Impact of panchratna juice in the management of diabetes mellitus: Fresh vs. processed product

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Many of the plant sources have excellent nutrient and non-nutrient properties which can be exploited to manage clinical conditions. The current study was undertaken to assess the impact of panchratna juice (Amla, Tulsi, Ginger, Mint and Turmeric) in the management of type 2 diabetes mellitus subjects. A total of 55 stable type 2 diabetics were enrolled from pathology laboratories and were given either fresh panchratna juice (n = 15) for 45 days or processed panchratna juice (n = 20) for 90 days and compared with diabetic controls (n = 20). Anthropometric profile, glycemic status and lipid profile were assessed on all the subjects at baseline, 45 days and 90 days. Students t test, paired t test and ANOVA were applied using Microsoft Excel. The results indicated that fresh panchratna juice or processed panchratna juice supplementation for long-term did not have any significant impact on the glycemic and lipemic status of diabetic subjects. Thus, the protective effects are lost with processing, and therefore, these plant sources should be consumed in their natural form.

Key words: Fasting blood sugar, glycosylated hemoglobin, lipid profile, panchratna juice, T2DM (Type 2 diabetes mellitus)

INTRODUCTION

Diabetes mellitus remains a major public health problem and prevention of diabetes still lies in the realm of future. For thousands of years, plants have played a significant role in maintaining human health and improving the quality of human life. Herbal medicine is increasingly gaining acceptance from the public and medical profession due to greater advances in the understanding of the mechanisms by which herbs positively influence health and quality of life.[1] Leading the way in the new understanding is the discovery of herbs as potent free radical scavengers-antioxidants.[2]

Many of the medicinal plants have active components and are extensively used in India. Some of the common medicinal plants like Tulsi (*Ocimum sanctum*), Ginger (*Zingiber officinalis*), Turmeric (*Curcuma longa*), Mint (*Mentha piperita*) and Amla (*Embilica officinalis*) have been used extensively for various therapeutic purposes. Individually, these plant products have clinical utility.[3-9] However, the impact of a combination of five products (Tulsi + Ginger + Turmeric + Mint + Amla = Panchratna) in the form of juice has not been investigated so far. The present study aimed to see the effect of panchratna juice supplementation (fresh and processed) on the metabolic control of type 2 diabetes mellitus (T2DM) subjects.

MATERIALS AND METHODS

Preparation of Fresh Panchratna Juice

All the ingredients namely Amla (*Embilica officinalis*), Tulsi (*Ocimum sanctum*), Mint (*Mentha piperita*), Ginger (*Zingiber officinalis*) and Turmeric (*Curcuma longa*) were purchased from the local Vadodara market. These products are cultivated in villages around Vadodara city. They were thoroughly washed under running water. Amla was deseeded. Leaves of Tulsi and Mint were separated from the stem. Ginger and Turmeric were peeled to remove the skin. The ingredients were then chopped into small pieces. All the ingredients were then ground into a fine paste in an electric mixer using minimum amount of water. The volume was made up to 150 ml with the addition of water. The entire procedure was standardized with varying volumes and quantities of ingredients. Based on sensory evaluation by a panel of judges using the hedonic scale, the most acceptable composition (150 ml fresh juice) was chosen for the study. As 30 g of pulp represented an equivalent amount of fresh juice ingredients, this amount of pulp was reconstituted to 150 ml.

Preparation of Processed Panchratna Juice

Pre-preparation of the ingredients was similar to that of the fresh panchratna juice. However, the processed pulp was made in the food industry (one batch) unlike fresh juice which was made daily. All the ingredients were ground into a fine paste in a pulverizer. The closed container containing the pulp was then put in hot boiling water in order to sterilize. The pulp was then bottled

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immediately in sterilized glass bottles with airtight lids. No preservative was added to the pulp. These bottles were stored in a refrigerator. Each bottle contained 150g of the pulp which sufficed for 5 days (30 g/day).

Selection of Subjects

For the study, fifty five stable T2DM subjects who gave verbal consent were enrolled from the pathology laboratories of the city. These subjects were then divided into three groups namely fresh, processed and control group based on the willingness of the subjects. With an average FBS value of 150 mg/dl, SD of 45 mg/dl and an expected 20% decrease in FBS at $p < 0.05$ level the minimum sample size required in each group would be 13. The fresh group (n = 15) was given 150ml of fresh panchratna juice daily for a period of 45 days. The juice was delivered daily at the subject's residence in the morning by the investigator and hence this was a supervised trial. The processed group (n = 20) received bottled panchratna pulp for a period of 90 days. The subjects were given one bottle, every five days and were asked to store it in the refrigerator once the seal was opened. The subjects in the processed group were asked to reconstitute the pulp by mixing 30 g of the pulp (two tablespoons) to 150 ml of water. For this, the patients were trained by the investigator through demonstrations. The fresh juice and the processed juice were consumed by the subjects daily in the morning. Since many of the patients in both the groups were hypertensives, they were asked not to add salt to the juice while drinking. The control group (n = 20) received no supplementation. During the course of supplementation subjects were asked not to alter their diet and medication. None of the subjects took other complementary or alternative medications during the study period. The baseline data was collected on general information, anthropometry, medical history and 24 hour dietary recall along with fasting blood sugar (FBS), lipid profile and glycated hemoglobin (HbA1C). FBS and lipid profile levels were quantified as per standard procedures using enzymatic kits. HbA1C was estimated using ion exchange resin method. Low density lipoprotein cholesterol (LDL-C), Very low density lipoprotein cholesterol (VLDL-C) and Non High density lipoprotein cholesterol (Non HDL-C) were estimated by calculations. All parameters were monitored at baseline, 45 days and 90 days. The study was approved by the Ethical Committee of the department of Foods and Nutrition, The M S University of Baroda, Vadodara (Approval No: FCSC/FND/ME 17; dated: 15/7/2006). Due to ethical reasons the control group received the bottled panchratna pulp after the intervention period.

Phytochemical Analysis of the Individual Ingredients

Phytochemical analysis was carried out on the leaves, stem and roots of the plants: *Embilica officinalis, Mentha piperita, Zingiber officinalis, Curcuma longa and Ocimum sanctum*. All the plants were procured from the local market. The plants were cleaned and washed under running water to remove dirt and other impurities and later washed with distilled water. They were then shade dried for a week and then oven dried at 60°C for two to three days. The dried powder was used for analysis of chemical constituents. Thin layer chromatography and paper chromatography was done to analyse flavonoids, phenolic acids, glycoflavones, quinones and steroids on all the samples.

Statistical Analysis

Results are expressed as Mean ± S.D. The significance of the data was evaluated using ANOVA, students't test and paired t test. The statistical analysis was carried out using Microsoft Excel.

RESULTS

The composition and the nutritive value of fresh panchratna juice shows that it is rich in Ascorbic acid and Beta Carotene, which are considered as good antioxidants [Table 1]. The results of the phytochemical analysis showed the presence of flavonoids in Amla, Tulsi and Mint. Phenolic acids and steroids were present in all the samples. Glycoflavones were found to be absent in all the samples [Table 2]. The main flavonoids present in amla were Quercetin and Gallic acid. Apigenin was found to be present in Tulsi leaves and the predominant flavonoid found in Mint leaves was Scutellarin. The major phenolic acids found in all the plant samples were Syringic acid, Vanillic acid, Mellilotic acid and Gallic acid [Table 3].

Table 1: Nutrient composition of panchratna juice

Ingredients	Amount (g)	Energy (Kcal)	Protein (g)	Fats (g)	CHO (g)	Fibre (g)	β Carotene (µg)	Vitamin C (mg)
Amla	50	29	0.25	0.05	6.85	1.7	1.12	300
Tulsi	10	-	2	0.7	4.9	2.3	NA	NA
Mint	20	9.6	0.96	0.12	1.16	0.4	54.8	5.4
Ginger	10	6.7	0.23	0.09	1.23	0.24	1	1.3
Turmeric	5	17.4	0.31	0.25	3.47	0.13	0.37	-
Final volume	150 ml with water							

CHO-Carbohydrate NA-Not available

Mint and tulsi leaves were also analyzed for the presence of volatile oils [Table 4]. The characteristic flavor and smell of these plants is due to the presence of volatile oils. The characteristic yellow color of turmeric is due to the presence of Curcuminoids. Ginger was analyzed for the presence of its active principles Gingerol and Shogoals. Tulsi leaves showed the presence of mucilage. Thus, the phytochemical analysis revealed a mixture of active components beneficial to health.

The diabetic subjects were in the age group of 50–65 years [Table 5]. Majority of the subjects were overweight as indicated by their body mass index (BMI). In all the three groups, female diabetics had higher BMI and waist circumference (WC) as compared to the male diabetics. The background information related to the risk factors showed that only two and one male subject consumed tobacco and alcohol respectively. None of the subjects smoked. Regular physical activity was not seen among the enrolled diabetic subjects. Thus the background variables were comparable between the three groups.

After 45 days of fresh Panchratna Juice supplementation, there was a transient fall in the FBS and HbA1C levels (7 % and 3 % respectively). These levels reverted back near to the baseline values at the end of washout period of 45 days [Figures 1 and 2]. When the subjects were categorized on the basis of their initial FBS levels, the percent reduction in the FBS and HbA1C levels was 9.15 % and 5.82% respectively in subjects with initial FBS >150 mg/dl in the experimental group as compared to 2.94 % and 0.43% in the control group [Table 6]. Supplementation of fresh panchratna juice did

Table 2: Phytochemical analysis of the plant ingredients of panchratna juice

Plant	Flavonoids	Phenolic acids	Steroids	Quinones	Glyco-flavones
Embilica officinalis (Amla)	+	+	+	+	-
Ocimum sanctum (Tulsi)	+	+	+	-	-
Mentha piperita (Mint)	+	+	+	+	-
Zingiber officinalis (Ginger)	-	+	+	+	-
Curcuma longa (Turmeric)	-	+	+	+	-

(+) stands for presence in the ingredient (-) stands for absence in the ingredient

Table 3: The distribution of various flavonoids and phenolic acids in the ingredients of panchratna juice

Name of plant	Part used	Flavonoids	Phenolic acids
Embilica officinalis	Fruit	3', 4' di OMe Quercetin Gallic acid	Gallic acid Syringic acid Vanillic acid
Ocimum sanctum	Leaves	4' OMe Apigenin Apigenin	Syringic acid Mellilotic acid
Mentha piperita	Leaves	4' OMe Scutellarin OH Scutellarin	Syringic acid Mellilotic acid Gallic acid
Zingiber officinalis	Root	Absent	Mellilotic acid Caffeic acid 2 OH. 5 OCH_3 β Resorcyclic (2,4 dihydroxy benzoic acid) γ Resorcyclic (2,6 dihydroxy benzoic acid) 2 OH 6.OCH_3
Curcuma longa	Root	Absent	Phydroxy benzoic acid Mellilotic acid Syringic acid Vanillic acid

Table 4: Presence of active components in plant ingredients of panchratna juice

Plant	Component present
Embilica officinalis	-
Mentha piperita	Volatile oil
Ocimum sanctum	Volatile oil
Curcuma longa	Curcuminoids
Zingiber officinalis	Shogoal, Gingerol

Table 5: Clinical profile of the diabetic subjects (Mean ± SD)

	Control	Fresh juice	Processed juice
n = 55	20	15	20
Sex			
Male	12	11	14
Female	8	4	6
Age (Y)			
Male	52±6	61±7	60±8
Female	57±9	54±9	65±9
Height (cm)			
Male	164±7.13	164±8.10	164±6.77
Female	157±3.35	156±4.27	155±4.01
Weight (Kg)			
Male	71.17±5.98	69.18±4.73	68.71±8.02
Female	69±2.88	76.50±5.20	65.17±9.83
Body mass index (Kg/m^2)			
Male	26.36±1.41	25.95±2.26	25.50±1.72
Female	28.25±1.63	31.71±3.04	27.29±3.77
Waist circumference (cm)			
Male	88.5±8.19	95.91±3.48	89.79±8.56
Female	92.88±10.45	103.5±5.80	90.67±7.37

not alter the lipid levels [Table 7]. Further, the levels were sustained after the washout period indicating no adverse or beneficial effects of panchratna juice. In line with the observations of lipid profile, the atherogenic indices also remained unaltered throughout the study period [Table 8].

When supplementation data for the processed panchratna juice was looked into it was observed that the fall in the FBS was 9.56% and 15.8% at the end of 45 and 90 days of supplementation. However, the reductions were not statistically significant. Similarly the reduction in the HbA1C was 3.22% and 5.87%, respectively, after 45 and 90 days. In the control group the FBS and HbA1C levels remained unaltered throughout the study period [Figures 1 and 2]. Based on the initial FBS levels, the percent drop in the FBS levels was 21.16% in subjects with initial FBS >150 mg/dl in the experimental group as compared to 4.8% in the control group [Table 9]. With a 21% reduction in FBS values the HbA1C levels fell by 8.08%. The non significant fall in the FBS and HbA1C values indicate that the reduction was a transient one and not a physiological one.

Long term supplementation with processed panchratna juice for a period of 3 months did not bring about any change in the lipid profile of the subjects [Table 7]. There was a non significant reduction (11.2%) in the values of triglyceride (TG). With the fall in TG values, the TG/HDL-C, which represents small dense lipoprotein showed a reduction (4.12 *vs.* 3.5) in the experimental group [Table 8]. This observation is important as small dense lipoprotein is more atherogenic and reduction in the levels may lower the risk for cardiovascular diseases.

DISCUSSION

Plants have played a significant role in maintaining human health and improving the quality of human life for thousands of years. A number of herbs such as amla, turmeric, ginger, tulsi have shown hypoglycemic and hypolipidemic properties. Few studies are available which

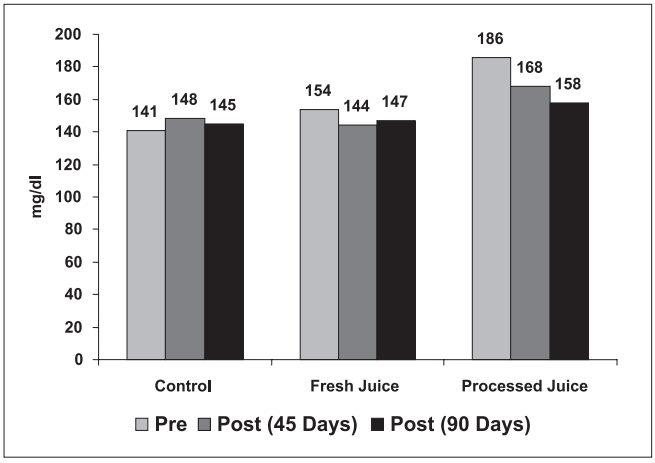

Figure 1: Impact of fresh and processed panchratna juice supplementation on blood sugar levels in type 2 diabetic subjects

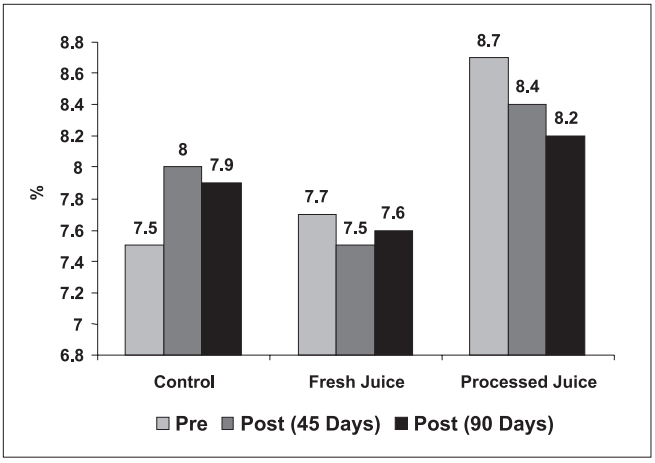

Figure 2: Impact of fresh and processed panchratna juice supplementation on HbA1C levels in type 2 diabetic subjects

Table 6: Impact of fresh panchratna juice supplementation on blood sugar and HbA1C levels in diabetics with initial fasting glucose levels >150 mg/dl (Mean ± SD)

	Control		Fresh juice	
	(n = 6) >150 mg/dl	(n = 14) <150 mg/dl	(n = 6) >150 mg/dl	(n = 9) <150 mg/dl
FBS				
Pre	199.60±40.22	115.31±17.55	201.48±38.5	122.7±11.73
Post (45 days)	193.73±56.52	127.68±19.80	183.03±50.2	118±16.20
Post (90 days)	189.85±54.45	125.51±18.08	189.72±42.79	119.12±15.73
F value	0.05	1.78	0.26	0.27
HbA1C				
Pre	9.46±0.90	6.63±1.76	8.92±1.18	6.96±0.26
Post (45 days)	8.95±1.41	7.62±0.89	8.40±1.36	6.93±0.29
Post (90 days)	9.10±1.08	7.32±0.59	8.50±1.31	6.99±0.42
F value	0.31	2.57	0.25	0.07

Post (90 days) in case of panchratna juice is washout period.

have investigated the influence of combinations of herbs on glucose levels and lipid profile of diabetic subjects. Therefore the present study was undertaken to assess the effect of panchratna juice supplementation on metabolic control of Type 2 Diabetes Mellitus (T2DM) subjects. Panchratna juice is a concoction of *Embilica officinalis, Ocimum sanctum, Mentha piperita, Zingiber officinalis and Curcuma longa.*

In the present study the major flavonoids identified in Amla were Gallic acid and 3′, 4′ di OMe Quercetin. Many studies have shown Quercetin as the most potent antioxidant. Another ingredient in the juice was tulsi. *Ocimum sanctum* is rich in volatile oils (Eugenol, cineole, borneol) besides Apigenin, syringic and Mellilotic acid. The medicinal parts of *Mentha piperita* include volatile oil, flavonoids, phenolic acids and triterpenes. Mint has high menthol content. The other active ingredients are menthone and menthyl acetate. Peppermint due to its volatile oil is used as an expectorant and decongestant and used to treat many ailments including asthma, bronchitis, sinusitis and cough. Mint oil is known to stimulate the gall bladder and secretion of bile. It is known to have analgesic, antiseptic and carminative activities.[10] Various studies have shown the antiemetic effect of ginger which is due to its active principles Gingerol and Shogoals. Ginger contains a digestive enzyme called Zingibain which exceeds papain (from papaya) in digestive potency. Turmeric or *Curcuma longa* has been used in Indian systems of medicine for a

Table 7: Impact of fresh and processed panchratna juice supplementation on lipid profile in Type 2 diabetic subjects (Mean ± SD)

	Control (n = 20)	Fresh juice (n = 15)	Processed Juice (n = 20)
TC			
Pre	192.6±38.16	167.63±32.18	187.23±36.00
Post (45 days)	188.6±38.32	164.69±30.39	179.31±28.28
Post (90 days)	187.7±39.28	165.13±30.33	183.03±57.50
F value	0.09	0.03	0.16
HDL-C			
Pre	43.2±6.31	40.38±5.56	43.65±6.06
Post (45 days)	43.3±6.63	41.48±5.36	45.25±5.68
Post (90 days)	43.5±6.52	40.77±5.55	45.35±4.14
F value	0.01	0.15	0.60
LDL-C			
Pre	115.8±29.32	100.21±36.06	108.06±29.08
Post (45 days)	112.4±32.64	97.87±31.86	99.61±27.10
Post (90 days)	111.2±32.42	96.87±33.64	106.15±51.82
F value	0.11	0.03	0.27
VLDL-C			
Pre	33.6±12.75	27.07±14.63	35.52±16.43
Post (45 days)	32.9±11.87	26.28±11.68	34.45±15.47
Post (90 days)	33.0±11.46	27.49±12.70	31.54±11.90
F value	0.02	0.03	0.39
TG			
Pre	168.2±63.76	135.39±73.12	177.58±82.15
Post (45 days)	164.5±59.34	131.44±58.38	172.25±77.37
Post (90 days)	165.0±57.32	135.33±63.52	157.68±59.52
F value	0.02	0.01	0.39

Post (90 days) in case of panchratna juice is washout period.

Table 8: Impact of fresh and processed panchratna juice supplementation on atherogenic indices in Type 2 diabetic subjects (Mean ± SD)

	Control (n = 20)	Fresh Juice (n = 15)	Processed juice (n = 20)
TG/HDL-C			
Pre	4.1±1.41	3.47±2.11	4.12±1.98
Post (45 days)	4.0±1.21	3.30±1.82	3.86±1.84
Post (90 days)	4.0±1.15	3.45±1.92	3.50±1.37
F value	0.04	0.03	0.59
LDL-C/HDL-C			
Pre	2.7±0.78	2.5±0.90	2.49±0.65
Post (45 days)	2.7±0.92	2.39±0.79	2.23±0.66
Post (90 days)	2.6±0.93	2.41±0.86	2.33±1.05
F value	0.07	0.06	0.52
TC/HDL-C			
Pre	4.5±1.01	4.2±0.87	4.32±0.75
Post (45 days)	4.4±1.09	4.01±0.77	4.00±0.71
Post (90 days)	4.4±1.12	4.11±0.85	4.03± 1.13
F value	0.08	0.20	0.72
TC/LDL-C			
Pre	1.7±0.16	1.80±0.53	1.79±0.31
Post (45 days)	1.7±0.23	1.79±0.48	1.88±0.37
Post (90 days)	1.7±0.21	1.83±0.54	1.86±0.36
F value	0.28	0.03	0.26

Post (90 days) in case of panchratna juice is washout period.

Table 9: Impact of processed panchratna juice supplementation on blood sugar and HbA1C levels in diabetics with initial fasting glucose levels >150 mg/dl (Mean ± SD)

	Control		Processed juice	
	(n = 6) >150 mg/dl	(n = 14) <150 mg/dl	(n = 10) >150 mg/dl	(n = 10) <150 mg/dl
FBS				
Pre	199.60±40.22	115.31±17.55	245.77±50.85	125.67±12.77
Post (45 days)	193.73±56.52	127.68±19.80	213.09±55.75	22.81±23.71
Post (90 days)	189.85±54.45	125.51±18.08	193.76±42.06	121.25±17.24
F value	0.05	1.78	2.77	0.14
HbA1C				
Pre	9.46±0.90	6.63±1.76	10.02±1.45	7.35±0.72
Post (45 days)	8.95±1.41	7.62±0.89	9.63±1.62	7.17±0.65
Post (90 days)	9.10±1.08	7.32±0.59	9.21±1.16	7.13±0.41
F value	0.31	2.57	0.81	0.35

long time. The rhizome contains curcuminoids, curcumin, di methoxy curcumin, bis- dimethoxy curcumin, 5′ methoxy curcumin which are natural antioxidants. Turmeric has anti inflammatory action which could be due to leukotriene inhibition. The hypoglycemic and hypolipidemic activity of turmeric, ginger, fenugreek seeds, holy basil leaves and curry leaves has been reported earlier.[11-16]

The active components of *Curcuma longa* such as curcumin and tetrahydrocurcumin (THC) also posses antidiabetic, anti-inflammatory and antioxidant activity and nephroprotective action.[11] The effect of THC on glycoproteins studied in normal and Streptozotocin (STZ) induced rats for 45 days showed decrease in levels of blood glucose and glycoproteins. Plasma insulin and sialic acid levels increased when treated with THC, establishing its beneficial effect on glycoproteins.[16]

Hyponidd is a herbomineral formulation composed of the extracts of ten medicinal plants (*Momordica charantia, Melia azadirachta, Pterocarpus marsupium, Tinospora cordifolia, Gymnema sylvestre, Enicostemma littorale, Emblica officinalis, Eugenia jambolana, Cassia auriculata and Curcuma longa).* Oral administration of hyponidd (100 mg/kg body weight and 200 mg/kg body weight for 45 days resulted in significant lowering of the blood glucose levels, and decreased the levels of glycosylated hemoglobin, plasma thiobarbituric acid reactive substances, hydroperoxides, ceruloplasmin and alpha-tocopherol in diabetic rats, thus exhibiting antihyperglycemic and antioxidant activity in STZ-induced diabetic rats.[17] Recently Ansarullah evaluated the antihyperlipidemic properties of a polyherbal extract consisting of *Cassia fistula L.*, Caesalpiacea, *Ocimum sanctum L.*, Lamiaceae, *Annona squomosa L.*, Annonaceae, *Terminalisa arjuna Roxb.*, Combretaceae, *Azadirecta indica A.* Meliaceae, *Aegle marmelose (L)* Correa ex Roxb *and Rutaceae* in Tyloxapol induced hyperlipidemic rats. Intragastric administration of polyherbal extract (500 mg/kg of body weight) significantly decreased plasma cholesterol, triglyceride, non-HDL-C and phospholipids levels and increased HDL-C levels. The polyherbal extract was comparable to the reference drug Lovastatin and can be used as an alternative therapeutic agent in the treatment of hyperlipidemia.[18]

Though the raw panchratna ingredients had a variety of bioactive compounds when the components were processed either for fresh juice or pulp no positive modulatory effects were seen on glycemic or lipemic status of diabetic subjects. However, supplementation showed some impact in subjects who had higher initial fasting sugar, indicating the potential of its individual ingredients and the juice as a whole for its hypoglycemic and hypolipemic properties. Further, the diabetic subjects reported no side effects. There is a need to look at the antioxidant status of the subjects after supplementation.

CONCLUSIONS

Various documentary evidences, along with the phytochemical analysis done by us reveal that the above five plant products have excellent nutritional properties and may impart positive health benefits. Further, our supplementation studies with whole fruit amla (35 g/day) for a period of 60 days or dry tulsi powder leaves (1 g/day) have shown significant improvements in the lipid profile of diabetic subjects.[3,4] Though we have not characterized the various ingredients from panchratna juice and the processed product, keeping in mind our earlier observations with fresh amla and dry tulsi powder, we feel that fresh whole products of herbs/fruits are better than the processed or juice form. It is possible that the active ingredients are lost during processing or the network of bioactive compounds present in the natural food may be disrupted while processing. In view of these observations, we recommend that the herbs/fruits be consumed in its natural form to get the desired effects on health. Further it is possible that higher amounts of the concoction may be required to bring about the desired effects. Future studies should focus on identifying the bioactive compounds in the fresh concoction and the processed product in order to quantify the amounts to be consumed for the desired effects.

Now-a-days, a number of herbal drinks and fruit and vegetable juices are available in the morning, near gardens where people go for morning walks. Among the various herbal drinks panchratna juice and wheat grass juice are very popular among the public. In view of these observations we need to sensitize and encourage people to consume whole fruits or herbs for potential health benefits.

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