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## Lifestyle Pattern and Food and Nutrient Intake among Hyperlipidemics

#### Choudhary Mayuri M.\*, Nalwade Vijaya M.\*\*

#### Abstract

Hyperlipidemia is one of the main risk factor associated with the high prevalence of cardiovascular complications. Present study was conducted to find out the lifestyle pattern and food and nutrient intake among hyperlipidemics. On the basis of lipid profiles, 30 hyperlipedemic subjects (men and women) of 40 to 60 years of age were selected from Parbhani city of Maharashtra State. The information was collected on family history, awareness and exercise pattern of hyperlipidemics. Food intake and nutrient intake was studied and nutrient adequacy was calculated. Results indicated that 66 per cent of the hyperlipidedmic subjects had family history of hyperlipidaemia and it was mostly inherited from mother. Maximum (23) number of subject experienced blood pressure fluctuation followed by breathlessness (21) and dizziness (20). More than 50 per cent subjects knowing the meaning and causes of hyperlipidemia. Majority (86.66 %) of the subjects performed daily walking as an exercise pattern followed by yoga and sports. Intake of pulses, green leafy vegetables, milk and milk products, fruits and sugar and jaggary was found to be more by the hyperlipidemic women than those by hyperlipidemic men. The diet of selected hyperlipidemic subjects was adequate in providing maximum nutrients in needed amounts as per the recommended dietary allowances for adults specially the adequacy of fat intake by men (201 %) and women (276 %) was highest among the nutrients. In conclusion, it can be said that modifications in the lifestyle and dietary pattern of hyperlipidemics needs to be followed to lead a healthier life.

Keywords: Hyperlipidemia; Food and Nutrient Intake; Awareness; Exercise Pattern.

#### Introduction

Health of all is an enduring vision that recognizes the oneness of humanity and therefore there is a need to promote health universally. According to World Health Organization, today's emphasis is on health expectancy rather than life expectancy. In Asian countries, due to globalization, urbanization and increasing life span there is a great change in life style of people during the past four decades. This change has led to a change in disease pattern as infectious diseases have gradually diminished and

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diet related lifestyle diseases such as obesity, diabetes, hypertention, cardiovascular diseases and some types of cancer have become emerging health problems in India (Musaiger, 2003).

Hyperlipidemia is one of the main risk factor associated with the high prevalence of cardiovascular complications. Globally, a third of ischaemic heart disease is attributable to high cholesterol. Raised total cholesterol is a major cause of disease burden in both the developed and developing world as a risk factor for Ischemic heart disease and stroke. Overall, it is estimated to cause 2.6 million deaths (4.5 % of total) and 29.7 million disability adjusted life years (WHO, 2016). The results of a study carried out in Tamil Nadu, Maharashtra and Jharkhand by Joshi et al., (2014) have disclosed that around 13.9 per cent population had hypercholesterolemia, 29.5 per cent had hypertriglyceridemia, 72.3 per cent had low HDL-C, 11.8 per cent had high LDL-C levels and 79 per cent had abnormalities in one of the lipid parameters. Obesity, diabetes and dysglycemia were the common significant risk factors for dyslipidemia. Asmare

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(2014) revealed that there are significant differences in blood lipid levels and the prevalence of hyperlipidemia between ethnic groups, different dietary habits, life style and level of physical activity, as well as genetic background. Also exogenous factors, such as dietary intake (fat, cholesterol), alcohol, use of contraceptives and other pharmacologic agents are indicated as the main secondary causes and risk factors of hyperlipidemia in adults. This indicates that there are many inter-related causes and risk factors which lead to development of hyperlipidemia and other chronic illnesses of metabolic syndrome. Keeping this in view the study was carried out with the following objectives.

- 1. To study the lifestyle pattern among hyperlipidemics.
- 2. To study the food and nutrient intake by the hyperlipidemics.

#### Methods

#### Selection of Hyperlipidemic Subjects

A total number of 50 subjects of 40 to 60 years of age were selected randomly based on their past medical history and blood cholesterol level, residing in Parbhani city of Maharashtra State. All the selected subjects were screened for their lipid profile in the *'Pranav* pathology laboratory' with the help of pathologist. On the basis of lipid profiles, 30 hyperlipedemic subjects were selected for the present study. Thirty hyperlipidemic subjects comprised of men (12) and women (18). The study was approved by institutional ethics committee and an informed written consent in local language was taken from all the subjects prior to their recruitment in the study.

#### Collection of General Information of the Selected Subjects

All the hyperlipidemic (30) subjects were personally interviewed by the investigator with the help of pretested questionnaire so as to elicit the information on their socio-economic background, age, sex, history of diseases, exercise pattern and awareness about hyperlipidemia and health problems.

# Determination of Intake of Food and Nutrient by the Selected Subjects

Food intake of the selected hyperlipidemic subjects per day was collected by 24 hour recall method. The type of food preparation and amount of each food preparation consumed by all the selected subjects and the raw ingredients used for each preparation was recorded. Quantities of food consumed were converted into raw equivalents of food items to determine the food intake per day. The intake of different nutrients per day by the selected hyperlipidemic subjects was then calculated from the food nutrient values using nutritive value of Indian Foods. (Gopalan *et al.*, 2004).Per cent adequacy of different nutrients in the diet of the selected hyperlipidemic subjects was calculated based on the recommended dietary allowances.

In order to draw valid conclusions from the present study, the collected data were consolidated, computed and exposed for statistical analysisusing Indian NARS (National Agricultural Research System) statistical computing portal (http://www.iasri.res. in/sscnars/).

#### **Results and Discussion**

Socio-economic status of the selected hyperlipidaemic subjects is given in Table 1. Out of 30 selected hyperlipidemic subjects, equal number of subjects were from 40-50 and > 50-60 years of age group. Majority (18) of the subjects were female and the remaining (12) were male. Maximum (66.66) per cent of subjects were belonging to nuclear family and the remaining 33.33 per cent were from joint family. Out of 30 hyperlipidaemic subjects, more per cent of subjects were graduate (26.66) and post graduate (26.66), whereas seven subjects had school education and five completed higher secondary education. Only two subjects were not educated.

A relatively high (66.66) per cent of subjects were belonging to income group of Rs. >25,000 to  $\geq$ 50,000. Thirty per cent of the selected subjects had a monthly family income of Rs.  $\geq$ 10000 to 25,000 and the remaining 3.33 per cent of the subjects were from families of monthly income of Rs. < 10,000. Maximum per cent of the selected hyperlipidaemic subjects were engage in doing service (43.33) followed by home maker (36.66) as an occupation. Even six subjects were found to be businessmen.

On the whole, results indicated that the subjects were belonging to 40 to 60 years of age. Majority of the selected subjects were female, graduate, from nuclear families and belonging to families of monthly family income of Rs. >25,000 to 50,000. Service was the main occupation followed by home maker among the selected hyperlipidemic subjects.

In random selection for hyperlipidemic subjects it

was noticed that more number of females were suffering from hyperlipidemia. The selected female subjects were of >40 years of age which is a menopausal stage. Similar observations were also recorded by Sartika et al., (2015). This is probably due to the influence of the hormone estrogen prior to menopause (Martins, 2001). Estrogen can reduce the lipase activity of the heart so that HDL cholesterol remains high, and can increase LDL receptor activity, thus lowering LDL cholesterol and total cholesterol (O'Meara, 2004). When a woman regularly menstruates, it will protected by the hormone estrogen, being less likely to develop dyslipidemia until reaching menopause. Reduced estrogen levels during menopause can cause elevated levels of cholesterol (Martins, 2001).

Out of 30 selected hyperlipidaemic subjects, it was found that a relatively high (66) per cent of subjects had a family history of hyperlipidaemia. Out of these, 36.66 per cent subjects reported that it was inherited from mother whereas 30 per cent inherited from father. It was reported that 17 and 13 subjects had a family history of blood pressure and acidity respectively. Even three subjects had a family history of heart disease (Table 2).

On the whole, results indicated that majority of the hyperlipidedmic subjects had family history of hyperlipidaemia and it was inherited from mother. Nwamarah *et al.*, (2016) also reported that a greater percentage of respondents had a strong family history of hyperlipidemia which was inherited from father (37.5). On the contrary, in the present study, majority of the respondents inherited hyperlipidemia from maternal side (36.66 %).

Hyperlipidemia is associated with genetic disorders. Most hereditary lipid disorders are common among generations of families with obesity problems. Some familial lipid disorders can directly result in over production of cholesterol by the body. Another condition called 'Familial Combined Hyperlipidemia (FCHL)' can lead to high cholesterol levels including high triglyceride levels. Another hereditary condition called 'Familial Defective Apolipoprotein B-100', can cause the LDL blood cholesterol (also called the 'bad' cholesterol)to increase and also raise total cholesterol levels. (Shoulders et al., 2004). It was found that more than 55 per cent subjects had a history of blood pressure. Even Nwamarah et al., (2016) found that 25 per cent of hyperlipidemic respondents had hypertention. Many studies have demonstrated that dyslipidemia, one of the strong predictors of cardiovascular disease, causes endothelial damage and loss of physiological vasomotor activity. (Nickenig, 2002; Nickenig and Harrison, 2002 and Wong *et al.*, 2006). The damage may manifest as elevated systemic blood pressure (BP).

Information regarding health complications experienced by the selected hyperlipidemic subjects is given in Table 3. It was found that more than 65 per cent subjects had some or the other health complication. Maximum (23) number of subject experienced blood pressure fluctuation followed by breathlessness (21) and dizziness (20). Only three hyperlipidaemic subjects had undergone through heart surgery and two suffered from brain stroke. Hypercholesterolemia can also influence blood pressure by potentiating the effects on the endothelium of the vasoconstrictors endothelin-1 and Angiotensin II. (Cardillo et al., 2000 and Wierzbicki et al., 2002). Salkic et al., (2014) reported that dizziness and breathlessness were the complications found in hypertensive subjects. These results are in line with the results of the present study.

Awareness regarding hyperlipidemia among hyperlipidemic subjects is presented in Table 4. Majority (17) of the subjects were aware about the hyperlipidemia, even 12 subjects knew the causes of hyperlipidemia. Newspaper, television, magazine and physician were the various sources from which the subjects acquired the information about hyperlipidemia. It was found that all the selected hyperlipidemic subjects acquired the knowledge regarding hyperlipidemia from physician. Beside this more number of subjects acquired the information by reading newspaper (18), magazine (15) and television (11).

In nutshell, results inferred that more number of subjects knew the meaning of hyperlipidemia and causes of hyperlipedemia. Physician was the main source for acquiring the information about hyperlipidemia followed by reading newspaper and magazine among the selected subjects. Nwamarah *et al.*, (2016) conducted a study in Enugu State University Teaching Hospital (ESUTH) of Nigeria and found that about 45 per cent of the out-patients had a fair knowledge of hyperlipidemia whereas in the present study more per cent of the subjects were found to be aware about hyperlipidemia. It may be due to their consciousness about their condition which might have helped in improving the awareness.

It is clear from the data presented in Table 5 that a relatively very high per cent (86.66) of hyperlipidemic subjects followed walking daily as their exercise pattern. Sports activity as exercise was performed by 10 per cent of the selected subjects while only running was practiced by 6.66 per cent subjects and sports by three subjects. One third of total subjects were found to be performing yoga.Prasanthi and Amirthaveni (2013) reported that more than 95 per cent of the hyperlipidemic subjects were engaged in doing some or the other exercise. Even in the present study more than 86 per cent of the selected hyperlipidemic subjects were following some or the other exercise pattern daily. Exercise helps and plays an important role as that of diet and medicine. People with hyperlipidemia who exercise regularly requires less medicine, as it lowers the blood pressure and serum cholesterol. So the lifestyle changes inturn helps in many ways to live and lead a healthy life (Prasanthi and Amirthaveni; 2013).

Average intake of different foods per day by the selected hyperlipidemic men and women is presented in Table 6. Among the selected 30 hyperlipidemic subjects, an equal number of subjects were reported to be vegetarian (15) and non-vegetarian (15) and majority of the subjects were following three meal pattern.

Consumption of cereals and pulses per day by the selected hyperlipidemic men was 312 g and 54 g while it was 259 g and 58 g respectively by women. Intake of green leafy vegetables (g), other vegetables (g) and roots and tubers (g) was found to be 40, 92 and 63 respectively by the hyperlipedemic men. The respective values recorded for the intake of corresponding foods by hyperlipidemic women were 55, 87.5 and 51.5. The hyperlipidemic men and women were found to be consuming on an average 153 ml and 243 ml of milk and milk products, 20 g and 23 g of fruits, 24 g and 23.75 g of fats and oil and 9 g and 11.25 g of sugar and jaggary respectively. It is obvious from the results that on an average the intake of cereals, other vegetables, roots and tubers, fats and oils were more by hyperlipidemic men than that of the women. On the other hand, the daily intake of pulses, green leafy vegetables, milk and milk products, fruits and sugar and jaggary was more by the hyperlipedemic women.

In conclusion, it can be said that the consumption of pulses, green leafy vegetables, milk and milk products, fruits and sugar and jaggary was found to be more by the hyperlipidemic women than those by hyperlipidemic men. However the mean intake of different foods between men and women did not differ significantly. Premakumari and Haripriya (2010) also studied the food and nutrient intake of obese patients having higher lipid profile. The values reported for the intake of milk and milk products and fat was found to be more than that of values recorded in the present study. On the other hand, intake of cereals, green leafy vegetables and other vegetables was more by the hyperlipidemic subjects of present study while consumption of pulses and fruits was almost same.

The mean values of different nutrients supplied from the daily diet consumed by the hyperlipidemic subjects are given in Table 7.

The amount of energy (Kcal), protein (g), fat (g), iron (mg), calcium (mg),  $\beta$ -carotene ( $\mu$ g), thiamine (mg), riboflavin (mg), niacin (mg) and vitamin C (mg) supplied from the diet of the selected hyperlipidemic men was 1939.76, 62.12, 50.31, 19.29, 589.47, 2057.94, 1.8, 0.95, 19.3 and 51.56 respectively. The respective values of the corresponding nutrients for the women were 1843.74, 60.75, 55.14, 17.11, 748.27, 2882.2, 1.6, 0.97, 16.5 and 63.48. The results also indicated that the amount of energy, protein, iron, thiamin and niacin provided from the diet consumed by hyperlipidemic men were found to be more than that by hyperlipidemic women. On the other hand, the intake of nutrients like fat, calcium,  $\beta$ -carotene (µg), riboflavin and vitamin C was more by the women as compared to men. Moreover significant difference was not observed between mean intake of all the nutrients supplied from the diet consumed by the selected hyperlipidemic men and women.

It was found that the values recorded for intake of energy by the hyperlipidemic subjects was almost same with the values reported by Kuriyan *et al.*, (2010) whereas a study conducted by Premakumari and Haripriya (2010) indicated higher values for energy intake by the obese subjects having higher lipid profile values. The values obtained for protein intake in the present study found to be higher than the values reported by Kuriyan *et al.*, (2010) and Premakumari and Haripriya (2010). Kuriyan *et al.*, (2010) revealed that fat intake by the hyperlipidemic subjects was less while Premakumari and Haripriya (2010) reported more intake of fat than the values recorded in the present study.

Per cent of adequacy in the intake of different nutrients per day by the selected hyperlipidemic subjects is given in Table 8.

The adequacy (%) of the nutrient intake by the selected hyperlipidemic subjects for energy (Kcal), protein (g), fat (g), iron (mg), calcium (mg),  $\beta$ -carotene (µg), thiamine (mg), riboflavin (mg), niacin (mg) and vitamin C (mg) was 84, 103, 201, 113, 98, 43, 150, 68, 121 and 129 per cent respectively. whereas, corresponding values of the nutrients among hyperlipdemic women were 97, 110, 276, 81, 125, 60, 160, 88, 137 and 159 respectively.

The values of percent adequacy in the intake of energy and protein by the men and women varied from 84 to 103 and from 97 to 110 respectively. The per cent adequacy values of the intake of both the nutrients by the women were found to be more than those by men. Adequacy (%) of fat in the diet of women (276) was more than men (201). The values of adequacy (%) in the intake of iron was less by the women while it was more by the men, but it was vice-versa with regard to the per cent adequacy in the intake of calcium. The diet was grossly deficient in supplying  $\beta$ -carotene followed by riboflavin in both men and women. The per cent adequacy of  $\beta$ -carotene and riboflavin was more among women (60 and 88) than that of men (43 and 68) but it was not significant. On

the other hand, it is evident from the results that the diet consumed by the hyperlipidemic men and women was more than adequate in supplying protein, fat, thiamin, niacin and vitamin C.

In conclusion, it is evident from the results that the diet consumed by the selected hyperlipidemic men and women was more than adequate in providing maximum nutrients in needed amounts as per the recommended dietary allowances for adults specially the adequacy (%) of fat intake was highest among the nutrients.

Socio-economic factors	Hyperlipidemi	Hyperlipidemic subjects	
	Number	Per cent	
Age in years			
40-50	15	50.00	
>50-60	15	50.00	
Sex			
Male	12	40.00	
Female	18	60.00	
Type of the family			
Joint	10	33.33	
Nuclear	20	66.66	
Educational status			
Illiterate	2	06.66	
School education	7	23.33	
Higher secondary	5	16.66	
Graduate	8	26.66	
Post-graduate	8	26.66	
Monthly family income			
Rs. ≤ 10000	1	03.33	
Rs. 10000 - Rs. 25000	9	30.00	
Rs. > 25000 to ≥ 50000	20	66.66	
Occupation			
Homemaker	11	36.66	
Service	13	43.33	
Business	6	20.00	

Table 1: Socio-economic status of the selected hyperlipidemic subjects (n=30)

Table 2: Family history of various diseases among the selected hyperlipidemic subjects (n=30)

Particulars	Hyperlipidemic subje	
	Number	Per cent
Hyperlipidemia		
Inherited from father	09	30.00
Inherited from mother	11	36.66
Blood pressure	17	56.66
Heart disease	03	10.00
Acidity	13	43.33

Table 3: Information regarding health complications experienced by the selected hyperlipidemic subjects (n=30)

Particulars	Hyperlipidemic subjects	
	Number	Per cent
Blood pressure fluctuations	23	76.66
Heart surgery	03	10.00
Brain stroke	02	06.66
Breathlessness	21	70.00
Dizziness	20	66.66

Particulars	Hyperlipidemic subjects		
	Number	Per cent	
What is hyperlipidemia?	17	56.66	
Causes of hyperlipidemia	12	40.00	
Information acquired from			
News paper	18	60.00	
Television	11	36.66	
Magazine	15	50.00	
Physician	30	100.0	

 Table 4: Awareness regarding hyperlpidemia among the selected hyperlpidemic subjects (n=30)

Table 5: Prevailing exercise pattern among the selected hyperlipidemic subjects (n=30)

Particulars	Hyperlipidemic subjects		
	Number	Per cent	
Exercise pattern			
Daily	24	86.66	
Twice a week	02	06.66	
Type of exercise			
Walking	24	86.66	
Running	02	06.66	
Yoga	10	33.33	
Sports	03	10.00	

Table 6: Average dail	y intake of different for	ods by the selected h	yperlipidemic subjects

'ť Food group (g) Average amount of food intake by the selected hyperlipidemic subjects Value  $Mean \pm SD$ Mean ± SD Men (n=12) Women (n=18) Cereals  $312.5\pm78$  $259.5 \pm 51.65$  $2.07^{NS}$  $0.29\,\mathrm{NS}$ Pulses  $54.5\pm32.36$  $58.25\pm36.28$  $55\pm79.30$ Green leafy vegetables  $0.58\,\mathrm{NS}$  $40\pm61.46$ Other vegetables  $92 \pm 73.76$  $87.5 \pm 83.59$  $0.15\,^{\rm NS}$  $0.95\,{\rm ^{NS}}$ Roots and tubers  $63 \pm 37.73$  $51.5\pm22.07$  $1.98\,{}^{\rm NS}$ Milk and milk product  $153 \pm 89.32$  $243 \pm 157.91$  $0.20\,\mathrm{NS}$  $20\pm33$ Fruits  $23 \pm 45.77$ Fats and oils#  $24 \pm 2.10$ 0.29 NS  $23.75 \pm 2.57$ Sugar and jaggary  $9 \pm 2.10$  $11.25 \pm 7.58$  $1.19\,^{\rm NS}$ 

NS - Non significant # - Visible fat only

(n=30)

Nutrient	Mean value of different nutrient intake by the selected hyperlipidemic subjects		't' Value
	Mean ± SD Men (n=12)	Mean ± SD Women (n=18)	
Energy (Kcal)	$1940 \pm 229$	$1844 \pm 226$	1.12 <sup>NS</sup>
Protein (g)	$62 \pm 10.66$	$61 \pm 10.1$	0.35 <sup>NS</sup>
Fat (g)	$50.31 \pm 4.78$	$55.14 \pm 14.41$	1.31 <sup>NS</sup>
Iron (mg)	$19.29 \pm 5.89$	$17.11 \pm 7.03$	$0.91^{NS}$
calcium (mg)	$589.47 \pm 253$	$748.27 \pm 245$	1.70 <sup>NS</sup>
$\beta$ – carotene (µg)	$2057.94 \pm 1897$	$2882.2 \pm 3806$	$0.78^{NS}$
Thiamine (mg)	$1.8 \pm 0.38$	$1.6 \pm 0.3$	1.53 <sup>NS</sup>
Riboflavin (mg)	$0.95 \pm 0.22$	$0.97 \pm 0.24$	0.23 <sup>NS</sup>
Niacin (mg)	$19.3 \pm 3.61$	$16.5 \pm 3.48$	2.11 <sup>NS</sup>
vitamin C (mg)	51.56 ± 37.25	$63.48 \pm 48.94$	$0.75^{NS}$

NS - Non significant

			(11 50)
Nutrient	Adequacy (%) of nutrients in the diet of the selected hyperlipidemic men and women		'Z' Value
	Men (n=12)	Women (n=18)	
Energy (Kcal)	84	97	1.25 <sup>NS</sup>
Protein (g)	103	110	0.69 <sup>NS</sup>
Fat (g)	201	276	1.12 <sup>NS</sup>
Iron (mg)	113	81	5.58**
Calcium (mg)	98	125	2.02 <sup>NS</sup>
$\beta$ – carotene (µg)	43	60	0.94 <sup>NS</sup>
Thiamine (mg)	150	160	0.29 NS
Riboflavin (mg)	68	88	1.34 <sup>NS</sup>
Niacin (mg)	121	137	$0.75  {}^{ m NS}$
Vitamin C (mg)	129	159	1.02  NS

**Table 8:** Per cent adequacy in the intake of different nutrients per day by the selected hyperlipidemic subjects (n=30)

NS-Non significant\*\*-Significant at 1 per cent level

#### Conclusion

On the whole, results indicated that among selected hyperlipidemic subjects, maximum subject experienced blood pressure fluctuation followed by breathlessness and dizziness. More number of subjects was aware about the meaning and causes of hyperlipidemia and physician was the main source for acquiring the information.

Maximum number of hyperlipidemic subjects performed daily walking as an exercise pattern followed by yoga and sports. Consumption of pulses, green leafy vegetables, milk and milk products, fruits and sugar and jaggary was found to be more by the hyperlipidemic women than those by hyperlipidemic men. However the mean intake of different foods between men and women did not differ significantly. The diet of the selected hyperlipidemic men and women was adequate in providing maximum nutrients in needed amounts as per the recommended dietary allowances for adults specially the adequacy (%) of fat intake was highest among the nutrients. This may be one of the important causes for occurrence of hyperlipidemia among the selected subjects. Hence lifestyle and dietary pattern needs to be modified to lead a healthier life by the hyperlipidemics.

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### Nutritional Composition and Sensory Quality of Cookies Incorporated with Little Millet (*Panicum Milliarae*)

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#### Abstract

Present study was carried out to formulate and evaluate the little millet based cookies for their nutritional and sensory acceptability to increase the utilization of millet in the preparation of healthy snacks. Two types of cookies were developed *viz* sugar cookies and salt cookies by replacing one part of refined wheat flour with little millet flour by repeated trials. Proximate composition, iron and calcium content were estimated and sensory analysis was done for all the samples to find out the effect of incorporation of millet. Cookies were packed in zip lock covers and stored at room temperature and refrigerator and sensory analysis was carried out till 30 days at the intervals of 10 days. Results showed that cookies were good sources of fat, protein and iron, especially salt cookies were high in protein and iron. There was no significant difference in sensory profile of the cookies prepared by incorporating millet flour in comparison with control products. The storage temperature and duration did not alter the sensory attributes as judged by statistical analysis.

Keywords: Nutritional Composition; Sensory Analysis; Storage Studies; Low Temperature Storage.

#### Introduction

Millets, alternatively called as coarse cereals, are small grains grown in low rain fall areas (Sambavi et al., 2015), they are staple food sources not only providing major nutrients like protein, carbohydrates, fat etc, but also provide fair amounts of minor nutrients (Singh, 2016), *Ragi* (finger millet), *Navane* (Foxtail millet), *Samai* (Little millet), *Haraka* (Kodo millet), *Banti* (Barnyard millet) are commonly used millets, largely cultivated in the Asian and African countries (Jayabhaye et al., 2014). Millets are called as low glycemic foods, as they release sugars slowly into the blood stream. Hence, they are good for diabetic people and also they are poor source of gluten, hence good for people with celiac disease (Karuppaswamy et al., 2013).

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In many developing countries malnutrition and other communicable diseases are increasing because of life style and consumption of diets high in calories, fat and low in protein (Singh, 2016). On account of this, there is an increasing level of awareness regarding health benefits of millets, hence consumers are attracted more towards traditional foods and want to know more about their nutritional benefits and role in prevention of certain non-communicable diseases.

Little millet (*Panicum milliarae*) is a small seeded cereal included among the minor millet. It is a nutritious but neglected crop. Little millet has a good storage stability and is of superior nutritional value; a good source of energy, protein, B complex vitamins, fiber and minerals. It is specifically a rich source of iron with 9.30 mg/100g (Gopalan et al., 2009).

Beneficial effect of consumption of little millet among humans are indicated by many investigators. Hypoglycemic action, hypolipidemic effect and faecal bulking effects of little millet have been determined and reported. It also contains phytochemicals such as phenolic acids, flavonoids, tannins and phytates (Krishnakumari and Thayumanavan, 1997).

Snacks food market, is one of the largest market in the world, continuous innovations in the new flavors and new products are expected to score huge gains in the upcoming years. Among the snacks food market, baking industry occupies wide area and demand is increasing for bakery products at the rate of 10.07% annually, so bakery products are the best vehicle to incorporate the small millets to enhance the nutritive value and for better health (Kamaljit et al., 2010). Refined wheat flour is the major ingredient in the bakery sector, however, it has low level of important micronutrients, which are removed during the process of refining. Studies indicate that refined wheat flour has lower levels of protein, fat, minerals, vitamins and dietary fiber in comparison to whole wheat flour (Oghbaei and Prakash, 2013, Oghbaei and Prakash, 2016). Comparatively refined wheat flour also contains less nutrients than millets (Gopalan et al., 2009).

Cookies are a group of foods which has long shelf life, generally accepted by all age groups and are available almost everywhere (Turner et al., 2010, Popov-Raljic et al., 2013). The main ingredients in cookies preparation are refined wheat flour, fat and sugar (Paret and Delcour, 2008). The objective of present study was to prepare cookies by partial replacement of wheat flour with little millet and analyze its nutritional and sensory quality.

#### Materials and Methods

Dehusked little millet (*Panicum milliarae*) was obtained from the local organic shop. Low trans fat margarine was procured from an organic source and all the other ingredients used for the formulation of products such as refined wheat flour, icing sugar, and other spices were brought from local market. All the chemicals and solvents used for the study were of analytical grade and were procured from E-Merck, Mumbai, SD Fine, Qualingens Fine Chemicals, Mumbai, and Nice chemical laboratories (Kolkata). Glass double distilled water was used for all the analysis.

#### Method

Dehusked little millet (*Panicum milliarae*) was cleaned and extraneous materials were removed. This cleaned millet was milled into fine flour in milling machinery and whole millet flour was used in the study to retain the fiber content.

Two types of cookies were prepared *viz.*, sugar cookies and salt cookies.

#### Sugar Cookies

Refined wheat flour (40 g) was sifted along with

baking powder (1/2 tsp.) and kept aside. Margarine (25 g) and icing sugar (35 g) was creamed together and folded well with flour and cardamom powder (2 no.). Cookies were rolled and baked in a preheated oven at 200°C for 10 minutes. In the experimental product, 20% of refined wheat flour was replaced with little millet flour.

#### Salt Cookies

Refined wheat flour (80 g), margarine (25 g) and other herbs and spices (green chili - 2 no., omum seeds -1/2 tsp, curry leaves - 5 g, coriander leaves - 6 g and salt (to taste) were mixed together. Curd (25 ml) was added to this mixture and kneaded well to make stiff dough. The dough was rolled and cookies were baked in preheated oven at 170°C for about 15 - 20 minutes. In experimental cookies, 40% of refined wheat flour was replaced with little millet flour.

#### Analysis of Nutrients

Cookies were analyzed for nutritional composition by standard techniques. Moisture was estimated by oven drying and repeated weighing of samples till a constant dry weight was obtained and moisture calculated by difference in weight (AOAC, 2005). Fat was extracted by Soxhlet distillation method using suitable organic solvent, and extracted fat was weighed after removing the solvent (Raghuramulu et al., 2003). Protein was determined by the Kjeldahl procedure which measures the nitrogen content of a sample. The digested sample was made alkaline with NaOH and the nitrogen is distilled off as ammonia. This is "trapped" in a boric acid solution and quantified by titration with a standard HCl solution. A conversion factor of 6.25 was used to convert the percent nitrogen to crude protein (AOAC, 2000). Total ash was estimated by incinerating the food sample in a muffle furnace at 600°C for 3 - 5 hours. It was weighed and the total ash obtained was converted to solution for measuring iron and calcium (Raghuramulu et al., 2003). Iron was estimated colorimetrically by method of Wong using potassium thiocyanate for color development. A standard curve was prepared by using ferrous ammonium sulphate. The iron content of sample was then read from the standard curve (Raghuramulu et al., 2003). Calcium was precipitated as calcium oxalate, the precipitate dissolved in hot dilute H<sub>2</sub>SO<sub>4</sub> and titrated against standard potassium permanganate (Oser, 1965).

#### Sensory Analysis

Cookies were randomly coded and subjected to

sensory analysis for appearance, color, texture, aroma, taste and overall quality by using the score card having 1-10 grading scale by semi-trained panel members (n = 30). The products were evaluated by the panel members in mid-morning and water was allowed in between the samples. Effect of incorporation of millet on sensory attribute was analyzed and the results were tabulated. To determine the alteration in the sensory quality during the storage, all samples were packed in the Zip lock cover, one set of samples were stored in the refrigerator (low temperature) and another set of samples were stored at room temperature. Sensory evaluation was done for the samples stored at both the temperature till the end of 30th day at the intervals of 10 day and results were recorded.

#### Statistical Analysis

The data were analyzed statistically using suitable tests. Standard deviation and t-test was used for the compositional analysis data and also post test carried out to see any significant differences between the samples. The sensory analysis data was subjected to t-test to determine the statistically significant differences among the products which were developed.

Table 1: Nutritional composition of cookies (per 100g)

#### **Results and Discussion**

#### Analysis of Nutritional Composition

Nutritional composition of cookies was analyzed and the results are presented in Table 1. Moisture content of the refined wheat flour (RWF) cookies and litte millet (LM) sugar cookies was found to be 3.17±0.08 and 3.64±0.02 respectively as there was no added water in the cookies during preparation, the moisture content was not high.

But in salt cookies curd was used to prepare the dough, hence, moisture content was in higher range (12.29±0.32 in RWF and 13.08±0.07 LM) as curd has 89% moisture (Gopalan et al., 2009). Fat content of sugar cookies was high in both RWF and LM flour cookies because of added fat, i.e margarine, and slightly lower fat content was found in salt cookies as lesser fat was used than sugar cookies. Addition of millet did not change the protein content significantly in salt and sugar cookies had more protein than sugar cookies. Kumar et al., (2015) and Hemalatha et al., (2006) reported that addition of millet flour resulted in an increase in protein and fiber content in millet incorporated biscuits.

Nutrients	<b>RWF Sugar Cookies</b>	LM Sugar Cookies	<b>RWF Salted Cookies</b>	LM Salted Cookies
Moisture (g)	3.17±0.08	3.64±0.02	12.29±0.32	13.08±0.07
Fat (g)	25.31±0.12	25.68±0.10	18.84±0.02	18.36±0.00
Protein (g)	5.40±0.11	5.30±0.00	9.96±0.21	10.06±0.11
Ash (g)	0.92±0.02	1.13±0.01	3.10±0.00	4.20±0.03
Calcium (mg)	15.43±0.03	33.89±1.56	54.69±0.78	70.10±0.77
Iron (mg)	2.07±0.06	2.28±0.12	3.95±0.46	6.65±0.30

RWF- Refined wheat flour, LM- Little millet.

Ash content of LM cookies was higher than RWF cookies as little millet is richer in mineral content. Increased iron content was seen in both sugar (2.28 mg/100g) and salt cookies (6.65 mg/100g). LM salt cookies has highest iron content due to the addition of greens, as green leafy vegetables are rich in iron (Gopalan et al., 2009).

Millet incorporated cookies has higher calcium content than RWF cookies and more calcium content was found in LM salt cookies. This enhancement could be due to addition of curd, which has a higher calcium content (Gopalan et al., 2009). Hemalatha et al., (2006) also reported a content of 3.30 mg of iron and 23 mg of calcium/100g in the 20% little millet incorporated cookies. Shiny et al., (2012) also reported an improved nutrient profile in the millet incorporated biscuits.

# Effect of Incorporation of Millet on Sensory Attributes of Cookies

Sensory characteristics of cookies are presented in the Table 2. Scores for all sensory attributes of cookies ranges from 9.0-9.1 $\pm$  0.8. Sensory evaluation of cookies for appearance, color, texture and flavor shows no significant difference between the RWF cookies and LM cookies. It ranged between 9.0-9.1 $\pm$ 0.7. Shiney and John (2012) also found that millet added biscuits were well accepted by panel members.

Sensory attributes	Sugar Cookies		Salt Cooki	ies
-	Refined wheat flour	Little millet	Refined wheat flour	Little millet
Appearance	$9.0 \pm 0.6$	$9.1 \pm 0.7$	9.1 ±0.7	9.3 ±0.7
p-value	-	0.350 ns	-	0.845 ns
Color	9.1±0.6	9.2±0.6	9.2 ±0.7	$9.2 \pm 0.7$
p-value	-	0.571 ns	-	0.550 ns
Texture	9.1 ±0.7	9.1 ±0.7	9.1 ±0.8	9.3 ±0.6
p-value	-	0.461 ns	-	0.855 ns
Flavor	9.1 ±0.8	9.1 ±0.7	9.1 ±0.7	9.3 ±0.7
p-value	-	0.377 ns	-	1.000 ns
Overall quality	9.1 ±0.7	$9.2 \pm 0.7$	9.1 ±0.7	9.1 ±0.7
p-value	-	0.566 ns	-	1.000 ns

Table 2: Effect of incorporation of millet on sensory attributes (Mean ± Standard deviation) of Cookies

P value indicates statistical significance between refined wheat flour and millet Cookies on application of Student's 'T' test., ns- not significant

#### Effect of Storage on Sensory Quality of Cookies

Effect of storage on sensory value of sugar cookies and salt cookies are presented in the Table 3 and Table 4. Comparison was done between the duration of storage with 0 day samples using Students T test and the P value are indicated in tables. It was also done between the samples stored at room and low temperature and appropriate statistical notations are indicated along with the attribute scores in tables. On the 10<sup>th</sup> day of storage period, products stored at the refrigerator and room temperature were given almost similar scores for all the attributes and no significant difference was observed in any of the attributes.

 Table 3: Effect of storage on sensory quality of Sugar cookies: comparison between samples stored at different temperature and with 0 day sample
 (P value)

Sensory attributes	Refined Whea	t Flour cookies	Little M	illet cookies	
5	R.T.	R.F.	R.T.	R.F.	
10th day					
Appearance	$9.0 \pm 0.8$	9.0 ±0.8 <sup>ns</sup>	$9.0 \pm 0.6$	9.0 ±0.5 n	
p-value	0.711 <sup>ns</sup>	0.729 <sup>ns</sup>	0.854 ns	1.000 ns	
Color	9.1±0.8	9.1 ±0.7 ns	9.1±0.7	9.2 ±0.8 n	
p-value	0.855 ns	0.698 ns	0.695 ns	1.000 ns	
Texture	9.1 ±0.7	9.1 ±0.8 ns	9.1 ±0.7	9.1 ±0.7 ns	
p-value	1.000 ns	0.863 ns	1.000 ns	0.593 ns	
Flavor	9.1 ±0.8	9.1 ±0.8 <sup>ns</sup>	9.2 ±0.7	9.1 ±0.8 n	
p-value	0.867 <sup>ns</sup>	0.867 <sup>ns</sup>	0.576 ns	1.000 ns	
Overall quality	9.1 ±0.7	9.2 ±0.5 <sup>ns</sup>	9.2 ±0.7	9.2 ±0.7 n	
p-value	0.854 <sup>ns</sup>	1.000 ns	0.597 <sup>ns</sup>	0.730 <sup>ns</sup>	
20th day					
Appearance	$9.0 \pm 0.8$	9.0 ±0.7 <sup>ns</sup>	$9.0 \pm 0.7$	9.0 ±0.6 n	
p-value	0.678 ns	0.583 ns	0.711 ns	0.436 ns	
Color	9.1±0.8	9.1 ±0.7 <sup>ns</sup>	9.1±0.7	9.2 ±0.8 n	
p-value	0.855 ns	0.701 ns	0.695 ns	1.000 ns	
Texture	9.1 ±0.8	9.1 ±0.7 <sup>ns</sup>	9.0 ±0.7	9.2 ±0.6 n	
p-value	1.000 ns	1.000 ns	1.000 ns	1.000 ns	
Flavor	9.1 ±0.7	9.2 ±0.7 <sup>ns</sup>	9.1 ±0.7	9.1 ±0.7 n	
p-value	0.867 ns	0.728 <sup>ns</sup>	0.576 ns	0.848 ns	
Overall quality	9.1 ±0.7	9.2 ±0.7 <sup>ns</sup>	9.2 ±0.5	9.2 ±0.7 n	
p-value	1.000 ns	0.520 ns	0.597 ns	0.713 ns	
30th day					
Appearance	$9.1 \pm 0.7$	9.1 ±0.6 <sup>ns</sup>	$9.0 \pm 0.7$	9.0 ±0.7 n	
p-value	0.243 ns	0.673 ns	0.056 ns	0.169 ns	
Color	9.1±0.7	9.1 ±0.7 <sup>ns</sup>	9.0±0.7	9.2 ±0.8 n	
p-value	0.333 ns	1.000 ns	0.706 ns	0.469 ns	
Texture	9.1 ±0.8	9.1 ±0.7 <sup>ns</sup>	9.1 ±0.7	9.1 ±0.7 n	
p-value	0.854 ns	0.855 <sup>ns</sup>	0.860 ns	0.593 ns	
Flavor	9.1 ±0.8	9.1 ±0.6 <sup>ns</sup>	9.1 ±0.7	9.1 ±0.7 n	
p-value	0.736 ns	0.725 <sup>ns</sup>	0.844 ns	1.000 ns	
Overall quality	$9.0 \pm 0.7$	9.2 ±0.8 ns	$8.0 \pm 0.7$	9.0 ±0.7 n	
p-value	0.698 ns	0.132 <sup>ns</sup>	0.730 ns	0.371 ns	

R.T.- Room temperature, R.F.- Refrigerator, P value indicates statistical significance on application of Student's 'T' test between 0 day and stored samples.

Notations with attributes indicate difference between sample stored at different temperatures, ns: not significant.

 Table 4: Effect of storage on sensory quality of Salt cookies: comparison between samples stored at different temperature and with 0 day sample
 (P value)

			Little Millet cookies		
Sensory attributes	Refined Wheat Flour cookies R.T. R.F.		R.T.	R.F.	
10th day					
Appearance	$9.0 \pm 0.6$	9.3 ±0.6 <sup>ns</sup>	$9.0 \pm 0.7$	9.2 ±0.7 <sup>ns</sup>	
p-value	0.560 ns	0.442 ns	0.687 ns	0.709 ns	
Colour	9.1±0.6	9. 2±0 <sup>ns</sup>	9.2±0.7	9.2 ±0.6 <sup>ns</sup>	
p-value	0.698 <sup>ns</sup>	1.000 <sup>ns</sup>	0.709 <sup>ns</sup>	0.698 ns	
Texture	9.1 ±0.7	9.1 ±0.7 ns	9.0 ±0.7	9.1 ±0.7 ns	
p-value	0.710 <sup>ns</sup>	1.000 ns	0.711 ns	1.000 ns	
Flavour	9.1 ±0.8	9.0 ±0.7 <sup>ns</sup>	9.2 ±0.8	$9.3 \pm 0.7$ ns	
p-value	1.000 ns	0.377 ns	0.714 ns	0.205 ns	
Overall quality	9.1 ±0.7	9.2 ±0.7 <sup>ns</sup>	9.2 ±0.7	9.2 ±0.7 <sup>ns</sup>	
p-value	1.000 ns	0.852 <sup>ns</sup>	0.571 <sup>ns</sup>	0.698 <sup>ns</sup>	
20th day					
Appearance	$9.1 \pm 0.7$	9.1±0.6 ns	$9.1 \pm 0.8$	9.1 ±0.7 <sup>ns</sup>	
p-value	1.000 ns	0.727 ns	0.426 ns	0.461 ns	
Colour	$9.1 \pm 0.7$	9.2 ±0.7 <sup>ns</sup>	$9.2 \pm 0.8$	9.2 ±0.6 <sup>ns</sup>	
p-value	1.000 ns	1.000 ns	1.000 ns	0.727 ns	
Texture	9.1 ±0.8	9.0 ±0.7 <sup>ns</sup>	9.1 ±0.7	9.1 ±0.7 <sup>ns</sup>	
p-value	0.860 <sup>ns</sup>	0.867 ns	0.711 ns	0.593 ns	
Flavour	9.1 ±0.7	9.2 ±0.7 <sup>ns</sup>	9.1 ±0.8	9.2 ±0.7 <sup>ns</sup>	
p-value	0.867 <sup>ns</sup>	0.867 ns	0.714 <sup>ns</sup>	0.704 ns	
Overall quality	9.3±0.7	9.2 ±0.7 <sup>ns</sup>	9.1 ±0.8	9.2 ±0.7 <sup>ns</sup>	
p-value	0.470 ns	1.000 ns	1.000 ns	1.000 ns	
30th day					
Appearance	$9.0 \pm 0.7$	9.1 ±0.7 <sup>ns</sup>	$8.9 \pm 0.7$	9.1 ±0.8 <sup>ns</sup>	
p-value	0.720 <sup>ns</sup>	0.281 ns	0.461 ns	0.490 ns	
Colour	8.9±0.8	9.0 ±0.8 <sup>ns</sup>	9.0±0.8	9.1 ±0.7 <sup>ns</sup>	
p-value	1.000 ns	1.000 ns	0.252 ns	0.461 ns	
Texture	9.1 ±0.8	9.0 ±0.8 <sup>ns</sup>	9.1 ±0.8	$9.0 \pm 0.8$ ns	
p-value	0.860 ns	0.311 ns	0.868 ns	0.609 ns	
Flavour	9.2 ±0.7	9.3 ±0.8 <sup>ns</sup>	9.1 ±0.7	9.1 ±0.7 <sup>ns</sup>	
p-value	0.662 ns	0.205 ns	1.000 <sup>ns</sup>	0.604 ns	
Overall quality	8.9±0.7	9.0 ±0.8 <sup>ns</sup>	9.2 ±0.7	9.1 ±0.7 <sup>ns</sup>	
p-value	0.264 ns	0.556 ns	0.367 ns	0.255 ns	

samples stored at different temperature and with 0 day sample (P value)

R.T.- Room temperature, R.F.- Refrigerator, P value indicates statistical significance on application of Student's 'T' test between 0 day and stored samples. Notations with attributes indicate difference between sample stored at different temperatures, ns: not significant.

On the 20<sup>th</sup> and 30<sup>th</sup> day of storage period, sensory scores slightly decreased from 9.3 to 8.0 for the products stored at room temperature in comparison with samples stored under refrigeration, however, the differences were not significant. Little millet contains phenolic compounds, which function as powerful antioxidants and increase the shelf life cookies by preventing them from rancidity (Krishnakumari and Thayumanavan, 1997).

#### Conclusion

Cookies prepared by incorporating little millet flour exhibited better nutritional composition. Addition of green leafy vegetables and curds to the salt cookies was highly beneficial in the enhancement of iron and calcium content. The sensory quality of cookies was also good, with no significant differences found in the acceptability of millet incorporated and standard cookies. There was no off flavor or off odor developed during the storage period at both the storage conditions. Hence in conclusion, it can be stated that addition of little millet in the preparation of cookies is a healthy option to utilize millet and provide essential nutrients without any alteration in the taste profile of the products with good shelf life.

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### Adverse Pregnancy Outcomes in Women with Polycystic Ovary Syndrome

#### Mohini Paliwal\*, Vandana Bharti\*\*, Kirti Tewari\*\*\*

#### Abstract

*Objective:* The aim of this study was to know the adverse pregnancy outcomes and find out its association with polycystic ovary syndrome. *Material and Methods:* A case-control study was conducted at fertility center in Indore city from June 2014 to September 2015. The study population included seventy five women diagnosed with PCOS and 75 women without PCOS followed from early pregnancy. Incidence of gestational diabetes mellitus (GDM), pregnancy induced hypertension (PIH), preeclampsia, pre term delivery and caesarean section were determined. Statistics was done using statistical package for the social science (SPSS) 21.0. *Results:* The prevalence of miscarriage rate (all spontaneous losses <20 wks) was notable higher among PCOS group. The prevalence of GDM was significantly higher in women with PCOS than that of women without PCOS (31.82% vs. 15.07%; p<0.05). The prevalence of preeclampsia was more among women with PCOS as compared to women without PCOS, but not significant (16.67% vs. 8.22%; p>0.05). The overall risk of pre term delivery was 27.27% in women with PCOS compared to 9.59% in women without PCOS, P<0.01. *Conclusion:* The incidence of GDM, PIH and pre term delivery were significantly higher in women with PCOS as compared to women without PCOS.

**Keywords:** Adverse Pregnancy Outcomes; Gestational Diabetes Mellitus; Polycystic Ovary Syndrome; Pregnancy Induced Hypertension.

#### Introduction

Polycystic ovary syndrome (PCOS) is one of the most common endocrine disorder in reproductive age women and its prevalence varies broadly from 6% to 15% in general population depending on ethnicity studied and criteria utilized (Kumarapeli et al. 2008; Mehrabian et al. 2011). It is characterized by the presence of oligo or amenorrhea, polycystic ovary, hirsutism, raised LH: FSH ratio, insulin resistance (IR) and compensatory hyperinsulinemia (The Rotterdam ESHRE/ASRM – sponsored consensus

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workshop group, 2004). IR has an important influence on the development of diabetes type 2 and hypertension (Perciaccante et al. 2006) . Abdominal overweight and obesity also important components of PCOS that affect 30%-70% of the PCOS population (Pasquali et al. 2006; Vrbikova et al. 2009). In women with PCOS, pregnancy is often complicated by gestational diabetes mellitus (GDM), pregnancy induced hypertension (PIH), preeclampsia and also the risk for a preterm delivery or a delivery by cesarean section is raised (Boomsma et al. 2006; Altieri et al. 2010; Kjerulff et al. 2011; Qin et al. 2013). The new born babies stay more frequently in a neonatal intensive care unit and perinatal mortality also occurs more frequently (Wang et al. 2015).

However, not only PCOS, but also obesity, which is frequently accompanied by PCOS, is an independent risk factor of these obstetric complications (Frene et al. 2015). A few studies have reported an increased risk of adverse pregnancy and neonatal outcomes in women with PCOS. The purpose of the study was to determine the adverse pregnancy outcome in women with PCOS.

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#### Methods

A case-control study was conducted at fertility center in Indore city from June 2014 to September 2015. The study population included seventy five women diagnosed with PCOS and 75 women without PCOS followed from early pregnancy. PCOS was diagnosed using the diagnostic criteria of the Rotterdam consensus 2003. All respondents were interviewed personally to obtain the relevant information. All parameters and pregnancy outcome were retrieved from medical records.

The diagnosis of gestational diabetes mellitus (GDM) was defined as fasting blood glucose level  $\geq$  95mg/dl and oral glucose tolerance test (75 gm glucose orally administered and plasma glucose measured after 2 hours)  $\geq$  155 mg/dl. Pregnancy induced hypertension (PIH) was defined as blood pressure reading  $\geq$  140/90 mmHg. Preeclampsia is defined as blood pressure reading  $\geq$  140/90 mmHg in combination with proteinuria more than 300mg/ 24 hrs after 20weeks of gestation. Preterm birth defined as delivery at less than 37weeks of gestation.

Statistics was done using statistical package for the social science (SPSS) 21.0. All the values were expressed as percentage and mean  $\pm$  standard deviation (SD), continuous variables were compared using the Z - test and categorical data using the chisquare test. P values of <0.05 were considered statistically significant.

#### Results

The demographic characteristics of the women with PCOS and without PCOS are summarized in Table 1. Women with PCOS were slightly older than women without PCOS (mean age  $31.23 \pm 4.40$  yrs Vs 29.63  $\pm$  4.09 yrs, p<0.01). The age at menarche was found slightly more among women with PCOS as compare to without PCOS ( $13.93 \pm 1.65$  yrs Vs  $13.37 \pm 1.34$  yrs, p<0.01). BMI prior to pregnancy was statistically significant between women with and without PCOS ( $26.08 \pm 4.91$  kg/m<sup>2</sup> Vs  $23.36 \pm 3.26$  kg/m<sup>2</sup>, p<0.01). The length of gestation was statistically significant between both groups ( $34.17 \pm 5.59$  wks Vs  $36.40 \pm 3.36$  wks, p<0.01).

Table 1: Demographic characteristics of the study population

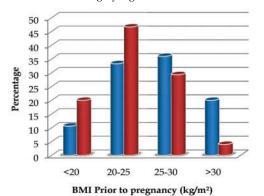
Characteristics	Women with PCOS (n=75) Mean ± SD	Women without PCOS (n=75) Mean ± SD	Z Value	P Value
Age (yrs)	31.23±4.40	29.63±4.09	2.305	< 0.05*
Age at Menarche (yrs)	$13.93 \pm 1.65$	$13.37 \pm 1.34$	2.281	< 0.05*
Pre pregnancy BMI (kg/m <sup>2</sup> )	$26.08 \pm 4.91$	$23.36 \pm 3.26$	3.997	< 0.01**
Length of gestation (weeks)	$34.17 \pm 5.59$	$36.40 \pm 3.36$	2.971	< 0.01**

\* Significant at 5% level; \*\* highly significant at 1% level; [BMI=Body mass index]

Table 2: Adverse pregnancy outcomes in women with and without PCOS

Adverse Pregnancy outcomes	Women with PCOS frequency (%)	Women without PCOS frequency (%)	Chi square value	P Value	
Miscarriage (<20 wks)	9/75 (12.00%)	2/75 (2.67%)	_	_	
GDM	23/66 (34.85%)	8/73 (10.95%)	11.414	< 0.01**	
PIH	21/66 (31.82%)	11/73 (15.07%)	5.478	< 0.05*	
Pre eclampsia	11/66 (16.67%)	6/73 (8.22%)	2.305	>0.05	
Pre term Delivery	18/66 (27.27%)	7/73 (9.59%)	7.35	< 0.01**	
Cesarean Section	52/66 (78.79%)	48/73 (65.75%)	2.921	>0.05	

\* Significant at 5% level; \*\* highly significant at 1% level



Women with PCOS
 Women without PCOS
 Women without PCOS

**Fig. 1:** Prevalence percentage of women with PCOS and without PCOS according to pre pregnancy BMI

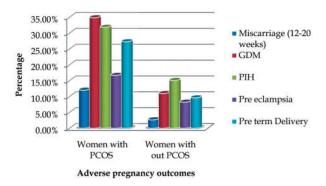


Fig. 2: Prevalence of adverse pregnancy outcomes in women with and without PCOS

Table 2 reveals the prevalence of adverse pregnancy outcomes and their association with PCOS in the study population. The miscarriage rate (all spontaneous losses <20 wks) in women with PCOS was 12.00% compared to 2.67% in women without PCOS. The percentage of women with ongoing pregnancy beyond 20 weeks who developed GDM was statistically significant in women with PCOS as compared to women without PCOS (34.85% vs. 10.95%, p<0.01). The prevalence of PIH was two times higher in women with PCOS than that of women without PCOS (31.82% vs. 15.07%; p<0.05). The prevalence of preeclampsia was more among women with PCOS as compared to women without PCOS (16.67% vs. 8.22%; p>0.05). The overall risk of pre term delivery was 27.27% in women with PCOS compared to 9.59% in women without PCOS, P<0.01. The prevalence of cesarean section was higher in both groups.

#### Discussion

The results of our study support the hypothesis that women with PCOS to have a higher risk of adverse pregnancy outcomes as compared to women without PCOS. We observed in our study that women with PCOS have an increased prevalence with increasing age of pregnancy. Similarly in previous study conducted by (Roos et al. 2011; Tehrani et al. 2011) also found that women with PCOS were slightly older than women without PCOS. Strong positive association was observed between pre pregnancy BMI and PCOS. PCOS was also associated with metabolic syndrome. This finding was consistent with study on pregnant women with PCOS (Baldani et al. 2012; Frene et al. 2014). The women with PCOS were observed to be at higher risk for developing GDM. These finding suggested that PCOS may be a predisposing factor of GDM (Paola et al. 2010; Roos et al. 2011; Wang et al. 2013). It is important to note that, among the obese population, the effects of PCOS on the presence of GDM were mitigated, while the incidence of GDM among the women with PCOS did not differ significantly from the women with out PCOS. These results suggested that obesity may also play a role in the development of GDM (Turhan et al. 2003). Pregnant women with PCOS have a greater risk of PIH compared to women without PCOS in the present study, which is agreement with other studies (Altieri et al. 2010; Wang et al. 2013). We did not find any association between PCOS and preeclampsia. Similarly the prevalence of preeclampsia appears to be two-fold greater in women with PCOS, but was not statistically significant (Bagegni et al. 2010). We found that incidence of pre term was significantly high in women with PCOS. A similar but not statistically significant result was found by Altieri et al. 2010. Sterling et al. 2015 reported that women with PCOS often required assisted reproductive technology to become pregnant, increasing the risk of multiple births and hypertensive disease, which are associated with pre term delivery. There are certain limitations must be noted. The sample size was small and confounding variables (age and BMI) that were not controlled. Longitudinal studies are required in future to establish the facts in this direction.

#### Conclusion

Our results suggest that women with PCOS were more likely to develop GDM, PIH and pre term delivery as compared to women without PCOS. We therefore suggest increased attention during antenatal care and delivery in women with PCOS.

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## Polyphenols in Polycystic Ovary Syndrome: A Perspective

#### E. Lakshmi\*, Abirami P.\*\*

#### Abstract

Polycystic ovary syndrome is one of the most common hormonal endocrine disorders that affects around 5-10 percent women of childbearing age and is characterized by abnormal hormone levels. Natural polyphenolic compounds found in plants known as flavonoids (catechins) and non-flavonoids (resveratrol, chlorogenic acids) have recently become very popular. This review article is an attempt to present the findings of scientific studies with regard to beneficial effects of polyphenols on humans affected by pcos. Relevant Papers were identified from Science Direct, Google Scholar and pubmed by using all combinations of the search terms related to polyphenol and pcos.

Keywords: Polycystic Ovary Syndrome; Polyphenol; Resveratrol; Catechins; Chlorogenic Acids.

#### Introduction

The female body is equipped with the power to create new life out of just a few microscopic cells. But along with that comes many physical burdens. Polycystic ovary syndrome (PCOS) is the most common hormonal condition in women across the globe.The three most common features of PCOS include irregular periods, excess androgen in the body, and polycystic ovaries that become enlarged and contain many fluid sacs.Women with pcos produce marginally higher amounts of testosterone and other male hormones than average[1]. There is evidence suggesting that elevation of these hormones contribute to infertility, weight gain, acne or excess body hair, associated [2] with other health issues as illustrated in Table1.

Table 1: Signs and Symptoms

- Hyperandrogenism
- Increased sympathetic nerve activity
- Altered GH/IGF-1 axis

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- Hypersecretion of LH
- Anovulation
- Visceral obesity
- Hypertension
- Insulin resistance
- Psychological stress
- LH, Luteinising hormone;
- GH, growth hormone;
- IGF, insulin growth factor

Raja-Khan N et al [3] addressed other risk factors to aggravate this condition: insulin resistance, obesity, hypertension, dyslipidemia, inflammation and subclinical cardiovascular disease. Anxiety, depression and reduced quality of life are also common. Pcos is characterized by lipidic blood profile, bigger adipocytes and insulin resistance. Studies performed on monkeys, sheep and rats demonstrate that prenatal androgenization is a cause of pcos [4]. In human, only one study has verified the presence of testosterone in the blood of babies from PCOS mothers [5].

Evidence suggests that vitamin D deficiency may be a causal factor in the pathogenesis of IR and the metabolic syndrome in pcos [6].

Research suggest [7] that the theca interstitial cells of the ovary are responsible for producing male hormones which are normally required for healthy ovarian function. However in pcos, the large amount of these cells causes a large amount of male hormones to be produced, causing problems with normal ovulation. This often results in delay in ovulation (and therefore in menstruation) or lack of ovulatory cycling altogether in more severe cases. These hormonal changes also greatly reduce egg quality and health. The two causes for the excessive overgrowth of these theca interstitial cells of the ovary are:

- 1. High levels of oxidative stress and
- 2. High levels of insulin in the local tissues.

Both these factors play a great role in the pathology of polycystic ovarian syndrome. Because pcos is a multifactorial pathology several treatments (Table 2) have been proposed [8].

Table 2:	Therapeutic	tools in	the	treatment of Pcos	
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1.	<b>Old Therapy</b> Oral contraceptives (OC) Anti-androgens
2.	<b>New Therapy</b> Insulin sensitizers: (Metformin Thiazolinediones)
3.	<b>Emerging Therapy</b> Statins Acupuncture Dietary products and nutrients. Vitamin D Herbal medicines Vitamin B12 and folate. Resveratrol AGEs low diet (atherogenic and endogenous) Phisical activity

#### Polyphenols in Pcos

Natural polyphenolic compounds found in plants known as flavonoids (catechins) and non-flavonoids (resveratrol, chlorogenic acids) have recently become very popular. There is evidence[9] suggesting that resveratrol, a natural plant polyphenol found in grapes, berries, and medicinal plants, which is sought after by nutritionists and biochemists for its potential anti-cancer, anti-oxidant and anti-inflammatory property , is now proved to lower levels of testosterone in pcos women. It balances lipid profile, decreases adiposity and improves insulin sensitivity. Tomatis. V et al [10] opines that Green tea (catechins, GTC) and coffee polyphenols (chlorogenic acids, CGA) have been associated with reduced diabetes and cardiovascular risk in Women with Polycystic Ovary Syndrome. Quercetin, derived from Chinese medicinal herbs such as hawthorn, has proved practical in the management of insulin resistance (IR )in women with polycystic ovary syndrome [11].

Resveratrol and other polyphenols (flavanols, flavonoids, anthocyanins and phytochemicals) are produced as part of the plant's defense system against infection [12]. The compound is found throughout the plant: in the vines, roots, seeds and stalks, but the highest concentration of resveratrol is in the grape skin. Some other rich plant sources of resveratrol include eucalyptus, spruce and lily, peanuts and berries of all forms (including blueberries, huckleberries and cranberries[13]. Content of resveratrol in grapes varies from 0.16 to 3.54  $\mu$ g/g; dry grape skin contains about  $24 \mu g/g$  of resveratrol. Resveratrol is also present in other berries and nuts. For example, cranberry raw juice contains about 0.2 mg/L. In other natural foods, the concentration of resveratrol varies in the range of  $\mu g/g$  (peanuts, pistachios) to ng/g (bilberries, blueberries). It has been documented that red wine contains a much greater amount of polyphenolic compounds than white wine. The concentration of resveratrol ranges from 0.1 to 14.3 mg/L in various types of red wine, while white wines contain only about 0.1-2.1 mg/L of resveratrol [14].

Dr. Bill Rawls a gynecologist practicing conventional medicine for over 20 years suggest Japanese knotweed as an excellent source of resveratrol, Once planted, the rhizome (root) expands rapidly, pushing out any other plants in the vicinity.

#### Bioavailability

Low solubility of resveratrol in water (<0.05 mg/mL), caused by its chemical structure, affects its absorption . In animals and humans, resveratrol is quickly metabolized in liver; in plasma it binds to lipoproteins and albumin, and this facilitates its entry to cells [15]. Resveratrol (Figure 1) presents itself in both *trans*- and *cis*- isomeric forms [16].

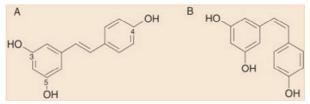


Fig. 1: Chemical structure of trans- (A) and cis-resveratrol (B).

The absorption and metabolism of resveratrol appears to be broadly similar to that of other polyphenols such as quercetin and catechin, although a number of factors may influence the pharmaco-kinetics of each.

#### Oxidative Stress and Inflammation

Resveratrol has been reported by Ketan RP [17] to decrease oxidative stress and attenuate inflammation, and these mechanisms may account for its health benefits in pcos. Oxidative stress occurs when an excess of reactive oxygen species (ROS) are generated from any of a variety of sources, including the mitochondrial electron transport chain and reduced nicotinamide adenine dinucleotide phosphate (NAD(P)H) oxidases. ROS can damage macromolecules and activate signaling pathways that include a number of inflammatory mediators . Inflammation, in turn, can lead to further oxidative stress in a cycle that contributes to the progression of many diseases. Current evidence of actions of resveratrol on the ovary in rat models by Wong et al [18] sugests that.

- Invitro- resveratrol inhibits proliferation and androgen production by theca-interstitial cells. Resveratrol also exerts a cytostatic, but not cytotoxic, effect on granulosa cells, while decreasing aromatization and vascular endothelial growth factor expression.
- In vivo- resveratrol treatment reduces the size of adipocytes and improves estrus cyclicity in the previously acyclic rat model of polycystic ovary syndrome (PCOS).

#### PCOS- Common Endocrine Disorder of Reproductive-Age Women

Some interesting findings of resveratrol on women with PCOS were reported in the journal of clinical endocrinology and metabolism from the study conducted by Beata Bet al[19] at the Poznan University of Medical Sciences in Poznan, Poland. The first randomized, double-blind, placebo-controlled clinical trial recruited a total of 30 women with PCOS randomly to two groups. They were treated daily with either 1,500 milligrams of resveratrol or placebo for 3 months to determine levels of testosterone and other androgen hormones. The study cited that resveratrol significantly reduced ovarian and adrenal androgen. The polyphenol called a phytoalexin was found to lower the levels of testosterone and dehydroepiandrosterone sulfate (DHEAS), another hormone that the body can convert into testosterone in pcos patients. The study also stated the effect to be, at least in part, related to an improvement of insulin sensitivity and a decline of insulin level. Finaly the study concluded that resveratrol (1,500 mg/day) significantly reduces total serum testosterone and DHEAS in women with polycystic ovary syndrome.

A study published in Science Daily - 2016 [20] has given a new insight into why some women have difficulty falling pregnant.Led by the research team Dr Simon Lane a research fellow at the University of Southampton involved taking immature mouse eggs and incubating them in follicular fluid taken from women who have endometriosis, in vitro. The researchers examined the amounts of ROS (Reactive Oxygen Species) that were generated and the ability of the egg to mature. They found the follicular fluid from women with endometriosis resulted in higher amounts of ROS and the ability of the egg to mature was blocked by endometriosis, Resveratrol when added to the follicular fluid taken from women who have endometriosis found ROS levels decreased and more eggs were able to mature.

In a 16-week, single-blind, unilateral crossover trial, by Tomatis V et al [21] on effects of green tea (catechins, GTC) and coffee polyphenols (chlorogenic acids, CGA) that recruited 12 PCOS women (mean  $\pm$  SD: age, 28  $\pm$  10 y; BMI, 35  $\pm$  7 kg/m<sup>2</sup>; fasting insulin, 97  $\pm$  52 pmol/L) who underwent 8 weeks of placebo treatment followed by 8 weeks of active treatment (tablets providing 2093 mg GTC and 220 mg CGA/ day) found a reduced waist circumference (MD:2.4cm; P = 0.02), altered eicosanoid profile. It is postulated that a combined intake of GTC and CGA may modulate eicosanoid pathways, thereby reducing inflammation and abdominal adiposity.

Insulin resistance (IR) is a clinical feature of polycystic ovary syndrome (PCOS). Wang Z et al [22] demonstrated that quercetin, derived from Chinese medicinal herbs such as hawthorn, decreases insulin resistance in a polycystic ovary syndrome rat model by improving inflammatory microenvironment. The underlying mechanism of quercetin potentially involves the inhibition of the Toll-like receptor/NFêB signaling pathway and the improvement in the inflammatory microenvironment of the ovarian tissue of the pcos rat model.

Brown adipose tissue has been a recent target for obesity prevention and therapy. Srujana R.et al investigated the novel mechanism through which a well-studied anti-obesity phytochemical resveratrol (RSV) induced browning of white adipose tissue (WAT). The study suggest that RSV-induces change in the polarity of macrophages leading to elevated levels of catecholamines that will induce browning of WAT and contributes to the anti-obesity effects of RSV.

Study by Hossein KJ et al [24] on the effect of pomegranate extract on hormonal changes caused by polycystic ovary syndrome in female wistar rats revealed that the phenolic compounds of pomegranate extract lead to reduced effect of testosterone hormone through inhabitation from formation of dihydrotestosterone receptor complex, reduce secretion of testosterone hormone and increased concentration of oestrogen in wistar rats. Resveratrol came to scientific attention as one part of a possible explanation for the french paradox – the low incidence of obesity and heart disease among french people who eat a relatively high-fat diet. A key factor attributed to this was the French custom of drinking wine with meals. The health benefits of red wine consumption have been attributed to polyphenols; resveratrol, found in high concentrations in red wine, is a major constituent of polyphenols [25].

How does all of this relate to Ayurveda? Thousands of years ago the rishis (sages) with their power of pratyeksha (direct perception) put together herbal super-formulas made with a synergetic blend of numerous fruits and herbs. Two of these are known as chyavanprash and darakchasava. Rammohan Rao a graduate of the California College of ayurveda opines that these tonics were designed to nourish the body and mind, promote longevity and support strong immune system function. These polyphenols promote free radical scavenging, protection by antioxidants, reducesLDL cholesterol levels, reproductive abnormalities, especially anovulation and there by impove womens health [26].

#### Conclusion

There is increasing interest in the dietary components of food and the possible benefits of polyphenolic compounds in women with pcos . Several studies confirm that these compounds have anti-androgenic properties and inhibitory effect on formation of dihydrotestosterone receptor complex. They also reduce secretion of testosterone hormone . These positive results have been shown also in animal models, and their combinations, in clinical applications. Natural polyphenols, may have positive metabolic effects. The road to good reproductive health and extended lifespan also exists in a glass of fruit juice, fruit extract, whole fruits, green leaves, nuts and seeds, all of which contains polyphenols and their derivatives .

Conflict of Interest: Conflict of interest declared none.

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