

# Effect of *P. serratifolia* Leaf Extract on Hematological Parameters and Standarization of Snacks using *P. serratifolia* Leaf Powder

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## Abstract

*Premna serratifolia* (also known as Pachumullai) belongs to the family Verbenaceae, is a medicinal plant known as Agnimantha in Ayurveda. It has potential hepatoprotective and cytotoxic activity. There are many phytochemical and bioactive compounds present in the plant parts of *Premna serratifolia*. In addition to this, it has higher antioxidant and antimicrobial activity which is beneficial to human health as a therapeutic adjuvant. However, the nutritional components in *P. serratifolia* have not been probed, though it was consumed during special rituals. Further the anti-anemic effect of *P. serratifolia* was not yet studied. The present research aimed at assessing the micronutrients (Iron, Phosphorus, Calcium, Ascorbic acid) in *P. serratifolia* leaf, and the ethanolic extract of *P. serratifolia* leaf at two different dosage (100mg and 200mg) was examined for hematological parameters (Hemoglobin, Red Blood Cell count and Mean Corpuscular Hemoglobin) in anemic rats and the value added snacks like biscuit, vadam and murukku were formulated at two different ratios by using *P. serratifolia* leaf powder. The results showed that *P. serratifolia* leaf extract had higher impact on the hematological parameters when compared with standard drug ferrous fumarate and the higher incorporation of *P. serratifolia* leaf powder helped to increase the nutrient content in formulated snacks.

**Keywords:** Anti-Anemic, Ethanolic Extract, Ferrous Fumarate, *P. serratifolia*, Value Added Snacks

## 1. Introduction

India has the world's highest prevalence of iron deficiency anemia among women, with 60 to 70 percent of the adolescent girls being anemic<sup>1</sup>. Anemic condition tends to have negative impact on physical performance, mostly work productivity due to reduced oxygen transport<sup>2</sup>. The first victim of lifestyle changes has been food habits. Consumption of junk food has increased manifold, which has led to a number of diseases related to nutritional deficiencies, and nutraceuticals play an important role in controlling them<sup>3</sup>. A term combining the words nutrition (a nourishing food or food component) and Pharmaceutical (a medical drug)

is a food or food product that provides health and medical benefits, including the prevention and treatment of diseases<sup>4</sup>.

India is endowed with an estimated 47,000 species of plant that include around 8000 plants which are known to have medicinal properties<sup>5</sup>. Among various medicinal plants *Premna serratifolia* is one of the medicinal plant belonging to the family of verbenaceae also known as Agnimantha in Ayurvedha. It is a large shrub or a small tree up to 9m in height with yellowish lenticulate bark, spinous large branches and yellowish brown woody aromatic root, leaves, simple, opposite, sometimes whorled, elliptic ovate<sup>6</sup>.

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The GC-MS chromatogram shows the presence of 22 major peaks with the retention time ranging between 2.72 and 34.54. The nature of the compounds obtained are of monoterpene oxide, monoterpene alcohol, sesquiterpene, terpene alcohol, alcoholic compound, peripionate compound, linolenic acid ester, diterpene, acetate compound, alkene compound, triterpene, unsaturated fatty acid ester, vitamin compound, ketone compound and acetate compound<sup>7</sup>.

The leaf extract of *P. serratifolia* was most susceptible anti microbes like *Klebsiella pneumoniae*, *Streptococcus epidermidis* and *Pseudomonas aeruginosa*<sup>8</sup>.

The alcoholic extract of *P. serratifolia* leaf has hepatoprotective activity against carbon tetrachloride induced hepatotoxicity in rats. According to Selvam *et al*<sup>9</sup>, the methanolic extract of *P. serratifolia* leaf has tumor cell suppression potential in 3 different cancer cell lines - breast cancer (MCF7), liver cancer (Hepg2), and lung cancer (A549) than the standard drug adriamycin.

## 2. Materials and Methods

### 2.1 Determination of Nutrient Content in *P. serratifolia* Leaf

*P. serratifolia* leaf was analysed for the micronutrients like Iron, phosphorus, calcium, ascorbic acid by using the standard procedures.

### 2.2 Processing and Preparation of Plant Extract

Fresh leaves of *P. serratifolia* were collected in the month of October to December from Erode and Karur district of Tamilnadu and used for the study. Cleaned fresh leaves were shade dried for 3-4 days and powdered. By using ethanol the air dried powder was concentrated, evaporated to dryness until semi solid masses was obtained.

### 2.3 Experimental Design for Animal Study

Healthy twenty eight days old albino Wister rats weighing about 100-200 g were used for the study. The rats were divided into five groups consisting of five animals in each group (Table 1).

### 2.4 Body Weight Measurements

The rats in each group were weighed on the initial day, 7<sup>th</sup>, 14<sup>th</sup> and 21<sup>st</sup> day and their weights were recorded.

### 2.5 Biochemical Examination of the Rats

About 1 ml of blood was collected from each rat by Tail vein method at 7 days interval for a period of 21 days. Local anesthesia was applied to the rats before drawing the blood. After collecting the blood sample, silver nitrate ointment solution was applied to stop the bleeding and the blood samples were used for assessing the hematological parameters on the initial day, 7<sup>th</sup>, 14<sup>th</sup> and 21<sup>st</sup> day.

**Table 1.** Animal groups (N=5)

| Animal grouping | Drug treatment        | Dosage treatment                    |
|-----------------|-----------------------|-------------------------------------|
| Group 1         | Normal control        | Standard feed                       |
| Group 2         | Anemic control        | Anemic condition with standard feed |
| Group 3         | Standard control      | Ferrous fumarate<br>0.23 mg/kg      |
| Group 4         | Leaf extract dosage 1 | 100 mg/kg                           |
| Group 5         | Leaf extract dosage 2 | 200 mg/kg                           |

Hemoglobin estimation was done by using Sahli's method and Red Blood Cell count was done by using Neubauer chamber method.

After determining, Hb and RBC, Mean Corpuscular hemoglobin (MCH) was calculated, by using the following equation

$$\text{MCH (Pg)} = [\text{Hb (gm\%)} \times 10] / [\text{RBC (cm.mm)}]$$

## 2.6 Preparation of Value Added Biscuit using *P. serratifolia* Leaf Powder

Proportions of ingredients used in value added biscuit is presented in Table 2.

Mix all ingredients into a dough by adding little amount of water, rolled and then cut into a desired shapes and fried in oil till golden brown colour was obtained.

## 2.7 Preparation of Value Added Rice Vadam using *P. serratifolia* Leaf Powder

The ingredients used in the preparation of value added rice vadam is presented in Table 3.

Grind Green chilly with rice flour and mix well by adding salt and water. Mix the leaf powder into the rice batter at desired proportion and cumin seeds, and then allow it to stand for overnight. Then steam cooked for 10-20 mints, molded into shapes, and sun dried for 1-2 days and then fried in oil.

## 2.8 Preparation of Value Added Murukku by using *P. serratifolia* Leaf Powder

The ingredients used in the preparation of value added Murukku is presented in Table 4.

Mix rice flour and roasted Bengal gram flour by adding salt and sesame seeds.

Make into a dough by adding little amount of water, then shaped by using a mould and fried in oil still golden brown colour.

## 2.9 Organoleptic Evaluation of *P. serratifolia* Leaf Powder Incorporated Products

The formulated recipes were organoleptically evaluated to estimate their acceptability like appearance, flavor, taste,

**Table 2.** Proportion of ingredients used in value added biscuit

| S.No | Ingredients  | Weight In Grams       |                                  |                |
|------|--------------|-----------------------|----------------------------------|----------------|
|      |              | Standard Biscuit<br>S | Leaf Powder Incorporated Biscuit |                |
|      |              |                       | S <sub>1</sub>                   | S <sub>2</sub> |
| 1    | Wheat flour  | 250                   | 250                              | 250            |
| 2    | Ghee         | 2 tsp                 | 2 tsp                            | 2 tsp          |
| 3    | Leaf powder  | -                     | 12.5                             | 25             |
| 4    | Chili powder | as required           | as required                      | as required    |
| 5    | Salt         | as required           | as required                      | as required    |
| 6    | Oil          | to fry                | to fry                           | to fry         |

S – Standard; S<sub>1</sub> – 10 : 0.5 (Wheat flour: *P. serratifolia* leaf powder);

S<sub>2</sub> – 10 : 1 (Wheat flour: *P. serratifolia* leaf powder).

**Table 3.** Proportion of ingredients used in value added rice vadam

| S.No | Ingredients  | Weight In Grams     |                                |             |
|------|--------------|---------------------|--------------------------------|-------------|
|      |              | Standard Vadam<br>S | Leaf Powder Incorporated Vadam |             |
|      |              |                     | S1                             | S2          |
| 1    | Rice flour   | 100                 | 100                            | 100         |
| 2    | Green chilly | 2 No                | 2 No                           | 2 No        |
| 3    | Leaf powder  | -                   | 10                             | 20          |
| 4    | Cumin seeds  | 1 tsp               | 1 tsp                          | 1 tsp       |
| 5    | Salt         | as required         | as required                    | as required |
| 6    | Oil          | to fry              | to fry                         | to fry      |

S – Standard vadam; S<sub>1</sub> – 10 : 1 (Rice flour : *P.serratifolia* leaf powder);

S<sub>2</sub> – 10 : 2 (Rice flour: *P.serratifolia* leaf powder).

**Table 4.** Proportion of ingredients used in value added murukku

| S.No | Ingredients               | Weight In Grams       |                                  |                |
|------|---------------------------|-----------------------|----------------------------------|----------------|
|      |                           | Standard Murukku<br>S | Leaf Powder Incorporated Murukku |                |
|      |                           |                       | S <sub>1</sub>                   | S <sub>2</sub> |
| 1    | Rice flour                | 200                   | 200                              | 200            |
| 2    | Roasted Bengal gram flour | 50                    | 50                               | 50             |
| 3    | Ghee                      | 2 tsp                 | 2 tsp                            | 2 tsp          |
| 4    | Leaf powder               | -                     | 12.5                             | 25             |
| 5    | Cumin seeds               | 1 tsp                 | 1 tsp                            | 1 tsp          |
| 6    | Chili powder              | as required           | as required                      | as required    |
| 7    | Salt                      | as required           | as required                      | as required    |
| 8    | Oil                       | to fry                | to fry                           | to fry         |

S – Standard; S<sub>1</sub> – 10 : 0.5 (Rice flour: *P.serratifolia* leaf powder);

S<sub>2</sub> – 10 : 1 (Rice flour: *P.serratifolia* leaf powder).

texture, colour. Organoleptic evaluation was carried out

by numerical score card method by using 25 semi trained panel members.

## 2.10 Computation of Nutritive Value for Formulated Snacks using *P. serratifolia* Leaf Powder

Comparison of nutrients like iron, phosphorous, calcium, ascorbic acid for the standard and formulated snacks using *P. serratifolia* leaf powder were calculated based on the ICMR requirement (2011).

## 3. Results and Discussion

### 3.1 Determination of Nutrient Content in *P. serratifolia* Leaf

Green leafy vegetables are mostly rich in micro nutrients, hence micronutrients such as iron, phosphorous, calcium and vitamin C were determined in *P. serratifolia* Leaf.

The mean iron content (Table 5) of *P. serratifolia* leaf was  $35 \pm 2.03$ mg/100g which is comparable with the iron content of amaranth (*tristis*) - 38.5mg/100 and cauliflower leaves - 40.0mg/100g. Though the iron content is high in *P. serratifolia* leaf, it remains underutilized due to lack of awareness and lack of popularity.

The mean phosphorus content of *P. serratifolia* leaf was  $34.66 \pm 3.83$ mg/100g and the mean calcium content

**Table 5.** Nutrient content of *P. serratifolia* leaf (100g)

| Nutrients     | Mean $\pm$ SD (mg) |
|---------------|--------------------|
| Iron          | $35.0 \pm 2.03$    |
| Phosphorus    | $34.66 \pm 3.83$   |
| Calcium       | $18.53 \pm 0.32$   |
| Ascorbic Acid | $31.33 \pm 0.97$   |

of *P. serratifolia* leaf was  $18.53 \pm 0.32$ mg/100g. The mean ascorbic acid content of *P. serratifolia* leaf was  $31.33 \pm 0.97$ mg/100g.

### 3.2 Body Weight Changes in Experimental Rats

#### 3.2.1 Effect of *P. serratifolia* Leaf Extract on the Body Weight of Experimental Animals

During 21 days study period, the body weights of selected animal groups were assessed and listed in Table 6.

**Table 5.** Effect of *P. serratifolia* leaf extract on the body weight of experimental animals

| Drug treatment | Dosage                     | Mean $\pm$ SD       |                     |                      |                      |
|----------------|----------------------------|---------------------|---------------------|----------------------|----------------------|
|                |                            | Body weight (g)     |                     |                      |                      |
|                |                            | 1 <sup>st</sup> day | 7 <sup>th</sup> day | 14 <sup>th</sup> day | 21 <sup>st</sup> day |
| G1 (n-5)       | Standard Pellet            | $168 \pm 2.56$      | $173.6 \pm 2.70$    | $178.8 \pm 3.21$     | $183.6 \pm 2.41$     |
| G2 (n-5)       | Standard Pellet            | $149.8 \pm 2.8$     | $148.4 \pm 3.32$    | $147.6 \pm 3.59$     | $148 \pm 2.82$       |
| G3 (n-5)       | Ferrous Fumarate 0.23mg/kg | $149.8 \pm 3.49$    | $140 \pm 2.47$      | $196.4 \pm 3.33$     | $218.8 \pm 2.16$     |
| G4 (n-5)       | Extract 100mg/kg           | $123.2 \pm 2.9$     | $108.8 \pm 2.99$    | $121.2 \pm 4.25$     | $126 \pm 5.95$       |
| G5 (n-5)       | Extract 200mg/kg           | $131.4 \pm 2.68$    | $127.8 \pm 2.8$     | $146.8 \pm 3.34$     | $154.2 \pm 3.37$     |

G1 – Normal control, G2 – Anemic control, G3 – Standard drug control, G4 – Dosage 1, G5 – Dosage 2

It is evident from Table 6 that body weight of G1 gradually increased whereas the body weight of G2 decreased during the study period after phenylhydrazine administration. Due to regular intake of standard drug ferrous fumarate the body weight of the animal showed maximum increase in body weight compared to the other experimental groups. *P. serratifolia* leaf extract administered in two dosages also increased the body weight of the experimental rats. Further, it was noted that the increase in body weight was more in G5 administered animals than G4.

### 3.2.2 Comparison of Body Weight Changes on Experimental Animal Groups

The comparison of body weight changes of animal groups before and after supplementation is given in Table 7.

From the Table 7 it is noted that except G2, all other groups showed significant difference. G3 showed significant difference at 5 per cent level between initial and 7<sup>th</sup>, 14<sup>th</sup> and 21<sup>st</sup> day of supplementation. In the groups G4 and G5, which received extract of *P. serratifolia* in two different dosages showed a significant at 1 per cent level between initial and 7<sup>th</sup>, 14<sup>th</sup> and 21<sup>st</sup> day of the study.

### 3.2.3 Comparison of Body Weight Changes between the Groups

The comparison of body weight changes between the animal grouping are listed in the Table 8.

From the Table 8 it was noted that when G3 supplemented group was compared with G2, statistical analysis showed a significant difference at 1 per cent level from 14<sup>th</sup> day onwards. In G4 and G5 groups supplementation with *P. serratifolia* at two different dosages, both the groups showed significant increase in body weight from 7<sup>th</sup> day onwards.

## 3.3 Hematological Parameters

### 3.3.1 Hemoglobin Level of Experimental Animals

The hemoglobin levels were assessed on the 1<sup>st</sup>, 7<sup>th</sup>, 14<sup>th</sup>, and 21<sup>st</sup> day and the values are shown in the Figure 1.

It is evident that hemoglobin level reduced when phenyl hydrazine was induced on the animal groups except G1. The hemoglobin levels in G1 were maintained at normal range. When standard drug and extract (dosage 1 and

**Table 7.** Comparison of body weight changes in experimental animal groups after supplementation

| Days  | Drug treatment |                    |             |             |             |
|---|----------------|--------------------|-------------|-------------|-------------|
|   | G1<br>(n-5)    | G2<br>(n-5)        | G3<br>(n-5) | G4<br>(n-5) | G5<br>(n-5) |
| Between and 1 <sup>st</sup><br>and 7 <sup>th</sup> day  | 3.58*          | 0.31 <sup>NS</sup> | 3.94*       | 4.17*       | 3.61*       |
| Between and 1 <sup>st</sup><br>and 14 <sup>th</sup> day | 6.63**         | 1.05 <sup>NS</sup> | 4.10*       | 5.46**      | 6.84**      |
| Between and 1 <sup>st</sup><br>and 21 <sup>st</sup> day | 6.70**         | 0.81 <sup>NS</sup> | 4.58*       | 6.43**      | 10.68**     |

G1 – Normal control, G2 – Anemic control, G3 – Standard drug control, G4 – Dosage 1, G5 – Dosage 2

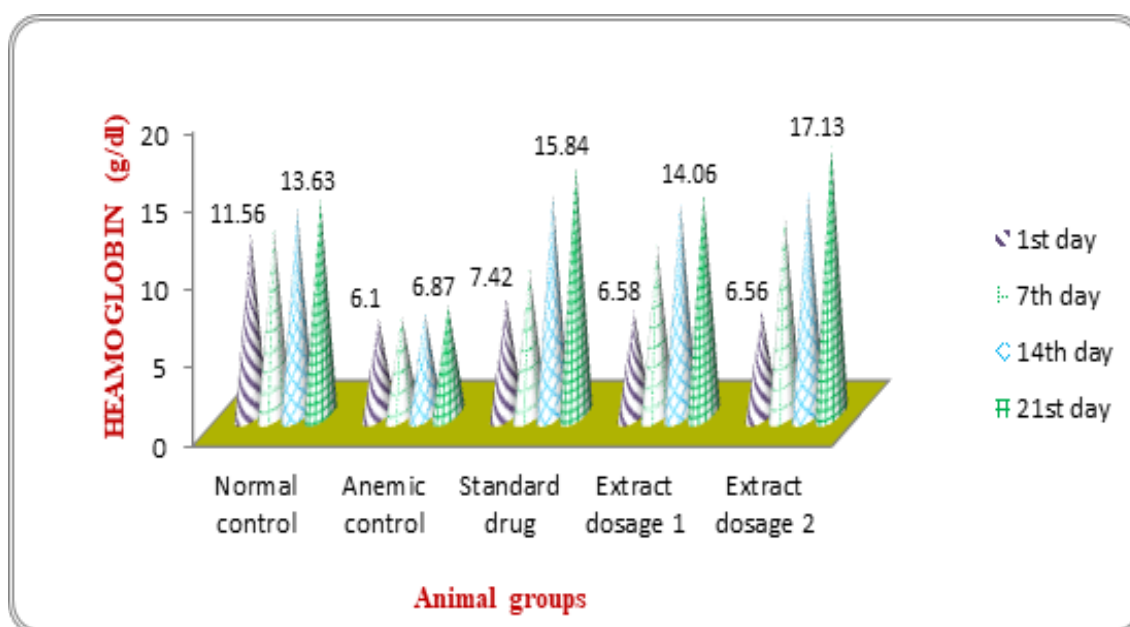
NS – not significant, \* – 5 per cent level significant, \*\* – 1 per cent level significant

**Table 8.** Comparison of body weight between the experimental animals [N=20]

| Animal groups | Days of supplementation |                    |                    |                    |
|---------------|-------------------------|--------------------|--------------------|--------------------|
|               | 1 <sup>st</sup>         | 7 <sup>th</sup>    | 14 <sup>th</sup>   | 21 <sup>st</sup>   |
| G2 vs G3      | 0.01 <sup>NS</sup>      | 0.49 <sup>NS</sup> | 4.95 <sup>**</sup> | 6.45 <sup>**</sup> |
| G2 vs G4      | 1.99 <sup>NS</sup>      | 3.29 <sup>*</sup>  | 3.59 <sup>*</sup>  | 4.17 <sup>*</sup>  |
| G2 vs G5      | 1.49 <sup>NS</sup>      | 3.68 <sup>*</sup>  | 4.06 <sup>*</sup>  | 4.55 <sup>*</sup>  |

G1 – Normal control, G2 – Anemic control, G3 – Standard drug control, G4 – Dosage 1, G5 – Dosage 2

NS – not significant, \* – 5 per cent level significant, \*\* – 1 per cent level significant

**Figure 1.** Effect of *P. serratifolia* leaf extract on Hemoglobin of experimental animals.

2) were given daily there was a gradual increase in the hemoglobin content. G5 showed greater improvement in hemoglobin level followed by G3 and G4. In G2, the hemoglobin content was maintain below the normal range after 21<sup>st</sup> day.

### 3.3.2 Comparison of Haemoglobin in Experimental Animals after Supplementation

It is evident from Table 9 that the hemoglobin levels of G1 and G2 did not show any significant difference between

**Table 9.** Comparison of hemoglobin levels in experimental animals after supplementation

| Days  | Drug treatment     |                    |                    |                     |                     |
|---|--------------------|--------------------|--------------------|---------------------|---------------------|
|   | G1<br>(n-5)        | G2<br>(n-5)        | G3<br>(n-5)        | G4<br>(n-5)         | G5<br>(n-5)         |
| Between and 1 <sup>st</sup><br>and 7 <sup>th</sup> day  | 1.87 <sup>NS</sup> | 2.75 <sup>NS</sup> | 4.23 <sup>*</sup>  | 3.46 <sup>*</sup>   | 4.08 <sup>*</sup>   |
| Between and 1 <sup>st</sup><br>and 14 <sup>th</sup> day | 4.24 <sup>*</sup>  | 2.30 <sup>NS</sup> | 13.36 <sup>*</sup> | 12.44 <sup>**</sup> | 20.70 <sup>**</sup> |
| Between and 1 <sup>st</sup><br>and 21 <sup>st</sup> day | 4.91 <sup>**</sup> | 4.90 <sup>**</sup> | 26.93 <sup>*</sup> | 15.93 <sup>**</sup> | 29.06 <sup>**</sup> |

G1 – Normal control, G2 – Anemic control, G3 – Standard drug control, G4 – Dosage 1, G5 – Dosage 2

NS – not significant, \* – 5 per cent level significant, \*\* – 1 per cent level significant

1<sup>st</sup> and 7<sup>th</sup> day of supplementation, whereas in rats supplemented with ferrous fumarate and *P. serratifolia* in two dosage statistical analysis revealed significant difference at 5 per cent level. Further statistical analysis showed significant difference between initial and 14<sup>th</sup> day in G1 at 5 per cent level and G2 did not show any significant difference between 1<sup>st</sup> and 14<sup>th</sup> day. In rats supplemented with standard drug and *P. serratifolia* in two dosages, significant difference was noted at 1 per cent in all groups between 1<sup>st</sup>

and 14<sup>th</sup> day of supplementation. In all the groups significant difference was noted in haemoglobin between 1<sup>st</sup> and 21<sup>st</sup> day of the supplementation.

### 3.3.3 Red Blood Cell Count of Experimental Animals

Red Blood Cell count was assessed on the initial day and at 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> days interval for a period of 21 days and the

**Table 10.** Effect of *P. serratifolia* Leaf Extract on RBC Levels in Experimental Animals

| Drug treatment | Dosage                        | Mean ± SD                   |                     |                      |                      |
|----------------|-------------------------------|-----------------------------|---------------------|----------------------|----------------------|
|                |                               | Red Blood Cell (cells/cumm) |                     |                      |                      |
|                |                               | 1 <sup>st</sup> day         | 7 <sup>th</sup> day | 14 <sup>th</sup> day | 21 <sup>st</sup> day |
| G1<br>(n-5)    | Standard Pellet               | 6.74 ± 0.26                 | 6.50 ± 0.48         | 6.97 ± 0.22          | 7.62 ± 0.37          |
| G2<br>(n-5)    | Standard Pellet               | 3.07 ± 0.10                 | 3.13 ± 0.04         | 3.21 ± 0.03          | 3.29 ± 0.08          |
| G3<br>(n-5)    | Ferrous Fumarate<br>0.23mg/kg | 3.43 ± 0.45                 | 4.77 ± 0.53         | 6.84 ± 0.29          | 7.89 ± 0.15          |
| G4<br>(n-5)    | Extract 100mg/kg              | 3.51 ± 0.41                 | 5.15 ± 1.04         | 7.39 ± 0.27          | 7.83 ± 0.31          |
| G5<br>(n-5)    | Extract 200mg/kg              | 3.59 ± 0.70                 | 6.95 ± 0.19         | 7.17 ± 0.35          | 8.85 ± 0.19          |

G1 – Normal control, G2 – Anemic control, G3 – Standard drug control, G4 – Dosage 1, G5 – Dosage 2



results are listed in Table 10.

It is evident that G4 and G5 had higher response to retain the normal range of RBC than G3. The regular administration of standard drug and extract in two different dosages increased the red blood cell count gradually and the G5 showed higher response compared with G4 on retaining the normal range of RBC from anemic status. In G2 the increase in RBC was slow but the range was below the normal level even after 21 days of the study period.

### 3.3.4 Comparison of RBC Count of Experimental Animals

The comparison of RBC count of the experimental animals between initial and every 7<sup>th</sup> day interval up to 21 days is given in Table 11.

From the above Table 11 it is evident that G1, G2 and G3 did not show any significant difference between initial and 7<sup>th</sup> day of supplementation, whereas G4 showed significant difference at 5 per cent level and G5 at 1 per cent level. Further comparison of RBC count between 1<sup>st</sup> and 14<sup>th</sup> day revealed significant difference in G1, G3, G4 and G5 statistically, however the significant difference was not evident in G2. The comparison of RBC count between 1<sup>st</sup> day and 21<sup>st</sup> day revealed significant difference in all the groups at 1 per cent level and in G2 at 5 per cent level.

### 3.3.5 Mean Corpuscular Haemoglobin Status of Experimental Animals

On selected animal groups the Mean Corpuscular Hemoglobin values were

calculated by using hemoglobin value and RBC count at 7 day interval for a period of 21 days and the values are listed in Table 12.

It is evident that all the experimental group had normal range of MCH value. In G3, G4 and G5 after inducing anemic, the regular administration of standard drug and leaf extract increased MCH in the 3 groups. The mean values for G3 was higher than G4 and G5 on 21<sup>st</sup> day. There was a gradual increase in MCH values in all the drug treated animals.

### 3.4 Comparison of Overall Acceptability Scores of Standards and Formulated Snacks using *P. serratifolia* Leaf Powder

Table 13 represents the comparison of overall acceptability scores of the formulated snacks using *P. serratifolia* leaf powder.

From the Table 13, it is evident that among the standard snacks, vadam received the highest score followed by stan-

**Table 11.** Comparison of RBC count of experimental animals

| Days  | Drug treatment     |                    |                     |                     |                     |
|---|--------------------|--------------------|---------------------|---------------------|---------------------|
|   | G1<br>(n-5)        | G2<br>(n-5)        | G3<br>(n-5)         | G4<br>(n-5)         | G5<br>(n-5)         |
| Between and 1 <sup>st</sup><br>and 7 <sup>th</sup> day  | 1.96 <sup>NS</sup> | 1.67 <sup>NS</sup> | 1.92 <sup>*</sup>   | 3.32 <sup>*</sup>   | 12.45 <sup>**</sup> |
| Between and 1 <sup>st</sup><br>and 14 <sup>th</sup> day | 4.23 <sup>*</sup>  | 2.60 <sup>NS</sup> | 12.22 <sup>**</sup> | 14.42 <sup>**</sup> | 10.18 <sup>**</sup> |
| Between and 1 <sup>st</sup><br>and 21 <sup>st</sup> day | 7.80 <sup>**</sup> | 4.08 <sup>*</sup>  | 26.11 <sup>*</sup>  | 16.05 <sup>**</sup> | 21.28 <sup>**</sup> |

G1 – Normal control, G2 – Anemic control, G3 – Standard drug control, G4 – Dosage 1, G5 – Dosage 2

NS – not significant, \* – 5 per cent level significant, \*\* – 1 per cent level significant

**Table 12.** Effect of *P. serratifolia* leaf extract on MCH levels in experimental animals

| Drug treatment | Dosage                        | Mean $\pm$ SD                     |                     |                      |                      |
|----------------|-------------------------------|-----------------------------------|---------------------|----------------------|----------------------|
|                |                               | Mean Corpuscular Haemoglobin (pg) |                     |                      |                      |
|                |                               | 1 <sup>st</sup> day               | 7 <sup>th</sup> day | 14 <sup>th</sup> day | 21 <sup>st</sup> day |
| G1<br>(n-5)    | Standard Pellet               | 14.36 $\pm$ 1.07                  | 16.33 $\pm$ 1.16    | 18.06 $\pm$ 0.63     | 18.43 $\pm$ 1.01     |
| G2<br>(n-5)    | Standard Pellet               | 16.39 $\pm$ 1.52                  | 17.72 $\pm$ 1.70    | 18.99 $\pm$ 1.80     | 20.97 $\pm$ 1.15     |
| G3<br>(n-5)    | Ferrous Fumarate<br>0.23mg/kg | 18.44 $\pm$ 3.68                  | 17.36 $\pm$ 1.26    | 19.48 $\pm$ 0.80     | 20.23 $\pm$ 0.47     |
| G4<br>(n-5)    | Extract 100mg/kg              | 15.85 $\pm$ 1.34                  | 17.67 $\pm$ 0.41    | 18.46 $\pm$ 0.73     | 18.97 $\pm$ 2.15     |
| G5<br>(n-5)    | Extract 200mg/kg              | 15.83 $\pm$ 1.14                  | 16.27 $\pm$ 0.73    | 17.55 $\pm$ 0.41     | 19.62 $\pm$ 0.76     |

G1 – Normal control, G2 – Anemic control, G3 – Standard drug control, G4 – Dosage 1, G5 – Dosage 2

**Table 13.** Comparative mean scores for overall acceptability of snacks using *P. serratifolia* leaf powder

| Formulated products | Variations Mean $\pm$ SD |                 |                 |
|---------------------|--------------------------|-----------------|-----------------|
|                     | S                        | S1              | S2              |
| Biscuit             | 4.44 $\pm$ 0.65          | 4.40 $\pm$ 0.49 | 4.32 $\pm$ 0.41 |
| Vadam               | 4.73 $\pm$ 0.41          | 4.56 $\pm$ 0.34 | 4.45 $\pm$ 0.47 |
| Murukku             | 4.65 $\pm$ 0.57          | 4.58 $\pm$ 0.45 | 4.11 $\pm$ 0.69 |

S – Standard

S1 – 10:0.5 (wheat flour :*P.serratifolia*leaf powder)

10:1 (rice flour :*P.serratifolia*leaf powder)

10:0.5 (rice flour :*P.serratifolia*leaf powder)

S2 - 10:1 (wheat flour :*P.serratifolia*leaf powder)

10:2 (rice flour :*P.serratifolia*leaf powder)

10:1 (rice flour :*P.serratifolia*leaf powder)

andard murukku for mean overall acceptability. Further, the overall acceptability scores of S for all formulated snacks received highest scores than S2. The least score for overall acceptability was obtained by S2 murukku. Hence addition of higher proportion of *P. serratifolia* leaf powder affects the overall acceptability scores of the product irrespective of the type of snacks.

## 4. Conclusion

From the above research, it can be concluded that *P. serratifolia* leaf contains iron, phosphorus, calcium and ascorbic acid. The leaf extract of *P. serratifolia* had higher impact of hematological parameters when compared with standard drug ferrous fumarate. Hence, the higher dosage (200mg) of *P. serratifolia* leaf extract has immediate and high response than the lower dosage. By using *P. serratifolia* leaf powder in various snacks could be formulated for easy consumption in day to day life. The higher incorporation of *P. serratifolia* leaf powder helps to increase the nutrient content in formulated snacks.

## 5. References

1. Biradar SS, Biradar SP, Alalagi AC, Wantamutte AS and Malur PR. Prevalence of anemia among adolescent girls, a one year cross sectional study, Journal of Clinical and Diagnostic Research. 2012; 6(3):372-7.
2. Zhang AS and Enns CA. Molecular mechanisms of normal iron homeostasis. Hematology American Society, Hematology Education Program. 2009; 1:207-14.
3. Pandey M, Rohit K, Verma and Shubhini AS. Nutraceuticals new era of medicine and health. Asian Journal of Pharmaceutical and Clinical Research. 2010; 3(1):11-15.
4. Biesalski HK. Nutraceuticals: The link between nutrition and medicine. Kramer K, Hoppe PP, Pocker L, editors. Nutraceuticals in health and diseases prevention New York. Marcel Dekker. 2001; p. 1-26. <https://doi.org/10.1201/9780203908174.ch1>. PMID:11370660
5. Rekha R. Antimicrobial activity of different bark and wood of *Premna serratifolia* Lin. 2010. International Journal of Pharma and Bio Sciences. 2010; 6(1):1-9.
6. Warriar PK, Nambiar VP, Ramankutty C. Indian Medicinal Plants. A compendium of 500 species, India. Orient Longman Publications. 1995; 4:348-52.
7. Kala SMU, Balasubramanian T, Soris T and Mohan VR. GC-MS determination of bioactive components of *Eugenia singampattiana* Bedd. International Journal of ChemTech Research. 2011; 3:1534-7.
8. Singh RC. Antimicrobial effect of callus and natural plant extracts of *Premnaserratifolia* L. International Journal of Pharmaceutical and Biomedical Research. 2011; 2:17-20.
9. Selvam TN, Venkatakrishnan V, Damoder KS, Elumalai P. Antioxidant and tumor cell suppression potential of *Premnaserratifolia* Linn. leaf. Toxicology International. 2012; 19:31-4. <https://doi.org/10.4103/0971-6580.94514>. PMID:22736900 PMCID:PMC3339242