Formulation of a herbal extract for anemia treatment and its effect on physical work and intelligence capacity in adolescent girls with iron deficiency in India

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Iron deficiency anemia poses a burden on the limited health care delivery system in India. Current pharmacological anti anemic agents cause undesirable side effect such as nausea, vomiting and dark stool. Herbal extract as a nutritional support therapy offer the potential for alternative treatment strategies. In this context, the goal of this study was to provide plant extract formulation for anemia treatment in adolescent girls in India. The herbal extract was prepared by slicing 500 mg of fresh amla or embelica officinals into small pieces and blended well and mixed with quarter litre of boiled and cold water and drain with sieve. Two medium sized pumpkin leaves or Cucurbita pepo leaves was blended well with 50 ml of water and the juice was drain out. Jaggery syrup was prepared by melting 1000 g jaggery in 250 ml of water. The herbal extract was prepared by adding amla juice (500 mL) and pumpkin leaves extract (50 mL) to the jaggery syrup. It was administered to the adolescent girls 30 mL per day prior to lunch for duration of 60 days with the help of an ounce glass. There was significant difference observed in the post test level of haemoglobin and physical work capacity between the control and experimental group after receiving the herbal extract. But it was found that only mild difference was observed in intelligence between the control and experimental group after receiving the herbal extract for sixty days. Combining the three extracts enhanced the level of haemoglobin which further improved the physical work capacity and intelligence. The formulation demonstrates more potential in combinational herbal medicines therapy in anemia management.

Key words: Herbal extract, jaggery, pumpkin leaves, amla.

INTRODUCTION

Iron is an essential element for blood production. It is used for formation of hemoglobin, oxygen transport, brain development, regulation of body temperature and muscle activity. The most severe consequence of iron depletion is iron deficiency anemia (IDA). Iron deficiency anemia is considered the most common nutritional deficiency worldwide. It generally results when the iron demands by the body are not supplied by iron absorption (Ekwagwu et
Individuals with IDA have inadequate intake, impaired absorption or transport, physiologic losses associated with chronological or reproductive age, or chronic blood loss secondary to disease. Accelerated development, hormonal changes, unhealthy food and starting of menstruation in girls are major causes of iron deficiency anemia in adolescence period. During this period, physical changes affect the nutritional needs from the body while changes in lifestyle may affect eating habits and food choices. In this context, adolescent nutrition is therefore important for supporting the physical growth of the body and for preventing future health problems (Gaja, 2011). Anemia is a major health problem throughout the world with an annual prevalence of 400 million. World Health Organization (WHO) stated in 2011 that in all South-East Asian countries, except Thailand, more than a quarter of girls are anemic. In the year 2015, WHO reported that the prevalence of anemia is attributed to iron deficiency estimating that the proportion of all anaemia amenable to iron was about 50% in women and 42% in children (National Health Mission and National Rural Mission, 2015). In India, the high prevalence rate of anaemia occurs in adolescent girls. National Rural Health Mission of India (2013) stated that a greater number of adolescent girls between 15-17 years suffer from anemia and its prevalence in India is high in rural community areas (WHO, 2011). According to WHO estimates, India is one of the countries in the world that has highest prevalence of anemia. The prevalence of anemia in Karnataka shows that 42.4% of women suffer from anemia, 26.7% with mild anemia, and 13.4% with severe anemia (Singh et al., 2013).

Hemoglobin (Hb) level is directly proportional to physical work capacity. In the case of decreased concentration of Hb, oxygen transport to the tissues becomes a hindrance, resulting in decrease in physical work capacity (Urmila et al., 2016). Iron deficiency leads to a reduction in neurotransmitter levels, impaired transmitter function, hypomyelination and delayed neuromaturation, or attributed to the reduction of oxygen delivery to the brain (Pavod, 2011). In India, there are National guidelines and standards of care for anemia in adolescents. However, the practice remains less satisfactory, which might partly be due to diverse religions, food habits, lifestyles, languages, cultures, and traditions (Rani, 2014). Possible side effect such as epigastric discomfort, nausea, diarrhea, or constipation may be attributed to a daily dose of iron at 60 mg or more.

The present study is based on community resources, since amla, jaggery and pumpkin leaves are locally available, cost effective can be easily stored and utilized by community people. Supplementation of locally available foods helps to reduce the prevalence of anemia at lower cost and useful to the community for combating anemia (Sukhdeep, 2016). Researcher shown that just 8.7 mg of natural vitamin C from amla is equivalent to 100 mg of synthetic vitamin C. Moreover, amla is an exception among fruits not only because of its high vitamin C content, but also because it contains substances which partially protect the vitamin from destruction on heating or drying (Mathur, 2005). Jaggery is a traditional Indian sweetener made from sugar cane. It has a characteristic dark color due to the presence of high iron content. It is an excellent source of iron and regular consumption of jaggery can help to improve the hemoglobin level. 100 g of raw jaggery contains 3 mg iron. Research studies have revealed that pumpkin leaves juice was effective in increasing the regeneration of hemoglobin. It is rich with vitamin C and Iron. 100 g of pumpkin leaves contains 18% of vitamin C and 12% of iron.

**MATERIALS AND METHODS**

**Plant collection**

The Amla and pumpkin leaves leaves were collected from Magadi village and jaggery was purchased at a local market in Maddur.

**Preparation of plant material**

The first step consisted of the preparation of amla aqueous extract. It was prepared by slicing 500 mg of fresh amla into small pieces (removal of the seeds) and blended well. This pulp was mixed with quarter liter of boiled and cooled water and a sieve was used to drain it. The second step was two medium sized pumpkin leaves blended well with 50 mL of water and the juice was drained out. The third step consisted of jaggery syrup preparation by melting 1000 mg jaggery in 250 mL of water. The Fourth step: the herbal extract (a nutritional support therapy) was prepared by adding the Amla juice (250 mL), pumpkin leaves extract (50 ml) to the jaggery syrup. The formulation was administered to the adolescent girls, 30 ml per day prior to lunch for 60 days with the aid of an ounce glass.

**Standardization of the herbal extract**

The herbal mixture was subjected to nutrient analysis. The extract has prepared as per the guidance of dieticians and ayurvedic doctor before supplementing the extract to adolescent girls. The result of the analysis showed nil alcohol content, 40% vitamin C and the level of Fe, 101.27 mg/kg.

**Participants**

An experimental design with pre-test and post test control group was used for the study. This study was conducted with adolescent girls after getting Institutional Human Ethics Committee of Saveetha University (011/01/2015/IEC/SU Dated 20/01/2015). Informed consent and assent was obtained from the adolescent girls and from their parents. The total sample consists of 120 adolescent girls between the age of 14-17, years studying at selected higher secondary school. The sixty participants from Sri Jagannadakaval Public School, Bangalore, India was taken as the control group and sixty participants from Gangothri Public School, Bangalore India were as the to experimental group.
The screening test was conducted in two steps.

(1) Check list: Observational check list was used to assess the signs and symptoms of iron deficiency anemia. It consists of 39 items to find out the presence or absence of major signs and symptoms of iron deficiency anemia. It was used to identify the adolescent girls with iron deficiency anemia and also degree of iron deficiency anemia. The degree of iron deficiency anemia was interpreted as mild (1-13), moderate (14-26) and severe (27-39).

(2) Confirmatory diagnostic test using Sahlis method: The participants who were identified by prior checklist were subjected to Sahlis method to estimate the level of hemoglobin in the blood. The sahlis hemoglobinometer was used to estimate the hemoglobin content in blood from the fingertip blood. The reading is interpreted as normal (≥12 g/dL), mild (9-11.9 g/dL), moderate (7.1-8.9 g/dL) and severe (<7 g/dL). This method was used as confirmatory diagnostic investigation for the selection of participants for the study. Adolescent girls with mild and moderate iron deficiency anemia were taken for the study.

**Phase II**

The structured questionnaire was used to collect demographic and clinical proforma. The participants who were identified by the checklist and Sahlis method were subjected to blood test analysis. The procedure of the blood test was explained to the participant. The participants were helped to assume comfortable position. The tourniquet was wrapped around the upper arm. The needle site was cleaned with spirit. The needle was inserted into the vein. 2 ml of blood was drawn. The tourniquet was removed from the hand. A gauze pad or cotton ball was put over the needle site. Pressure was applied to the site and the bandage was applied. 2 mL of blood was collected and transferred to ethylene diamino tetra acetic acid, anticoagulant (EDTA) vacutainer. The blood hemoglobin estimation was conducted in scan point diagnostic centre (NABL accredited laboratory) at Bengaluru. The level of hemoglobin was interpreted as normal (≥12 g/dL), mild (9-11.9 g/dL), moderate (7.1-8.9 g/dL) and severe (<7 g/dL).

**Phase III**

Culture fair intelligence non verbal test: It was used to assess the intelligence level of adolescent girls (Rao et al., 2013). The test has 4 sub tests (Test 1 series with 12 items, Test 2 Classification with 14 items, Test 3 matrices with 12 items, Test 4 conditions with 8 items). Total items is 46. The row scores was compared with the score given in the scoring table. Level of intelligence was interpreted as very superior (130 and above), superior (120-129), high average (110-119), average (90-109), low average (80-89), borderline (70-79) and extremely low (69 and below).

**Phase IV**

Harvard step test was used to assess physical work capacity among adolescent girls in the control and experimental groups. It was developed by Brouha et al. (1943) It was used to measure physical fitness and a person's ability to recover after a strenuous exercise. The test computes the capability to exercises continuously for extended interval of time without tiring. In this study, the researcher created stool at height of 33 cm and width 38 cm. The subjects were advised to step up and down on the modified Harvard steps of 33 cm height once every two seconds approximately 30 step up per minute (30/min) for 5 min. Pulse has recorded 1st, 3rd and 5th minute during the test. Was kept as PR1, PR2 and PR3 which means PR1 (pulse rate 1), 1 min after exercise, PR2 (pulse rate 2), 3 min after exercise, PR3 (pulse rate 3), 5 min after exercise, from the above calculation, physical fitness index was computed as

\[ PFI = \text{Duration of exercise in seconds} \times \frac{100}{2} (\text{Pulse 1+2+3}) \]

The physical fitness index score was interpreted as excellent (>96), good (83-96), average (68-82), low average (54-67) and poor (<54).

**Phase V**

Herbal extract was administered during the short interval break between 10.30-10.45 am for 60 days from Monday to Saturday.

**Phase VI**

After 60 days, post-test was including blood test to estimate Hb, Harvard step test and culture fair intelligence non verbal test were carried out with the same procedure.

**Statistics**

Non-parametric test (Wilcoxon-signed rank test and Mann-Whitney U-test) was used to find out effectiveness of intervention on physical work capacity and intelligence.

**RESULTS**

**Effect of herbal extract on level of haemoglobin**

Table 1 represents the comparison of pre test and post
test level of haemoglobin of the studied group. The comparison of pre and posttest level of haemoglobin within the control group was done using paired ‘t’ test and 0.469 (p=0.641) was obtained. The comparison of experimental pre and post test level of haemoglobin using paired ‘t’ test value was 3.96 (p<0.001). In the experimental group, there was a rise in mean value from 9.942 to 10.99 which shows that the herbal extract was effective in improving the level of haemoglobin among the adolescent girls. The comparison of pretest level of haemoglobin of the control and experimental groups using unpaired ‘t’ test value was 0.02 (p=0.984). The comparison of post test level of haemoglobin of the control and experimental groups using unpaired ‘t’ test value was 4.56 (p<0.001). Hence, there was significant difference observed in the post test level of haemoglobin between the control and experimental groups after receiving herbal extract.

**Effect of herbal extract on level of intelligence**

The comparison of pre and post test level of intelligence within the control group was done using Wilcoxon-signed rank test and value 0.352 (p=0.725) was obtained. The comparison of experimental pre and post test level of intelligence using Wilcoxon-signed rank test was done, value 3.65 (p=0.003) was obtained. Experimental group had mean value of 97 in pretest and 99 in posttest. The comparison of pre test level of intelligence of control group and experimental group using Mann-Whitney ‘u’ test value was 0.571 (p=0.568), there was no significant difference observed between the pre test level of intelligence of the control and experimental group. The comparison of the post test level of intelligence of the control group and experimental group using Mann-Whitney ‘u’ test showed 2.433 (p=0.015) (Table 2).

**DISCUSSION**

In the present study, there was a difference in the mean level of haemoglobin between the pre and post test among experimental group and the mean difference observed was t = 3.96 (p<0.001). And also, posttest means score of experimental group was higher than the control group after the intervention. It shows that the level of hemoglobin in the blood increased after the administration of herbal extract for 60 days. A similar study was conducted on impact of leaf concentrate and iron folic acid supplementation on blood profile of anemic adolescent girls. The study found that a statistically significant improvement had taken place in haemoglobin level, as well as other blood parameters (Vijayalakshmi and Selvasundari, 1983).

The result of the present study revealed that intervention was very effective in improving the physical work capacity. In the experimental group, there was rise in median value from 72 to 97. There was a difference in the level of physical work capacity between the pre and posttest among experimental group. The post test level of physical work capacity was higher in experimental group than the control group after the intervention. It shows that level of physical work capacity improved after the herbal intervention for sixty days. This study is consistent with study conducted on young adult women to find relationship between iron deficiency anemia and energy expenditure found that iron deficiency anemia has an effect on the physical work capacity by reducing the availability of oxygen to the tissues and interfering with work efficiency Gardner et al., 1977). The effect of iron supplementation on physical work performance in children and adolescents was reported. The physical work performance outcomes studied were heart rate, treadmill endurance times blood lactate and oxygen consumption. The study concluded that iron supplementation has positive effect on physical performance of children and adolescents (Gardner et al., 1977).

In the present study and the experimental group, there was rise in mean value (97) in pre test and 99 in posttest, this indicated that intervention of the experimental group was effective in enhancing the level of intelligence. There was only mild difference in the level of intelligence between the pretest and posttest among experimental group. So, the study proved that there was no much

**Table 2. Effectiveness of herbal extract on level of intelligence.**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group</th>
<th>Median (Q3-Q1)</th>
<th>Wilcoxon –signed rank test</th>
<th>Mann-Whitney ‘U’-test</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control pre test</td>
<td>97(100.5-92)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Control post test</td>
<td>97(102.5-92.5)</td>
<td>Z=0.352</td>
<td>Z=0.571</td>
</tr>
<tr>
<td></td>
<td>Experimental pre test</td>
<td>97(100-94)</td>
<td>p=0.725</td>
<td>P=0.568</td>
</tr>
<tr>
<td></td>
<td>Experimental post test</td>
<td>99(112-94)</td>
<td></td>
<td>Z=2.433</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>P=0.015</td>
</tr>
</tbody>
</table>
improvement in the intelligence after the administration of intervention for sixty days. IQ is influenced by various factors like heredity, nutrition, education, drugs, stress, peer group etc and cannot suggest any one factor for the improving the level of intelligence (Sen and Kannani, 2006). It was in tune with a study conducted on micronutrient status, cognition and behaviour problems in childhood showing the association between iron status and intellectual ability or scholastic performance. Iron supplementation appeared to improve mental development score in adolescents (Sachdev et al., 2005).

Conclusion

This study was carried out to determine the anti-anemic effect of herbal extract prepared from amla, pumpkin leaves and jaggery. The plant extract medicine had maximum response in enhancing the level of haemoglobin which further improved the physical work capacity and intelligence. The formulate product can be standardised and used as a potential anti anemic medicine.

CONFLICT OF INTERESTS

The authors declare that there is no conflict of interest.

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REFERENCES