



## **Impact of Pearl Millet Based Supplementary Food on Biochemical and Cognitive Profiles of School Children (5-6 Year Old)**

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### **Authors' contributions**

*This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.*

### **Article Information**

DOI: 10.9734/EJNFS/2020/v12i730245

#### Editor(s):

(1) Dr. Hudson Nyambaka, Kenyatta University, Kenya.

#### Reviewers:

(1) Yuanita Indriasari, Universitas Brawijaya, Indonesia.

(2) A.D.N.T. Kumara, South Eastern University of Sri Lanka, Sri Lanka.

Complete Peer review History: <http://www.sdiarticle4.com/review-history/58498>

**Original Research Article**

**Received 25 April 2020**

**Accepted 01 July 2020**

**Published 07 July 2020**

### **ABSTRACT**

**Aims:** To study the effect of pearl millet based supplementary food on biochemical and cognitive profiles of school going girl children.

**Methodology:** Two groups of 30 girl children in the age group of five to six year each were selected. The total number of children participated in the present study was 60. The first group constituted non supplemented (control); the second group of children (experimental) was supplemented 100 g of pearl millet based supplementary food mix in the form of biscuits along with their home diet for a period of 100 days. The biochemical profiles like haemoglobin, serum protein and serum retinol were analyzed and cognitive test also carried out before and after the administration of supplementary food.

**Results:** The mean increments in biochemical parameters such as haemoglobin, serum protein and serum retinol was higher in experimental group than the control group. At the end of the experiment the mean increment of Raven's score was 1.07 in control and 1.36 in experimental

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group. In Weschler's scale memory score has increased from 72.15 to 72.83 in control and 74.46 to 76.13 in experimental group. Binet Kamat's test of mental ability was also observed an increase in the score at the end of the study period. The haemoglobin level was positively correlated with cognitive development of the children in both control and experimental group.

**Conclusion:** Supplementation with pearl millet based supplementary food mix has improved the biochemical and cognitive development of the selected children. Long term feeding trials with supplementary food mix could improve the cognitive development. Incorporation of the green leafy vegetables powder and carrot powder in different type of foods has positive effect on the cognitive development of school children.

*Keywords: Supplementation; pearl millet; biochemical; children; cognitive.*

## 1. INTRODUCTION

Micronutrient deficiency is a serious public health concern in most of the developing countries. Those which are of greatest public health significance include iron, vitamin A and iodine deficiency disorders. Severe vitamin A deficiency (VAD) can cause eye damage and is the leading cause of childhood blindness. Deficiency of vitamin A, accounts for blindness in 250000 to 500000 children a year according to estimates by the World Health Organization. Globally 127.2 million preschool children are affected by vitamin A deficiency which represents 25 per cent of preschool children in high risk region of the developing world. Forty four per cent of them live in South and South East Asia [1]. Iron deficiency anaemia is also a significant public health problem in India. Anaemia is a serious concern for children because it can impair cognitive development, stunt growth, and increase morbidity from infectious diseases. Overall, 59 percent of children had some degree of anaemia (haemoglobin levels below 11.0 g/dl). Twenty-eight percent of children had mild anaemia, 29 per cent had moderate anaemia, and 2 per cent had severe anaemia [2]. If micronutrient deficiencies are made good it will improve Intelligence Quotient (IQ) by 10-15 per cent; reduce maternal deaths by one third; decrease child mortality by 40 per cent; eliminate nutritional blindness and increase work capacity by 40 per cent. Cognition, or one's ability to perceive, think and remember, is influenced by many factors, one of which is nutrition. Protein Energy Malnutrition (PEM), a multiple nutrient deficiency syndrome, is associated with a variety of cognitive and behavioural deficits [3]. Nutrition is a modifiable factor that can influence cognitive development in children. Supplementary feeding programmes constitute the most promising and effective measures to overcome the nutritional problem among children and improve the quality of their performance in school. So there is an

urgent need to promote healthy weaning practices and the consumption of nutritionally sound, low cost supplementary foods to prevent the development of nutritional deficiency among infants and young children in developing countries. The pearl millet (*Pennisetum glaucum*) is one of the most widely grown millet in India. It is grown in about 7.6 million hectares and yielding 9.1 million tons of grains per year [4]. Pearl millet grain contains starch (61.78%), crude protein (10.96%), fat (5.43%), ash (1.37%) and dietary fiber (11.49%). The calcium, magnesium, phosphorus and iron content of pearl millet are 27.35, 124.00 and 289.00, 6.42 mg, respectively [5]. Hence, the pearl millet is dense in essential nutrients and can be utilized along with pulses and oil seeds for the preparation of supplementary foods. Carrot and green leafy vegetables can be incorporate with the supplementary mix to enrich the  $\beta$ -carotene and iron content of the product. The  $\beta$ - carotene content of carrot is 5423  $\mu\text{g}/100\text{ g}$  [5]. The iron and carotene content of araikeerai are 8.9 mg and 5716  $\mu\text{g}/100\text{ g}$  respectively [6]. The Supplementary foods provide those additional nutrients that are lacking in day to day meals. Hence an attempt was undertaken to study the effect of pearl millet based supplementation on biochemical and cognitive profiles of school children.

## 2. MATERIALS AND METHODS

### 2.1 Procurement of Raw Materials

Pearl millet, Araikeerai (*Amaranthus dubius*), carrot, roasted bengal gram dhal, roasted groundnut and jaggery were purchased in bulk from the local market of Madurai, Tamil Nadu.

### 2.2 Preparation of Supplementary Food Mix

The supplementary food mix was prepared on the basis of a standardized supplementary food,

'Kuzhandai Amudhu' composition [7]. The pearl millet, Bengal gram, ground nut and jiggery were taken in the ratio of 30:20:10:20 and 5.0 per cent of carrot and araikeerai (*Amaranthus dubius*) powder were also incorporated. The flow chart for the preparation of pearl millet based supplementary food mix is given in Fig. 1.

### 2.3 Preparation of Biscuits

Supplementary food mix (100 g), baking powder (0.2 g) and cardamom powder (0.2 g) were mixed together, then sieved twice. Shortening (40 g) and powdered sugar (50 g) were creamed, blended with flour and made into dough. The dough was then rolled, biscuits were cut, baked at 160°C for 15 minutes, cooled and evaluated for its proximate composition.

### 2.4 Physico-Chemical Analysis

Protein, fat,  $\beta$ -carotene, calcium and iron content of supplementary food mix and biscuits were determined by AOAC method [8].

### 2.5 Selection of Study Area

It was planned to conduct the study in semi-rural area. For the present study Loosi Bery Noble special girl's school, at Moonrumavadi in Madurai district, Tamilnadu was selected, since this school served the community characterized by low socio-economic status. The school was located within 2.0 km from MGR (Mattuthavani) bus stand of central Madurai. Before starting the study, relevant informations were collected to confirm that there were no noteworthy

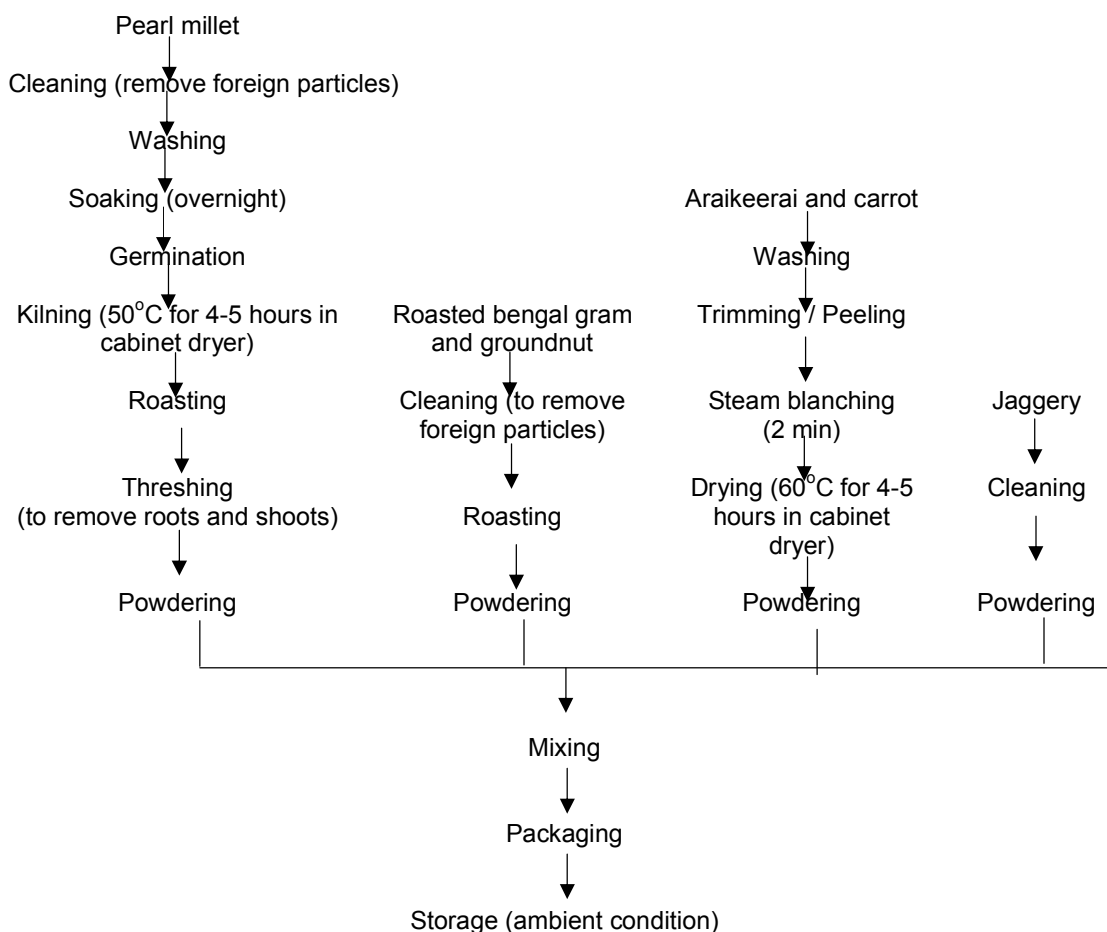


Fig. 1. Flow chart for the preparation of pearl millet based supplementary food mix

differences in characteristics between the children in the selected school or their environment that might cause confounding. The children in selected school represented a socio-economically homogenous population from lower income.

## 2.6 Study Design

To study the impact of the intervention, two groups of 30 girl children in the age group of five to six year each were selected. The total number of children participated in the present study was 60. The first group constituted non supplemented (control); the second group of children (experimental) was supplemented 100 g of pearl millet based supplementary food mix in the form of biscuits along with their home diet for a period of 100 days. The biscuits were distributed twice daily (5 biscuits for each serving) except Saturday and Sunday. To maximize the bioavailability of biscuits it was given without other foods between 11.00 AM and 12.00 Noon and 3.00 and 4.00 PM at a time intervals of two hours after breakfast and two hours before lunch and also two hours after lunch. Both the control and experimental group of children were given mebendazole (500 mg) before the supplementation to treat the worm infestation.

## 2.7 Inclusion and Exclusion Criteria

Inclusion criteria were children of age 5-6 years, those children whose parents were willing to participate in study. Exclusion criteria were children aged less than 5 years and more than 6 years, those Children with congenital diseases, history of metabolic diseases, chronic diseases, physical and mental impairment that could influence their growth. Children who were too agitated and unwilling for anthropometric measurements were excluded from the study.

## 2.8 Biochemical Assay

Subsamples (10 children) were randomly selected from each group (control and experimental) for the assay of biochemical parameters like haemoglobin, Serum protein and Serum retinol [9]. It was estimated before and after the administration of supplementary food to the control and experimental groups.

## 2.9 Classification of Children According to Biochemical Profiles

Children were classified into Normal, Mild, Moderate and severe based on haemoglobin

level [10] and categorized into satisfactory, marginal and deficient according to their Serum retinol level [11].

## 2.10 Cognitive Development Test

An authorized clinical psychologist Dr. Rawlin Chinnan trained the researcher for conducting the cognitive tests. Binet Kamat's test of mental ability, Weschler's scale memory and Raven's coloured progressive matrices tests were used to assess the attention, concentration and memory of the children. These tests were conducted before and after the supplementation of the supplementary food.

## 2.11 Statistical Analysis

The general information of study subjects were quantified, classified, tabulated and expressed in percentages. The paired 't' test was used for pre and post treatments comparison [12]. Correlation was used for determining relationship of selected independent variables with dependent variables [13].

## 3. RESULTS AND DISCUSSION

### 3.1 Chemical Composition of Supplementary Food Mix and Biscuits Prepared from Supplementary Food Mix

The protein, fat, calcium, iron and  $\beta$ -carotene ( $\mu\text{g}$ ) content of pearl millet based supplementary food mix were 15.85 g, 9.85 g, 490.20 mg, 9.13 mg and 8,048  $\mu\text{g}$  per 100 gram and the corresponding value of biscuits prepared from supplementary food mix were 15.71 g, 30.14 g, 490.20 mg, 9.11 mg and 8,021  $\mu\text{g}$ , respectively (Table 1).

### 3.2 Classification of Children According to Haemoglobin Levels

Children were classified according to WHO classification which is given in Table 2. According to these classifications 10.0 per cent of the children in the experimental group fell in normal category ( $\text{Hb} \geq 11.50 \text{ g / dl}$ ) after completion of the study. Before supplementation, in control and experimental group, 50 per cent and 20 per cent were in the mild deficient category and the remaining 50 per cent and 70 per cent were in the moderate deficient category. After supplementation, 60 per cent and 30 per cent of experimental group children improved to mild and moderate category, but no change was observed in the control group. This trend is

indicative of the fact that food source contained other haemopoietic substance beneficial to the improvement in the total nutritional picture of the individual. Chandrasekhar and George [14] reported that the increment in haemoglobin level was 1.2 g in the papaya fed group and 1.4 g in retinol fed group. No significant difference in blood haemoglobin level was noticed in the control group. Vijayalakshmi and Priya [15] reported that the difference in haemoglobin values recorded by the experimental group before and after supplementation was statistically significant at one per cent level. But the difference between initial and final haemoglobin values recorded by the control group was not statistically significant. A Similar trend was observed in the present study also.

### 3.3 Categorization of Children According to Serum Retinol Levels

Serum retinol level of children classified according to Olson (1984) before and after supplementation is presented in Table 3. From

the results it was inferred that none of the children in two groups fell in deficient (<10 µg / dl) category. Eighty per cent and 70 per cent of the children in the control and experimental group before supplementation were in the marginal category and the remaining children in the satisfactory category. After the supplementation, cent per cent of the children in the experimental group was shifted to "satisfactory category". The shift in the grades of the experimental group indicated that continuation of supplementation over a longer period will undoubtedly improve the situation favourably. Similar results were reported by Vuong et al. and Chandrasekhar and George [16,14].

### 3.4 Effect of Supplementation on Biochemical Parameters

The effect of supplementation on biochemical parameters viz., haemoglobin, Serum protein and Serum retinol are given in Table 4.

**Table 1. Chemical composition of supplementary food mix and biscuits**

Nutrients	Supplementary food mix	Biscuits (prepared from supplementary food mix)
Protein (%)	15.85±0.24	15.71±0.39
Fat (%)	9.85±0.17	30.14±0.62
Calcium (mg)	490.20±4.67	490.20±5.34
Iron (mg)	9.13±0.19	9.11±0.27
β-carotene (µg)	8,048±109.51	8,021±49.11

**Table 2. Classification of children according to haemoglobin levels**

Haemoglobin (g / dl)	Control group		Experimental group	
	Initial	Final	Initial	Final
Normal (> 11.5)	-	-	-	1(10.0)
Mild (11.0 – 11.4)	5 (50.0)	5 (50.0)	2 (20.0)	6 (60.0)
Moderate (8.0 – 10.9)	5 (50.0)	5 (50.0)	7 (70.0)	3 (30.0)
Severe (< 8.0)	-	-	1 (10.0)	-
Total	30 (100.0)	30 (100.0)	30 (100.0)	30 (100.0)

*Figures in parentheses represent percentage to the total*

**Table 3. Categorization of children according to serum retinol levels**

Serum retinol (µg/dl) levels	Control group		Experimental group	
	Initial	Final	Initial	Final
Satisfactory (20-50)	2 (20)	2 (20)	3 (30)	10 (100)
Marginal (10-20)	8 (80)	8 (80)	7 (70)	-
Deficient (< 10)	-	-	-	-
Total	10 (100)	10 (100)	10 (100)	10 (100)

*Figures in parentheses represent percentage to the total*

**Table 4. Effect of supplementation on Biochemical parameters of selected children**

Groups	Biochemical parameters			
	Initial	Final	Difference	't' value
<b>Haemoglobin (g/dl)</b>				
Control	9.80 ±1.4751	9.84±01.6728	0.04	0.038063 NS
Experimental	8.98±1.4183	10.00±1.8417	1.02	1.26758 NS
<b>Serum protein (g / dl)</b>				
Control	6.27±1.4751	6.39±1.6728	0.12	0.1693 NS
Experimental	6.13±1.4183	7.96±1.8417	1.83	7.72652**
<b>Serum retinol (g / dl)</b>				
Control	17.26±0.9525	19.72±1.2641	2.46	4.27837**
Experimental	17.20±1.2491	22.72±1.7726	5.45	8.75227**

\*\* Significantly differed at 1 % level and NS- Non significant

### 3.4.1 Increments in haemoglobin (g / dl) levels

The initial haemoglobin levels of control and experimental group were 9.80 and 8.98 g / dl respectively. The mean haemoglobin level of control group at the initial was higher than the experimental group. At the end of the study period, the haemoglobin level of the control (9.80 g / dl) was slightly increased to 9.84 g / dl and a difference of only 0.04 g / dl. However, the mean increment in the haemoglobin level (1.02 g / dl) was registered by the experimental group which could be attributed for supplementation of iron by green leafy vegetables. After the supplementation, the mean haemoglobin level was increased to 0.04 g / dl in control and 1.02 g / dl in the experimental group. This result agreed with [17] who reported that a total of 119 children showed a rise in haemoglobin levels after haematinic supplementation. The mean rise in haemoglobin was 0.64 g / dl in 91 anaemic children and 0.08 g / dl in 68 non-anaemic children. The rise was significant ( $P < 0.01$ ; paired test) after supplementation. Another study conducted by [18] reported that iron supplementation had a positive effect on the haemoglobin level and the difference in the haemoglobin levels shows a 5.0 per cent level of significance. The effect of iron supplementation was more pronounced in children with initial lower haemoglobin values. Bagyalakshmi and Vijayalakshmi [19] reported the impact of ICDS on the health status of children. The mean haemoglobin value recorded at the beginning of the study for the children was 7.8 g / dl. At the end of the year, it was found to increase to 9.8 g / dl. Effect of supplementation of Multiple Micronutrient Fortified Candy (MMFC) was assessed for haemoglobin levels of school children (3-6 yrs) in urban slums of Vadodara. Similar situation was observed in the present investigation too.

### 3.4.2 Increments in serum protein levels

The mean serum protein level of both the control and experimental group was found to be lesser than the standard value of 8.0 g / dl. After the supplementation there was an appreciable increase in serum protein of all the children in the experimental group. The increase in serum protein was 0.12 and 1.83 g / dl for control and experimental group respectively.

### 3.4.3 Increments in serum retinol levels

Initially the mean serum retinol among the children in the control and experimental group was 17.26 and 17.20  $\mu\text{g} / 100 \text{ ml}$  respectively. After supplementation, the level was increased to 19.72 and 22.72  $\mu\text{g} / 100 \text{ ml}$ . From the study reports the increase in serum retinol levels of the experimental group was higher than the control group. This shows the effectiveness of  $\beta$ -carotene rich food in combating vitamin A deficiency. Devadas and Saroja [20] reported an increment of serum retinol levels in 2½ to 6 years old children from 11.4 to 24.7 and 12.3 to 25.5 after 3 months of supplementation with papaya and amaranthus [5]. A similar trend was noted in the present investigation too. Mercado et al. [21] reported an increase in serum retinol levels from 0.68 to 1.06  $\mu\text{mol} / \text{L}$  after supplementation for 12 weeks [8]. Another study conducted by Ramalakshmi et al. [22] studied the impact of short term food supplementation of school children with special reference to vitamin A and haemoglobin. The results of the study indicated that serum vitamin A and haemoglobin status showed significant improvement after the supplementation. Aggarwal and Khanna [23] studied the impact of Leaf Powder Concentrate (LPC) supplementation on vitamin A and iron status of children aged one to three years. Six months of supplementary feeding led to

significant improvement in the vitamin A status of preschoolers more effectively than non-LPC fortified supplements. About 400 rural school children (6-12 years) from villages of Udaipur, Southern part of Rajasthan were screened by clinical examination for their vitamin A and iron deficiency, were included for intervention study conducted by pre-post, control-experimental design. Each child was fed one serving of biscuit (providing 8.62 mg of iron and 835.3  $\mu\text{g}$  of  $\beta$ -carotene) per day for 100 days. Study revealed that serum vitamin A and haemoglobin level increased significantly among children of experimental group as compared to control group after intervention ( $P < 0.05$ ) [24].

### 3.5 Effect of Supplementation on the Cognitive Tests

Table 5 shows the mean increment in cognitive scores. At the end of the experiment, the mean increment of Raven's score was 1.07 in control and 1.36 in the experimental group. In Weschler's scale memory score was increased from 72.15 to 72.83 in control and 74.46 to 76.13 in the experimental group. Binet Kamat's test of mental ability was also observed that an increase in the score at the end of the study period. Results showed that the mean increments in cognitive development were higher in the experimental group than the control group. Even though the mean increments in the cognitive development of both the group was statistically significant. This might be due to the environmental factors such as demographic, socio-economic, health, social, behavioural and motivational influences or genetic influences. The statistical analysis revealed that the children who received supplementary food showed a significant difference in cognitive tests. Results of the study showed that the supplementation had positive effects on children's cognitive performance. The study conducted by a researcher [25] revealed that children who received the supplement with meat showed significantly greater gains on Raven's progressive matrices than all other groups. Results of this study suggested that supplementation with animal source food has a positive effect on Kenyan children's cognitive performance. Bruner et al. [26] showed that girls who received iron performed better on a test of verbal learning and memory than girls in the control group. Black et al. [27] observed that the iron and zinc administered together and with other micronutrients had a beneficial effect on infant motor development. Sanstead et al. [28]

stated that neuropsychologic performance and growth were most improved after treatment with zinc with micronutrients. Chandrasekhar and Rani observed that the majority of children (97 to 100%) in group I given 62 g of soy protein isolate (SPI) based food mix showed good cognition concerning all the criteria studied, except stringing beads, folding paper horizontally and building tower [29]. Suboticanec et al. confirmed that effect of iron supplementation on cognitive function by a double blind intervention trial in nine year old mildly anaemic school children. Iron supplementation resulted in a statistically significant improvement in total Weschler's Intelligent Scale for Children - Revised (WISC-R) score [30].

### 3.6 Correlation between the Haemoglobin Level and Cognitive Development of the Selected Children

The correlation between the haemoglobin and cognitive development of the selected children was 0.794257, 0.961322 and 0.761329 in control and 0.921768, 0.932279 and 0.946939 in the experimental group (Table 6). The haemoglobin level and cognitive development were positively correlated. Significant associations with haemoglobin and cognitive developments were found in the selected children in both the control and experimental group. Based on the results, it was concluded that iron deficiency leads to cognitive impairment. The improvement in haemoglobin status helps to improve cognitive development. A study of Egyptian children 8 to 11 years old found that those with iron deficiency anaemia made more errors and were less efficient in the "Matching Familiar Figure Test" than non-anaemic children. Moreover, treatment with 50 mg of oral ferrous sulphate (2.0 mg elemental iron) reduced errors and improved efficiency scores [31]. Another study conducted by [32] examined the association between early childhood anaemia and mild or moderate mental retardation at 10 year of age. Results showed an increased likelihood of mild or moderate mental retardation associated with anaemia. The findings of the present study are in line with that of [32] who studied the effect of iron supplementation and anthelmintic treatment on iron status, anaemia and development of children aged 6-59 months. Results indicated that the iron supplementation improved motor and language development of preschool children in rural Africa. The effects of iron on motor development were limited to children with more severe anaemia (baseline haemoglobin concentration  $< 90 \text{ g / L}$ ).

**Table 5. Mean increments of the cognitive test scores in selected children**

Cognitive tests	Mean scores							
	Control group				Experimental group			
	Initial	Final	Difference	't' value	Initial	Final	Difference	't' value
Raven's coloured progressive matrices	12.06 ± 4.3710	13.13 ± 3.6030	1.07	6.72817**	13.8 ± 4.6360	15.16 ± 3.6339	1.36	6.45767**
Weschler's scale memory	72.15 ± 2.8523	72.83 ± 2.7123	0.68	8.80354**	74.46 ± 3.9517	76.13 ± 3.8534	1.67	11.2288**
Binet Kamat's test of mental ability	80.4 ± 6.1885	82.06 ± 6.3285	1.10	12.7778**	82.01 ± 6.3732	84.36 ± 6.1680	2.35	17.2135**

*\*\*Significantly differed at 1 % level*



**Table 6. Correlation between the haemoglobin level and cognitive development of the selected children**

Cognitive tests	Haemoglobin levels	
	Control group	Experimental group
Raven's colour progressive matrices	0.794257**	0.921768**
Weschler's scale memory	0.961322**	0.932279**
Binet Kamat's test of mental ability	0.761329*	0.946939**

\*\*Significantly differed at 1 % level and \*significantly differed at 5 % level

#### 4. CONCLUSION

The pearl millet based supplementary food is rich in protein, calcium, iron and  $\beta$ -carotene. The supplementary food (100 g) met the 78, 81.70, and 70 per cent of the recommended dietary allowance for protein, calcium and iron content respectively of the selected children in the age group of 5 to 6 years. Approximately 40 g of the supplementary food is enough to fulfill the recommended dietary allowance for  $\beta$ -carotene of the children. The iron play a vital role in the formation of haemoglobin and prevents anaemia. The supplementation with pearl millet based supplementary food mix has improved the biochemical profiles such haemoglobin, serum protein and serum retinol level of the children. The haemoglobin level and cognitive development of the children are positively correlated. So the supplementation improved the cognitive profiles like attention, concentration memory and mental ability of the children. Hence, the study concluded that incorporation of the green leafy vegetable powder and carrot powder in a supplementary food mix enrich the  $\beta$ -carotene and iron content of the product and have a positive effect on the cognitive development among the school children.

#### CONSENT

As per international standard, parental written consent has been collected and preserved by the author(s).

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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