Cannabinoids also modulate cell survival and growth of B-lymphocytes and fibroblasts.²¹

However, several authors have documented conflicting reports regarding the effect of cannabis use on the biomarkers of oxidative stress.²²⁻²³ Previously, some authors in this area had noted the effect cannabis smoking on both the kidney function and lipid profile levels in cannabis smokers.²⁴⁻²⁵ However, there is paucity of information regarding the effect of cannabis smoking on antioxidant status of cannabis smokers especially in the present area under study. Therefore, the present study is geared towards evaluating the effect of cannabis smoking on vitamin C and E levels of male cannabis smokers in Nnewi, Nigeria.

Materials and Methods

Study Area

This study was carried out in Nnewi North Local Government Area of Anambra State, Nigeria.

Study Design

This is a cross-sectional study designed to assess the levels of vitamin C and E among cannabis smokers in Nnewi North, Anambra State, Nigeria. The protocol was explained to the intending participants and those who gave informed consent were randomly recruited. A total of hundred (100) male participants aged between 18 and 45 years (50 cannabis smokers and 50 non-smokers) were recruited for the present study. The height, weight and body mass index (BMI) of the participants were measured and feeding habit, life style and age of the subjects were obtained using a structured questionnaire. Afterwards, 5mls of venous blood sample was collected from each participant into plain containers and used for laboratory analysis. The serum vitamin C and E levels were estimated using standard methods as described by Omaye et al²⁶ and Jadoon et al²⁷ respectively.

Ethical Consideration

This was obtained from the Faculty of Health Sciences and Technology Ethics Committee of Nnamdi Azikiwe University, Nnewi, Nigeria.

Inclusion Criteria and Exclusion Criteria

Apparently healthy male cannabis smokers aged 18 to 45 years were included in the present study but cannabis users taking vitamins C and E supplements, diabetics and

alcoholics, female cannabis users and those younger than 18 years or older than 45 years were not allowed into this study.

Statistical Analysis

The data obtained from this study was organized and subjected to statistical analysis using Statistical packages for social science (SPSS) version 20. Student t-test was used to compare differences between groups and was deemed significant at $P < 0.05$.

Results

Table 1 showed that there was a significant difference in the mean age of the subjects between the cannabis smokers (29.76 ± 5.03) and control group (24.40 ± 4.10) at $p < 0.05$. The weight of the subject showed that there was no significant difference between the cannabis smokers (62.28 ± 5.09) and control group (62.72 ± 6.22), ($p > 0.05$). Also, the result of BMI showed no significant difference between cannabis smokers and control group ($p > 0.05$).

Table 2 results show the independent t-test analysis comparing the level of Vitamin C and E level of cannabis smokers and non-smokers. There was a significant decrease in the mean level of vitamin C in cannabis smokers compared with non-smokers ($p = 0.000$). Also, the mean level of Vitamin E differed significantly between cannabis smokers and non-smokers ($p = 0.000$).

Discussion

Oxidative stress is considered as an imbalance between pro- and antioxidant species, which results in molecular and cellular damage.²⁸ Smoking may impact negatively on the antioxidant status of the body as a result of its ability to generate free radicals which may induce damage to cells by passing its unpaired electron resulting in oxidation of cell components and molecules.²⁹

Findings from this study showed no significant differences in mean weight and body mass index of cannabis smokers in comparison with non-smokers. The mechanism that explains the current result is not quite clear. This is in agreement with the report of some previous studies.³⁰⁻³¹ The present finding is not in keeping with the report of some previous studies.³²⁻³³ The result showed a significant reduction in the mean serum levels of both vitamin C and E in cannabis smokers than in non-smokers. The reason for the decreased serum levels of these vitamins (C and E) may be attributed to the generation of reactive oxygen species (ROS) by cannabinoid leading to increase in oxidative stress which tends to deplete the antioxidant balance. Oxidative stress leads to changes in the cellular structure and function which may ultimately culminate in cell death.³⁴ Due to the paucity of information regarding the present study, we could not make a direct comparison with any previous finding involving the effect of cannabis smoking on serum vitamin C and E levels. However, our findings corroborate the previous findings of some studies on the effect of cigarette smoking on antioxidant status.³⁵⁻³⁷

Table 1: Anthropometric indices in subjects studied

Groups	Age (years)	Weight (kg)	Height (m)	BMI (Kg/m ²)
Cannabis smokers (n=50)	29.76±5.03	62.28±5.09	1.69±0.10	22.19±3.22
Control (n=50)	24.40±4.10	62.72±6.22	1.69±0.00	22.12±2.94
p-value	0.000	0.722	1.000	0.918
t-value	0.820	0.022	0.000	0.300

*Statistically significant at $p < 0.05$; CS=Cannabis smokers.

Table 2: Level of serum vitamin C and E between smokers of cannabis sativus and Non-smokers

Variables	Groups	Mean±SD	p-value	t-value
Vitamin C	CS (n=50)	0.82±0.33	0.000	7.980
	Control (n=50)	1.55±0.33		
Vitamin E	CS (n=50)	11.92±1.45	0.000	7.624
	Control (n=50)	15.57±2.03		

*Statistically significant at $p < 0.05$; CS=Cannabis smokers

Conclusion

This study has shown significant reductions in the serum levels of Vitamin C and E in cannabis smokers than in non-smokers. Thus, antioxidant supplementation may be necessary in the diet of cannabis smokers as a way of boosting their antioxidant status.

Conflict of Interest: None.

References

- Matthew JH, Christine AR, David TS, Jack H. Smoking Tobacco. *Br Popular Culture* 2019;1800-2000.
- Baggio S, Decline S, Studer J, Mohler-Kuo M, Daepfen JB. Routes of administration of cannabis used for non-medical purposes and associations with patterns of drug use. *J Adolesc Health* 2014;54:235-40.
- Hartsel JA, Races J, Hickory B, Makriyannis A. Cannabis sativa and hemp. *Nutraceuticals, Efficacy, Saf Toxicity J* 2016;53:735-54.
- Zias J, Stark H, Sellgman J, Levy R, Werker E, Breuer A, Mechoulam R. Early medical use of cannabis. *Nat* 1993;363(6426):215.
- Xiaozhai L, Clarke RC. The cultivation and use of hemp (*Cannabis sativa* L.) in ancient China. *J Int Hemp Assoc* 1995;2(1):26-33.
- Pringle H. Ice age community may be earliest known net hunters. *J Sci* 1997;277:1203-4.
- de Padua LS, Bunyaprafatsara N, Lemmens MJ. Plant Resources of South-East Asia. Medicinal and Poisonous Plants 1999; 1(12):167-175.
- WHO (2016). Management of substance abuse: cannabis. Retrieved May, 2019.
- Yuan Z, Wei Z, Jia-qing Y, Min L, Yu Z, Xin S, Ying-lin M, Xue-song F, et al. A review of the extraction and determination methods of thirteen essential vitamins to the human body: An update from 2010. *J Mol* 2018;23:1484.
- Khayat S, Fanaei H, Ghanbarzahi A. Minerals in pregnancy and lactation: A Review Article. *J Clin Diagn Resolution* 2017;11:1-5.
- Halliwel B, Rafter J, Jenner A. Health promotion by flavonoids, tocopherols, tocotrienols and other phenols: direct or indirect effects? Antioxidant or not?. *Am J Clin Nutr* 2005;81:268-76.
- Vattasery GT, Smith WE, Quach HT. Ascorbic acid, glutathione and synthetic antioxidants prevent the oxidation of vitamin E in platelets. *J Lipids* 1989;24:299-304.
- Burton GW, Ingold KU. Beta-carotene: an unusual type of lipid antioxidant. *Sci J* 1984;224:569-73.
- Cary L, David R, Alexander PS, Dushenkov IR. The Composition of Hemp Seed Oil and Its Potential as an Important Source of Nutrition. *J Nutraceuticals* 2000;2(4):2000.
- Stone W, Papas A. Tocopherols and the etiology of colon cancer. *J Natl Cancer Inst* 1997;89(14):1006-14.
- Lampi A, Hopia A, Piironen V. Antioxidant activity of minor amounts of tocopherol in natural triacylglycerols. *J Am Oil Chemists Soc* 1997;74(5):549-55.
- Maione S, Costa B, Di Marzo V. Endocannabinoids: A unique opportunity to develop multitarget analgesics. *Pain* 2013;154:S87-S93.
- Bertha KM. Update of Cannabis and its medical use. 37th Expert Committee on Drug Dependence 2015;3-41.
- Adams IB, Martin BR. Cannabis: pharmacology and toxicology in animals and humans. *J Addict* 1996;91:1585-1614.
- Hampson AJ, Grimaldi M, Axelrod J. Cannabidiol and Δ^9 -tetrahydrocannabinol are neuroprotective antioxidants. *J Sci* 1998;95:8268-73.
- Chen Y, Buck J. Cannabinoids protect cells from oxidative cell death: a receptor independent mechanism. *J Pharmacol Expert Ther* 2000;293(3):807-12.
- Sarafian TA, Magallanes JAM, Shau H, Tashkin D, Roth MD. Oxidative stress produced by marijuana smoke: an adverse effect enhanced by cannabinoids. *Am J Respir Cell Mol Biol* 1999;20:1286-93.
- Bloomer RJ, Butawan M, Smith NJG. Chronic marijuana smoking does not negatively impact select blood oxidative stress biomarkers in young, physically active men and women. *Health* 2018;10:960-70.
- Analike RA, Emekwue L, Ogbodo EC, Ezeugwunne IP, Onoh JO, Amah UK, et al. Evaluation of renal function of apparently healthy male cannabis smokers of College of Health Science and Technology, Azikiwe University, Nnewi Campus, Anambra State, Nigeria. *Int J Sci Eng Invention* 2018;4(1):1-6.
- Okwara JE, Onoh JO, Ogbodo EC, Ezeugwunne IP, Emekwue L, Amah UK, et al. Evaluation of serum lipid profile in male cannabis smokers of College of Health Science and Technology, Azikiwe University, Nnewi Campus, Anambra State, Nigeria. *Int J Health Sci Res* 2018;8(2):1-6.
- Omaye ST, Turnbull JD, Sauberlich HE. Selected methods for the determination of ascorbic acid in animal cells, tissues and fluids. *Methods Enzymol* 1979;62:3-10.
- Jadoon S, Waseem A, Yaqoob M, Nabi, A. Flow injection Spectrophotometric determination of vitamin E in pharmaceuticals, milk powder and blood serum using potassium ferricyanide-Fe(III) detection system. *Chin Chem Lett* 2010;21(6):712-5.

28. Conti V, Izzo V, Corbi G, Russomanno G, Manzo V, De Lise F. Antioxidant supplementation in the treatment of aging-associated diseases. *Front Pharmacol* 2016;7:24.
29. Bansal AK, Bilaspuri GS. Impacts of oxidative stress and antioxidants on semen functions. *Vet Med Int* 2010;2011:686137.
30. Rodondi N, Pletcher MJ, Liu K. Marijuana use, diet, body mass index, and cardiovascular risk factors (from the CARDIA study). *Am J Cardiol* 2006;98:478–84.
31. Cluny NL, Keenan CM, Reimer RA, Le Foll B, Sharkey KA. Prevention of diet-induced obesity effects on body weight and gut microbiota in mice treated chronically with Δ^9 -Tetrahydrocannabinol. *PLoS ONE* 2015;10(12):e0144270.
32. Hayatbakhsh MR, O'Callaghan MJ, Mamun AA, Williams GM, Clavarino A, Najman JM. Cannabis Use and Obesity and Young Adults. *Am J Drug Alcohol Abuse* 2010;36(6):350-6.
33. Rajavashisth TB, Shaheen M, Norris KC. Decreased prevalence of diabetes in marijuana users: cross-sectional data from the National Health and Nutrition Examination Survey (NHANES) III. *Br Med J Open* 2012;2:e000494.
34. Jancsó Z. Study of oxidative stress and the antioxidant defense system after heavy metal treatment in carp and in streptozotocin-induced diabetic rats [Dissertation]: submitted to Department of Biochemistry and Molecular Biology, Faculty of Science and Informatics, University of Szeged, Szeged 2015.
35. Hauwa'u AB, Abdullahi D, Gaddafi ID. Effects of cigarette smoking on lipid peroxidation and serum antioxidant vitamins. *IOSR J Pharm Biol Sci* 2015;12(2 Ver. IV):40-4.
36. Okwara JE, Ibe CS, Ogbodo EC, Analike RA, Onyegbule OA, Oguaka VN, et al. The effects of cigarette smoking on the serum levels of some antioxidants (Vitamin C and E) amongst male smokers in College of Health Sciences, Nnamdi Azikiwe University, Nnewi Campus, Anambra State, Nigeria. *Int J Sci Res Manag* 2018;6(1):6-11.
37. Veysel KC, Imge EE, Gorsel Y. Vitamin E and antioxidant activity; its role in slow coronary flow. *Cardiovasc J Afr* 2013;24(1):360-4.