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DEVELOPMENT OF HEALTH MIX FOR THE MANGEMENT OF POLY CYSTIC OVARY SYNDROME (PCOS) IN WOMEN

*Shubhi Agarwal, **Dr.Usha Devi C, ***Vidya Lal K

Abstract: Polycystic Ovary Syndrome (PCOS) is commonly seen in young women of childbearing age and is mainly characterised by fulfilment of at least two of the following criteria: oligo and/or anovulation, clinical and/or biochemical signs of hyperandrogenism and/or polycystic ovaries (Rotterdam consensus 2003). Studies show that weight loss help in relieving menstrual irregularities in overweight and obese women. Garcinia Cambogia can help women with PCOS in weight loss .The health mix was developed using Garcinia Cambogia in 3 variations – 0.5 gram, 1.0 gram and 1.5 gram as the upper intake level has not been established yet. A nutri-bar was developed using the health mix. Sensory evaluation, proximate analysis and microbial analysis were done for the standard and the variants. The shelf life of the mix was estimated to be 30 days through microbial analysis and peroxide value which was well within the BIS standards. The AOAC methods were adopted for estimating the macronutrient content of the health mix. The mix provided appropriate amount of complex carbohydrates, protein, fibre and reduced amounts of fat as per FDA standards.

Key words: Polycystic ovarian syndrome (PCOS), Garcinia Cambogia, overweight, obese, health mix, weight loss, nutri bar, reduced fat

INTRODUCTION:

Polycystic Ovary Syndrome (PCOS) is a common disorder seen in women of reproductive age. It was first described by Stein and Leventhal in the year 1935 which linked the presence of amenorrhea, polycystic ovaries and hyperandrogenism. PCOS involves irregular ovulation

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in combination with excess androgens and possibly polycystic ovaries. Most of these patients

have clinical evidence of hyperandrogenismincluding hirsutism and/or acne, and/or

androgenic alopecia. Women with PCOS are often overweight or obese, and many have a

family history of diabetes or insulin resistance. This places women with PCOS at greater risk

for type 2 diabetes mellitus (DM), and possibly hypertension, abnormal lipid levels, and

cardiovascular disease. Other than health consequences, these girls are also prone to

psychological implications.

An international consensus definition of PCOS has now been established which requires the

fulfilment of at least two of the following criteria: oligo and/or anovulation; clinical and/or

biochemical signs of hyperandrogenism; and/or polycystic ovaries. (Rotterdam Consensus,

2003)

The prevalence of PCOS in India is 9.13% among adolescent girls aged 15-18 years (Ram et

al, 2011) and it is slowly becoming one of the leading causes of irregular menses and

infertility.

The increase in number of women with PCOS requires an early diagnosis to help in the

treatment and prevent further complications. There is no single inciting factor that triggers the

syndrome and the exact pathophysiology of PCOS is yet to be elucidated. Other than genetic

factors, environmental factors like diet and lifestyleplay a role in the makeup of the PCOS

phenotype. A reduction in 5% total body weight has been linked to reduced insulin levels,

improved mental functions, reduced testosterone levels and improved symptoms of acne and

hirsutism. (Moran et al,2004)

Modifications in the diet and usage of certain products can help in weight loss for PCOS

women. One such product that aids in weight reduction is Garcinia cambogia.

Garcinia gummi-gutta or Kudampuli (Garcinia cambogia) is a fruit that grows widely in

Southeast Asia and in India. The fruit resembles a small pumpkin and varies from green to

yellow in colour. The fruits are ovoid in shape and two inches in diameter. It is commonly

known as "Malabar tamarind" and is widely dried and used in Kerala and in some parts of

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Karnataka for cooking purposes. It is too sour to be eaten raw and is used in curries. Usually the ripe fruit is halved or sectioned and spread in thin layers, dried in the sun for three to seven days to moisture level of about 15 - 20 % and smoked. Other names: Marathy - Dharambe; Telengue - Simachinta; Tamil - Kodakkapuli; Kannada. - Upagimara, simaihunase; Malayalam - Kodampuli.

The active ingredient in Garcinia Cambogia that aids in weight loss is Hydroxycitric acid (HCA) which contributes to the sour taste. The chemical structure of HCA is quite similar to citric acid seen in citrus fruits. Garcinia cambogia contains low amounts of protein, fibre, pectin, reducing sugar and some amounts of anthocyanin pigments when dried. HCA is said to reduce the conversion of carbohydrates into fats. This is done by blocking the enzyme citrate lyase which converts citrate to cholesterol and fat respectively. It suppresses appetite thereby reducing weight gain while increasing the production and storage of glycogen. HCA reduces the availability of acetyl coenzyme A, the building block for fatty acid and cholesterol synthesis (Greenwood *et al*,1991). It aids in the reduction of cholesterol over the reduction of fatty acids. HCA is said to have a thermogenic effect on the body which also aids in weight loss but there is no scientific evidence to prove this statement.

Garcinia cambogia is available in dried forms and as supplements which are being widely popularised as a tool for weight loss. No studies using Garcinia cambogia have been done on humans but many clinical trials have been done on animals to show some significant results in weight loss. Thus, it is said that a low calorie diet along with exercise can show significant weight loss when Garcinia fruit or supplements are consumed. HCA is seen as a way of reducing the level of obesity.

No reports of acute and chronic toxicity have been reported due to the consumption of Garcinia both in the fruit form and as a supplement. It has been proven safe for human consumption and has been used in traditional Indian cookery. The upper intake level for HCA has not been identified. Another major advantage of using HCA for weight loss is that it does not affect the Central Nervous System by reacting with the catecholamine neurotransmitters dopamine and norepinephrine which in turn releases adrenaline (Ohiaet al, 2002).

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Currently, there are no food products available in the Indian market for women with PCOS

though there are varieties of products available for weight loss. The health mix was therefore

formulated keeping weight loss in mind along with the aim of supplying the required

nutrients. It can be incorporated into various food products consumed on a daily

basis. Therefore, an attempt was made to study the acceptability, proximate analysis, shelf life

and microbial load of the health mix.

MATERIAL AND METHODS:

Material and methods

Selection and procurement of raw materials

The raw materials were selected on the basis of health benefits and easy availability of the

ingredients.

The major ingredient used for the study includes:

Dried rind of Garcinia fruit procured from the local supermarket.

Materials for the preparation of health mix:

The current study aimed at selection of the following ingredients for preparation of health

mix:Oats, Wheat bran, Gram flour, Jaggery, Dark chocolate, Flax seeds.

Preparation and standardization of the health mix:

The health mix was tried in different proportions and the best outcome was selected for

further variations with Garcinia. Various recipes were looked at before narrowing it down to

the recipe for health mix from padhuskitchen.com. The basic health mix was made from

various millets and nuts.

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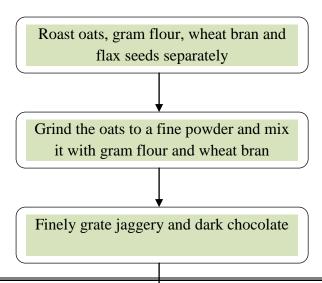
The health mix was prepared with the incorporation of Garcinia cambogia. The health mix made has an addition of jaggery and dark chocolate which were finely grated to make the product more palatable. The health mix was then developed using these ingredients in clean and sterile equipments.

Health mix Recipe (Garcinia cambogia)

Ingredients:

- Rolled oats 30 grams
- Wheat bran -10 grams
- Gram flour 15 grams
- Dark chocolate 15 grams
- Jaggery 25 grams
- Flaxseed 5 grams

The procedure for developing the product is as follows:



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Mix all the ingredients together and store in air-tight container in a dry place

Preparation of variants

Three variants were decided upon and added to the standard. As the upper intake level for

Garcinia and HCA has not been established, the Garcinia powder was taken on par with the

amount present in the supplements available in the market.

Preparation of garciniacambogia powder

Garcinia cambogia is available in the market in smoke dried forms which are added to

curries. The raw form is too sour and often inedible. Each raw fruit weighs around 70-75

grams. The fruit when dried, weighs about 20- 25 grams. In order to obtain in a powder form

to add it to the mix, it was oven dried at 100° C for 10 mins and ground to a fine powder in a

mixer grinder. The powder thus obtained was added to the health mix in three types of

variants

Addition of garcinia powder to health mix

The powder was added into three types of variants:

0.5 gram

1.0 gram

1.5 gram

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The additions were made to 100 grams of the mix respectively. 1.5 gram of Garcinia was chosenas the higher limit beyond which it would be unacceptable due to its strong sour flavour.



Plate 1. Garcinia cambogia powder

Development of Nutri-bar from health mix

Nutri bar was made in three different variations using Garcinia in the amount of 0.5gm, 1.0gm and 1.5gm respectively.100 grams of the health mix was used to make the standard and variations of the nutri bar respectively. There was no addition of any other ingredients. The nutri bar was made in kadai following the method used to make traditional sweets. Control product was made by traditional method of cooking.

Method:

In a pan add the water. Bring to a boil. To this, add the health mix and stir continuously. On a greased plate, make into the shape of bars. Cool and serve.

Table 1 Formulation of Nutri bar

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	Amount (gm)				
Ingredients	Standard	Variation 1	Variation 2	Variation 3	
Rolled oats	30	30	30	30	
Wheat bran	10	10	10	10	
Gram flour	15	15	15	15	
Dark chocolate	15	15	15	15	
Jaggery	25	25	25	25	
Flaxseed	5	5	5	5	
G. cambogia	-	0.5	1.0	1.5	



Plate 2.Nutri bar

Sensory evaluation of nutri bar was done using a 9 -point hedonic scale with semi-trained panellists (confirmed cases of PCOS). The market acceptability of the nutri bar made from the health mix was also checked. Various energy and health bars in the market were chosen based on affordable prices and uniqueness of the bars. Two different bars with Garcinia

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cambogia (0.5 grams) were used for the evaluation. One bar was made replacing the gram flour with whey protein while the other bar retained all the ingredients. The various bars chosen for sensory evaluation were – Yoga bar, Nilgiris millet bar, Crunch bar along with the Garcinia bars. They were procured from a super market, BengaluruThe evaluation was conducted in a clean room with a suitable environment for sensory evaluation which included odour free atmosphere and water in cups to prevent carryover tastes. The room was well lit and served in plates/bowls that did not influence the colour of the food.

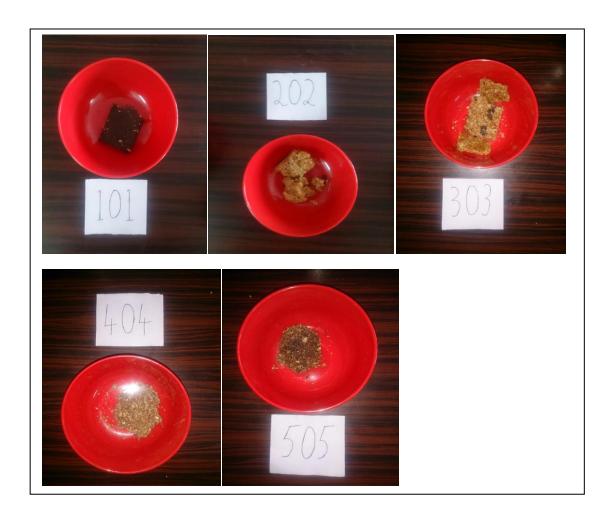


Plate 3.Sensory evaluation to check acceptance of nutri bar

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PROXIMATE ANALYSIS

The standard health mix and the variants were analysed in order to obtain the nutrient

composition. The macronutrients analysed were moisture, ash, protein, fat, crude fibre,

carbohydrate and energy using AOAC methods.

Estimation of Moisture (AOAC, 2000)

Moisture content is known to affect the physical and chemical aspects of food and plays a

major role in the shelf life of the product. An ideal product is required to have low moisture

content. Moisture also affects the freshness and storage of the food as moisture content can

determine the quality of food prior to consumption.

Estimation of total ash (AOAC, 2000)

Ash content represents the mineral content of a food product. The ash constituents include

potassium, sodium, calcium and magnesium, which are present in larger amounts. 3.6.3

Estimation of protein (AOAC, 2000)

The protein content of the sample was estimated using the Kjeldahl method.

Estimation of fat (AOAC, 2000)

Fat was estimated as crude extract with the solvent evaporated and residue weighed.

Estimation of crude fiber (AOAC)

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Crude fiber is a measure of the quantity of indigestible cellulose, lignin, and other

components present in foods. Crude fibre is estimated by acid and alkali treatment in the

sample free of moisture and fat.

Computation of carbohydrate (AOAC, 1995)

Carbohydrate in food can be calculated only after the other components have been estimated.

The content was computed using differential method.

Carbohydrate (g/100g) = 100 - [Protein (g) + Fat (g) + Fibre (g) + Ash (g) + Moisture (g)]

3.6.7 Computation of energy (AOAC, 1995)

Energy needs to be estimated in order to know if the food product is an energy dense product

or not. It was computed by factorial method for the samples.

Energy (kcal) = [protein (g) \times 4] + [carbohydrate (g) \times 4] + [fat (g) \times 9]

SHELF LIFE STUDY

The standard health mix and the variants were analysed in order to obtain the shelf life and

storage stability of the product. The analysis done composed of storage and microbial

analysis and peroxide value determination.

Microbial Analysis

Microbial analysis is done in order to study the shelf life of the product. The health mix was

sealed and stored in plastic covers and opened only during the time of testing. Serial Dilution

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of each sample was done and plating was done using pour plate and spread plate method. Nutrient Agar was used for pour plate method while Potato Dextrose Agar (PDA) was used for spread plate method. The dilutions were used in triplicates and the Colony Forming Units (CFU) in each plate was counted.

Pour plate method was done using 1 ml of the dilutions 10⁻⁷, 10⁻⁸, 10⁻⁹ and incubated at 35°C to 37°C for 24 hours. This is used to estimate the CFU for bacteria. Spread plate method was done using 0.1 ml of the dilutions 10⁻⁴, 10⁻⁵, 10⁻⁶ and incubated at room temperature (25 to 28°C) for 24-48 hours. This estimates the fungal count of the food.



Plate 4. Serial dilution and Plating done under Laminar Air Flow

Determination of peroxide value

Peroxide value (PV) is used as a measurement of the extent to which rancidity reactions have occurred during storage. Other methods are available but peroxide value is the most widely used. PV which is the concentration of peroxide in an oil or fat is useful for assessing the

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extent to which spoilage has occurred. The PV test is used to assess and study the shelf life of the product.

RESULTSAND DISCUSSIONS

SENSORY EVALUATION

The sensory evaluation was conducted using a 9 point hedonic scale with the attributes such as appearance, colour, texture, aroma, taste and overall acceptability. The results were calculated and tabulated.

Table.2Sensory attributes of Nutri bar

Nutri bar	Appearance	Texture	Colour	Odour	Taste	Overall
						acceptability
Standard	6.93 ± 1.64	6.86 ± 1.46	6.93 ± 1.66	6.86 ± 1.43	7.30 ± 1.43	7.16 ± 1.42
Variation 1	7.06 ± 1.44	7.16 ± 1.34	7.43 ± 1.07	6.90 ± 1.30	7.23 ± 1.43	7.30 ± 1.34
Variation 2	6.60 ± 1.30	6.80 ± 2.12	6.10 ± 1.56	6.30 ± 1.51	6.80 ± 1.42	6.76 ± 1.25
Variation 3	6.26 ± 1.36	6.36 ± 1.30	6.30 ± 1.18	6.10 ± 1.24	5.73 ± 1.68	6.06 ± 1.46

It can be seen that the nutri bar with variation 1 was preferred best among all the variations with the highest mean in all attributes. The standard was preferred next followed by variation 2 and variation 3 with a mean of 6.76 and 6.06 respectively.

Table.3 Overall acceptability between the Nutri bar variations

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Variation	Mean	Std. Deviation	F statistic	P value	Multiple compariso n
Standard	7.17	1.42	4.89	0.003*	
Variation 1	7.30	1.34			0.717 ^{ab NS}
Variation 2	6.77	1.25			0.252 ^{ac*}
Variation 3	6.07	1.46			0.004 ^{ad**}

Note: a- Standard; b - Variation 1; c - Variation 2; d- Variation 3

NS- not significant

The table shows that there was a significant difference (F=4.89, P=0.003) at 5%. This means that overall acceptability was not significant between the standard and variation 1. There was significance at 1% between standard and variation 3 with a mean of 6.07.

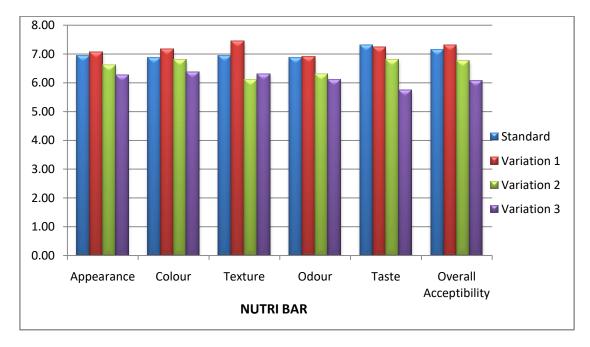


Fig.1 Sensory evaluation of Nutri bar

^{**}significance at 1%

^{*}significance at 5%

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The above figure depicts the sensory attributes for the standard and variants of Nutri bar. variation 1 was the most preferred when comparing the overall acceptability of the products.

SENSORY EVALUATION AND ACCEPTANCE LEVEL OF NUTRI BAR

Table. 4 Sensory attributes of the various bars

Nutri bar	Appearance	Colour	Texture	Odour	Taste	Overall acceptability
Yoga bar	6.71 ± 1.90	6.81 ± 1.69	6.05 ± 2.04	6.10 ± 1.92	5.43 ± 2.36	6.22 ± 1.98
Crunch bar	6.14 ± 1.88	6.24 ± 1.70	5.86 ± 1.68	5.67 ± 2.01	6.00 ± 2.12	5.98 ± 1.88
Nilgiris bar	6.76± 1.00	6.81 ± 0.87	6.62 ± 1.36	6.90 ± 1.30	6.76 ± 1.76	6.77 ± 1.26
Garcinia nutri bar	7.05 ± 1.02	7.00 ± 0.95	6.90 ± 1.04	7.48 ± 0.75	7.76 ± 0.77	7.24 ± 0.91
Garcinia whey bar	6.33 ± 1.56	6.14 ± 1.62	6.38 ± 1.16	6.76 ± 1.37	7.00 ± 1.41	6.52 ± 1.43

From the table it is clearly understood that both the Garcinia bars were preferred by the semi-trained panelists. Garcinia nutri bar was liked best with a mean of 7.24 followed by Nilgiris bar with a mean of 6.77

Table.5 Overall acceptability between the bar variations

Variation	Mean	Std. Deviation	F statistic	P value	Multiple comparison
Garcinia nutri bar	7.24	0.91	2.113	0.085*	
Yoga bar	6.22	1.98			$0.038^{\mathrm{ab}\mathrm{NS}}$
Crunch bar	5.98	1.88			0.009 ac**
Nilgiris bar	6.77	1.26			0.017 ^{ad *}
Garcinia whey bar	6.52	1.43			0.071 ae ***

Note: a- Garcinia nutri bar; b- yoga bar; c- crunch bar; d- Nilgiris bar; e - Garcinia whey bar

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NS- not significant

**significance at 1%

*significance at 5%

***significance at 10%

The above table clearly shows that there was a significant difference (F=2.113, P=0.085) at

10 % between Garcinia nutri bar and Garcinia whey bar. There was no significant difference

between the nutri bar and yoga bar with a mean of 6.22. 5% significance was seen between

the nutri bar and Nilgiris bar. With a mean of 5.98, significance was seen at 1% between

Garcinia nutri bar and crunch bar.

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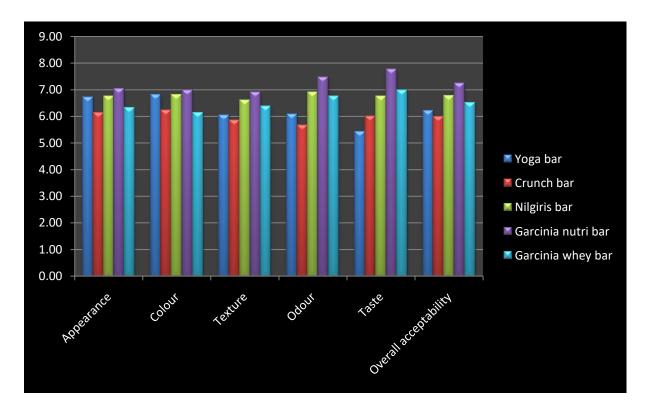


Fig 2. Comparison of various bars with Garcinia bars

From the figure it is seen that Garcinia nutri bar was best preferred with the highest attributes among all the various bars that were present. Garcinia nutri bar was overall acceptable among the market bars.

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PROXIMATE ANALYSIS

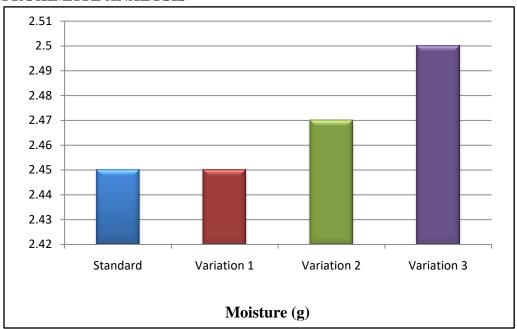


Fig 3. Comparison of moisture content (g/100g) of the standard and variations using Garcinia cambogia $\ \ \,$

The moisture level was seen to be the highest in variation 3 with a moisture level of 2.5g/100g. Variation 2 had a moisture level of 2.47g/100g and variation 1 had 2.45g/100g on par with the standard. This suggests that Garcinia powder maybe hygroscopic in nature.

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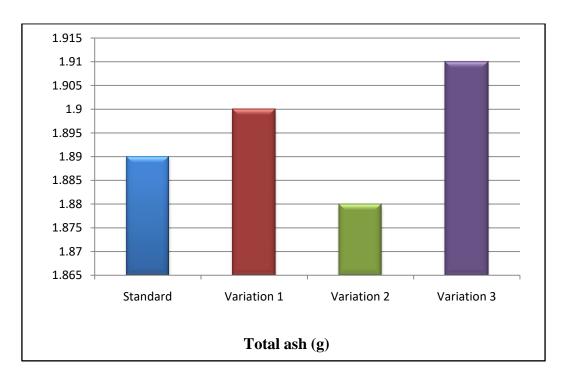


Fig.4 Comparison of total ash content (g/100g) of the standard and variations using Garcinia cambogia

The ash content of the standard was found to be 1.89g/100g. Variation 1 had 1.90g ash; variation 2 had 1.88 g of ash. The highest ash content was seen on variation 3 with 1.91 g ash per 100g

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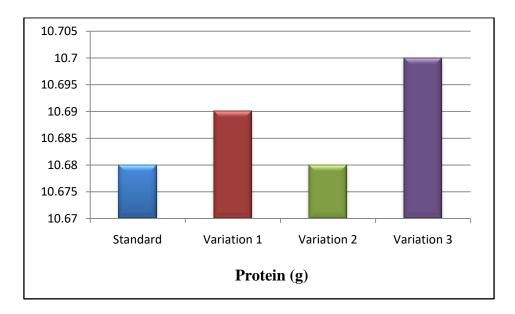


Fig.5 Comparison of protein content (g/100g) of the standard and variations using Garcinia cambogia

The protein content was similar in all the variants. The standard had 10.68g/100 g protein while variation 2 had a slightly higher amount of protein at 10.69g/100 g. Variation 2 had the same amount of protein as the standard and variation 3 has 10.70g protein

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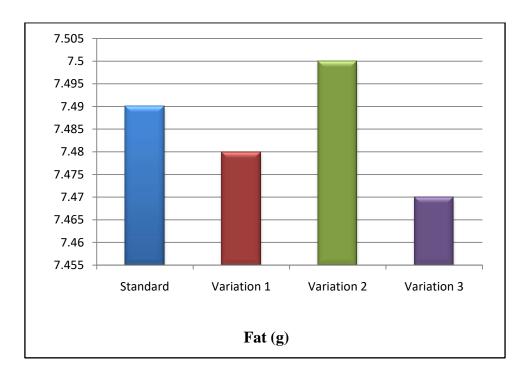


Fig.6 Comparison of fat content (g/100g) of the standard and variations using Garcinia cambogia

The fat content of the standard was found to be 7.49g/100g. Variation 1, 2 and 3 had similar fat content with 7.48g, 7.50 g and 7.47 g respectively. The product has reduced amounts of fat in comparison to Food and Drug Administration (FDA) standards

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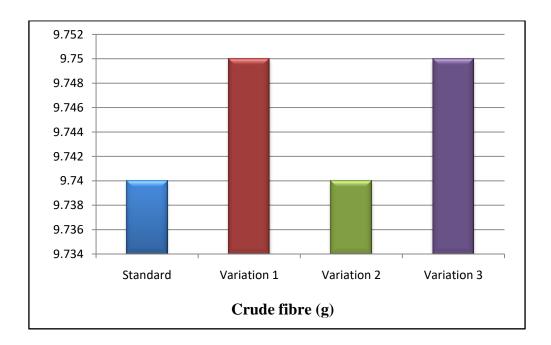


Fig.7Comparison of crude fibre content (g/100g) of the standard and variations using Garcinia cambogia

The fibre content of the variation 1 and variation 2 was found to be 9.75g/100g and 9.74g/100g respectively, Variation 3 had the same fibre content as variation 1 while standard had 9.74 g of fibre.

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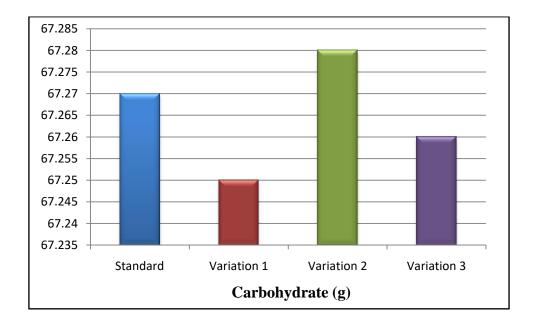


Fig 8. Comparison of carbohydrate content (g/100g) of the standard and variations using Garcinia cambogia

The carbohydrate content of the standard was 67.27g/100g. Variation 2 had the highest amount at 67.28g/100g. The values of variation 1 and 3 were also similar and found to be 67.25 g and 67.27 g respectively. There is appropriate amount of carbohydrates in the form of complex carbohydrates.

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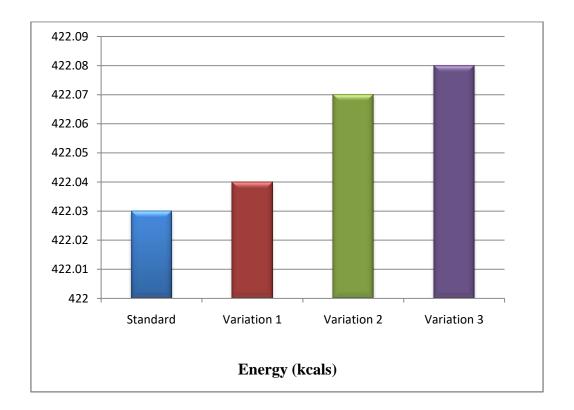


Fig 9. Comparison of total calorie content (Kcal/100g) of the standard and variations using Garcinia cambogia

The figure depicts the total calorie content of the health mix. The standard had 422.03 Kcal/100g. Variation 1, 2 and 3 has similar calories values at 422.04 Kcals, 422.07 Kcals and 422.08 Kcals respectively.

Table.6 Proximate values of health mix

Nutrient	Proximate value
Energy (Kcal)	422.03
Protein (g)	10.68
Fat (g)	7.49
CHO (g)	67.27
Fibre (g)	9.74

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The table shows the complied proximate values of the standard health mix.

4.5 SHELF LIFE STUDY

4.5.1 MICROBIAL ANALYSIS (DAY 1 TO DAY 30)



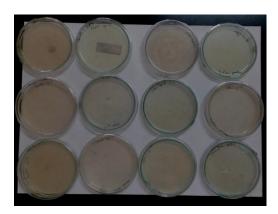


Plate.5 Microbial analysis on day 1

Day 1

Plate: Standard, Variation 1, Variation 2 and Variation 3

(Nutrient Agar- dilution 10^{-7} , 10^{-8} , 10^{-9})

Day 1

Plate: Standard, Variation 1, Variation 2 and Variation 3

(Potato Dextrose Agar - dilution 10^{-4} , 10^{-5} , 10^{-6})





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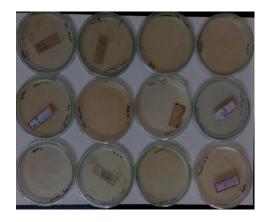


Plate
6.
Micr
obial
analy
sis on
day
15

Plate.7 Microbial analysis on day 30

Day 30

Plate: Standard, Variation 1, Variation 2 and Variation 3

(Nutrient Agar- dilution 10^{-7} , 10^{-8} , 10^{-9})

Day 30

Plate: Standard, Variation 1, Variation 2 and Variation 3

(Potato Dextrose Agar - dilution 10^{-4} , 10^{-5} , 10^{-6})

The shelf life study demonstrated a good keeping quality for all the products for 30 days.

This is contributed by aseptic preparation and storage of the health mix.

Table.7 Microbial analysis of the standard and variations of health mix

Variation of health mix	Day	Appearance of bacterial and fungal colonies
	1	None
Standard	15	None
	30	None
	1	None
Variation 1	15	None
	30	None
	1	None

Plate: Star Variation

(Nutrient Ag

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Variation 2	15	None
	30	None
	1	None
Variation 3	15	None
	30	None

The table shows the microbial analysis of standard and variations of the health mix for 30 days.

MICROBIAL ANALYSIS (DAY 45 TO DAY 60)

Table.8 Microbial analysis of the standard health mix

Day	Test	Result	
45	Total Bacterial Count	900 cfu/100g	
	Total Fungal Count	20 cfu/100g	
60	Total Bacterial Count	20,900 cfu/100g	
	Total Fungal Count	730 cfu/100g	

The above table shows the total bacterial and fungal count estimated by Chennai mettex lab on the 45th day and 60th day respectively.

DETERMINATION OF PEROXIDE VALUE

Table 9.Peroxide value of the standard and variations of health mix

Variation of health mix	Day	Peroxide value (mEq/kg)
	1	4.6
Standard	15	5.0
	30	5.4
	1	4.5
Variation 1	15	4.9
	30	5.1
	1	4.6
Variation 2	15	4.9
	30	5.0
	1	4.5

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Variation 3	15	4.8
	30	5.2

The peroxide value was estimated on the 1st day, 15th day and 30th day and the values were found to be well within the Bureau of Indian Standards (BIS) limits.

CONCLUSION

The developed health mix can positively treat the symptoms of PCOS, aid in weight loss and regulate body metabolism. From the microbial analysis it was confirmed that the health mix is GRAS(generally regarded as safe) which can be consumed on an average for a month. Garcinia cambogia is the next trend in weight loss and is gaining importance across the globe for its ability to reduce fat and suppress appetite. PCOS is becoming a common disorder seen in women of reproductive age. A lot of women are unaware of the condition and the number of women and young girls affected are increasing day by day. Garcinia cambogia can be the cure for PCOS with respect to weight loss. The health mix can be incorporated in various recipes using creativity.

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