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CITRUS FRUITS AS A SOLUTION TO NICOTINE ADDICTS

[1]Agneswari. S, [2]Sreeya G. Nair, [3]C. Christo Queensly, [4]Meera Sabari. V

[1]Assistant Professor, Department of Zoology, S.T. Hindu College, Nagercoil, affiliated to Manonmaniam Sundaranar University, Tirunelveli, Tamil Nadu, India.
[2]Assistant Professor, Department of Zoology, Sree Ayyappa College for Women, Chunkankadai, affiliated to Manonmaniam Sundaranar University, Tirunelveli, Tamil Nadu, India.
[3]Assistant Professor, Department of Zoology, The American College, Madurai, India.
[4]Department of Biotechnology, Noorul Islam College of Arts & Science, Kumaracoil, affiliated to Manonmaniam Sundaranar University, Tirunelveli, Tamil Nadu, India

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ABSTRACT

Smoking is one of the bad habits of men which is a significant risk factor for developing many diseases. Various medications and counseling can overcome this bad habit, but having a good diet with fruits and vegetables protect the body from smoking related complications because of their nutritive value and bioactive compounds. Hence, our study was intended to compare and assess the phytochemical constituents, vitamin C content, antibacterial and antioxidant activities of Citrus fruits such as *Citrus sinensis*(Orange), *Citrus limon* (Lemon), and *Vitis vinifera* (Grapes) so as to suggest the best fruit for nicotine patients.The findings of the study clearly suggest *Citrus limon* (Lemon) as the best citrus fruit among the three citrus fruits, with regard to its phytochemical content, Vitamin C content, antibacterial and antioxidant activity.

Key words: Smoking, nicotine, Citrus fruits, Phytochemicals, Antimicrobial activity, Antioxidant activity.

INTRODUCTION

Smoking is a practice in which a substance is burned and the resulting smoke breathed into be tasted and absorbed into the blood stream. Most commonly the substance is the dried leaves of the tobacco plant which have been rolled into a small square of rice paper to create a small , round cylinder called a cigarette. Smoking generally has negative health effects because smoke inhalation inherently possess challenges to various physiologic processes especially respiration. In case of cigarette smoking active substances are contained in a mixture of aerosol particles and gases and include the pharmacologically active alkaloid nicotine, a drug primarily responsible for a persons addiction to tobacco products including cigarettes. The addiction to cigarettes and other tobacco products that nicotine causes is similar to the addiction produced by using drugs such as heroin and cocaine (Hatsukami *et al*, 2008).[6]Smoking is the leading cause of many diseases such as lung cancer, heart attack, COPD, erectile dysfunction, and birth defects.. The term nicotine is named after a Frenchman, Jean Nicot who introduced tobacco to France in 1560.Smoking harms nearly every bodily organ and organ system in the body and diminishes a persons overall health. Smokers are at the higher risk of developing pneumonia, tuberculosis, and other airway infections. In addition smoking causes inflammation and impairs immune function. (Atlanta, 2014)[1].The majority of smokers ailments are

the result of free radicals. Though free radicals are formed during everyday functions such as breathing and eating , environmental stress factors such as smoking accelerate their production. Vitamin deficiency is yet another problem associated with nicotine addiction. Smokers have below normal levels of vitamin C as much as 40% lower in pack- a-day smokers. It has been noted that smokers are ill more often, directly related to low vitamin C levels. Vitamin C deficiency also leads to reduced iron absorption and blood cell production thus causing **vitamin C deficiency anaemia.**

Nicotine once ingested is absorbed and metabolized by the liver to byproducts such as cotinine, norcotinine etc (Nakajima *et al*, 2002)[7]. Nicotine acts via 3 major mechanisms , producing physiological and pathological effects on a variety of organ systems(Dani *et al.* , (2001)[3].Thus nicotine is one of the most toxic of all poisons and has a rapid onset of action. Healthy foods and lifestyle changes can help one eliminate the nicotine from body. Some of the best fruits to remove the nicotine from body includes broccoli, orange juice, lemon juice, strawberry, carrot juice, grape juice, kiwi fruit, spinach, water, milk, ginger, cabbage etc. Research suggest that restoring vitamin C in the body may also help reduce cigarette cravings. Citrus fruits of Rutaceae family are the more commonly known vitamin C rich foods(Oranges, grapes, & nectarines).Citrus fruits and juices are important sources of bioactive compounds including antioxidants such as ascorbic acid, flavonoids, phenolic compounds and pectins that are to human nutrition(Fernandez-Lopez. J et al in 2005).[4]

The aim of the present study was to compare between three types of citrus fruits viz: *Citrus sinensis, Citrus limon, and Vitis vinifera* to suggest the best one among the three for use by nicotine addicts to compensate the nutritional demand of body in response to nicotine.

MATERIALS AND METHODS

Collection of Citrus fruits : Matured *Citrus sinensis* (orange; MSP-I), *Citrus limon* (Lemon; MSP-2), and *Vitis vinifera* (Grapes; MSP-3) fruits devoid of any visual signs of bruises , cuts, blackening or infestations collected from local market at Nagercoil were used as samples and confirmed at the Research Department of Botany, S.T.Hindu College, Nagercoil.

The fruits were cleaned externally using 70% alcohol and the edible portion was subjected to a mixer for preparing fresh juice in laboratory. 100g of juice of fruits were weighed and then extracted separately with 200 ml 80% methanol and acetone yielding corresponding fractions using ordinary extraction. The extract was filtered using Whatman filter paper (No. 1) and concentrated in vacuum at 40°C using a rotary evaporator and kept at 4°C. Standarad procedures were employed to analyse extracts of three different Citrus fruits as described by [5], [9], [11], [12].

1. Phytochemical Screening

The methanol and acetone extracts collected was analysed qualitatively for the presence of various metabolites, minerals and vitamin C followed by quantitative estimation of total carbohydrates, proteins, aminoacids, and Vitamin C content.

2. Antibacterial activity of Citrus fruits

Antibacterial activity of methanol extract of *Citrus sinesis, Citrus limon* and *Vitis vinifera* (50µl) were performed by Kirby Bauer agar well(6mm) diffusion method against respiratory pathogens such as *Pseudomonas aeruginosa* and *Klebsiella pneumoniae* likely to infect nicotine addicts.Clinical strains used for the study was collected from Vivek clinical laboratory, Nagercoil. Commercially available discs of ofloxacin was used as positive control and DMSO was used as negative control. This assay was done in triplicates and the results were expressed in terms of diameter of the zone of inhibition.

3. Antioxidant activity of Citrus fruits

Antioxidant activity of fruit samples were done by ferrous iron chelating assay.Reaction mixtures containing 0.1ml of the methanolic extract of 3 fruits at different concentrations (2,4,8,12,16,20mg/ml), 0.1ml of 2Mm ferric choride and 0.2ml of 5Mm ferrozine were incubated at 37ºC for 10 min. After adding 1.5ml double distilled water to the mixture, the absorbance at 562nm was measured. This assay was done in triplicates. The percentage of ferrous iron chelating ability is expressed by [1-(test sample absorbance/blank sample absorbance] x 100.

Results and Discussion

1. **Phytochemical analysis**

The preliminary phytochemical analysis (Table 1) of citrus fruit juice using methanol and acetone revealed the presence of many phytochemicals in methanol extract than acetone in all the three Citrus fruits. Following qualitative analysis fruit extracts were quantitatively analysed for estimating the total protein, amino acid, carbohydrates, and vitamin C content.(Table 2 & 3).Among the three citrus fruits, Citrus limon (lemon) was found to posses higher amount of phytochemicals and vitamin C content. Sabari & Sujitha in 2012[8] analysed and reported the presence of various phytochemicals suchas proteins, aminoacids, carbohydrates, vitamin, alkaloids, flavonoids, saponins, minerals such as phosphate, chloride etc in three fruits such as papaya, custard apple, & grape fruit using water, acetone and benzene as solvents.

2. **Antibacterial activity of Citrus fruits**

Antibacterial activity testing against respiratory pathogens such as *Pseudomonas aeruginosa* and *Klebsiella pneumoniae* revealed maximum activity with *Citrus limon* with a diameter of zone of inhibition of 16mm and 17mm against both pathogens, while orange and grapes exhibited no effect.(Table 4). This result correlates with the results of Adham in 2015[2] who evaluated the antimicrobial activity of juice extracts of *Citrus limon*, *C. sinensis* and *C.paradisi* by agar well diffusion method against *Staphylococcus aureus, Streptococcus mitis, K. pneumoniae* etc and reported highest activity with extracts of *Citrus limon* ranging from 15mm-22mm.

3. **Antioxidant activity of Citrus fruits**

The citrus fruit extracts tested for their ability to scavenge the free radicals using ferrous iron chelating assay , at various concentrations, also revealed *Citrus limon* as the best antioxidant possessing citrus fruit than the rest two fruits(Table 5). Gorinstein etal, in 2001[10], estimated the vitamin C content of lemon, orange, and grape fruit extracts and compared with their total radical–trapping antioxidative potential. Their results revealed significant difference in the vitamin C content with extracts of lemon showing higher content than orange and grapes. Similarly, the antioxidant potential was also found significantly higher in lemon extracts than the rest two citrus fruits, thus correlating with the findings of present work

Conclusion

The above findings clearly suggest lemon *Citrus limon* (MSP-2) as the best Citrus fruit among the three compared Citrus fruits, with regard to its phytochemical content, Vitamin C content, antibacterial & antioxidant activity.
This significant results provide a fruitful information to nicotine addicts revealing the fact that consumption of lemon juice continuously can increase the detoxification of nicotine present in the body of smokers , because of its highest Vitamin C content and antioxidant activity. In addition , smoking related respiratory infection can also be controlled with lemon extracts . Yet, more fruitful factors associated with this fruit, if any, has to be reckoned in future by the scientific community for the benefit of society. Further, a detailed investigation of nicotine and

activity of lemon under invivo condition is required so as to design a powerful drug which can be made available to the market for use by nicotine addicts

S NO.	Compound	Fruits					
		MSP 1		MSP 2		MSP 3	
		M	A	M	A	M	A
1	Carbohydrate	+	+	+	+	+	+
2	Amino acid	+	+	+	+	+	+
3	Protein	+	+	+	+	+	+
4	Vitamin	+	+	+	+	+	+
6	Phenolic compounds	+	+	+	+	+	+

M-Methanol, A-Acetone, '+'Positive, '-' Negative.

Table : 1 Preliminary analysis of phytoconstituents in citrus fruits

S.No.	Name of Sample	Name of the Test		
		Carbohydrates (mg / dl)	Protein (mg / dl)	Amino acid (mg /dl)
1	MSP 1	25	30	30
2	MSP 2	45	40	45
3	MSP 3	20	20	25

Table:2 Quantitative estimation of primary metabolites in citrus fruits

S.No.	Name of Sample	Vitamin C (mg / dl)
1	MSP 1	44
2	MSP 2	47
3	MSP 3	21

Table: 3 Quantitative estimation of vitamin C in citrus fruits

Sample	Zone of inhibition (mm)	
	Pseudomonas aeruginosa	Klebsiella pneumonia
MSP 1	No effect	No effect
MSP 2	16	17
MSP 3	No effect	No effect
Positive control	25	26
DMSO	_	_

Table: 4 Antibacterial activity of citrus fruits

S.No.	Sample	Percentage of ferrous ion chelated Concentration		
		1 mg / ml	2 mg / ml	3 mg / ml
1	MSP 1	39 ± 0.47	70 ± 0.47	81 ± 0.47

2	**MSP 2**	**72.3 ± 0.94**	**77 ± 0.81**	**84 ± 0.81**
3	**MSP 3**	**16.3 ± 0.94**	**58 ± 0.47**	**64.3 ± 0.94**

Table: 5 Antioxidant activity of citrus fruits

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