Effect of *Opuntia elatior* Mill. (*Nagaphani*) in the management of *Pandu Roga* w. s. r to iron deficiency anemia; an open labeled randomized standard controlled clinical trial

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Abstract

Background: *Pandu Roga* described in *Ayurveda* bears resemblance to the clinical features of anemia. The fruits of *Nagaphani* (*Opuntia elatior* Mill.) of family *cactaceae*, are consumed by the inhabitants of Gujarat as a hematinic agent to treat general debility and anemia. **Aim and objective:** To evaluate the efficacy of the *O. elatior* fruit juice in *Pandu Roga* with special reference to iron deficiency anemia. **Materials and method:** Present clinical study was an open labeled, randomized trial, in which 105 patients of *Pandu Roga* (iron deficiency anemia), were divided into two groups using the coin method of randomization. Group A received *Opuntia elatior* fruit juice, 20 ml twice a day with lukewarm water before meal. Group B received *Punarnava Mandura* (standard control)*,* well accepted after this a herbo-mineral fromulation, two tablets of 500 mg each, twice a day with *Takra* (butter milk) before meal. Duration of intervention was 2 months in both groups and a follow-up of 1 month. The assessment was done based on the clinical signs and symptoms and hematological parameters such as Hb% (hemoglobin %), total red blood cell (TRBC), serum iron, serum ferritin and total iron-binding capacity (TIBC), before and after completion of the treatment. The data were subjected to statistical tests (Wilcoxon signed-rank test and paired *t*-tests, co-efficient of variance (CV)), and analyzed. **Results:** Both trial drug and the standard drug produced a statistically significant increase in Hb%, TRBC and serum Iron. The standard drug provided statistically significant increase ($P < 0.05$) in TIBC, whereas a statistically insignificant ($P > 0.05$) effect was observed in the trial group. Both trial and standard drug provided statistically significant ($P < 0.001$) relief in *Panduta* (pallor), *Daurbalya* (general weakness), *Akshikutashotha* (periorbital edema) and *Pindikodveshthana* (calf muscle cramps). When compared between the groups, the standard drug was found to be more effective than the trial drug. **Conclusion:** Fruit juice of *Opuntia elatior* is effective in iron deficiency anemia as it increases the hemoglobin, TRBC levels in blood along with providing relief in *Panduta* (pallor), *Daurbalya* (general weakness) *Akshikutashotha* (periorbital edema).

Keywords: Anemia, cactus pear, hathlo-thore, *Nagaphani*, *opuntia elatior*, *panduroga*, prickly pears, *Punarnava Mandura*

Introduction

The World Health Organization estimates that 1.62 billion people globally are affected with anemia which corresponds to 24.8% of the world population.[1] In India, anemia affects an estimated 50% of the population.[1] *Ayurveda* delineates *Pandu Roga* as a disease entity developing due to depletion of *Rasadhatu* (plasma)*,* which in turn results in deficient production of *Raktadhatu* (blood tissue). *Pandu Roga* may occur as a prime disease or as a complication of other disease or as a sign of a disease.[2] It has resemblance with the clinical picture of anemia.[3] It is the most predominant blood cell deficiency disorder and a global public health problem affecting both developing and developed countries

with major consequences for human health as well as social and economic development.[4] According to the WHO, anemia is the condition in which the hemoglobin content of blood is lower than normal as a result of deficiency of one or more essential nutrients.[5] In the conventional system of medicine,

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iron and folic acid supplements are used in the treatment of anemia, which usually produces gastritis and other side effects such as nausea, constipation, and diarrhoea which further deteriorates the health of the patient.[6] There is an urgent need to find out herbal drugs which are easily available, effective, and low in cost to manage this disease condition. *Ayurveda* describes both single as well as compound formulations for the management of *Pandu Roga. Punarnava Mandura*,[7] one of the most regularly prescribed drugs for the management of *Pandu Roga*, has been established through various research works,[8-10] for its effectiveness in managing the clinical conditions of *Pandu Roga*.

Opuntia elatior Mill.(*cactaceae*), a folklore plant known as *Nagaphani* or *Hathlo-thore* is available in many parts of India and widely prevalent in the state of Gujarat.[11] Fruits of *Opuntia elatior* are being used by the local people of Gujarat, to combat anemia and general debility, irrespective of age. The drug has been found safe through acute and chronic toxicity studies,[12] and effective in managing anemia through experimental studies on animals.[13,14] Further the drug has also been found effective in managing *Pandu Roga* (anemia) in the geriatric population.[15] It is pivotal to validate this traditional practice, scientifically, to treat *Pandu Roga*. In the present clinical research, *Opuntia elatior* fruit juice has been assessed for its efficacy in the management of *Pandu Roga* (iron deficiency anemia) in comparison to *Punarnava Mandura,* the standard control drug.

Materials and methods

The study, conducted from Feb 2015 to March 2016, got approved by Institutional Ethics Committee (No. PGT/7/-A/ethics/2014-15/2652; dated December 19, 2014) and was registered in the Clinical Trials Registry of India (CTRI/2015/02/005547; dated February 16, 2015). Patients visiting the outpatient department and inpatient department, Institute for Post-Graduate Teaching and Research in Ayurveda (IPGT and RA), Jamnagar, were thoroughly examined for clinical signs and symptoms of *Pandu Roga* along with the necessary hematological and biochemical investigations. Subjects were enrolled for the study considering the criteria of inclusion, after getting their full consent. The registered patients were allocated into two groups, using the coin method of randomization.[16] Group A received *Opuntia elatior* fruit juice, group B received *Punarnava Mandura* tablets for 2 months. Follow-up was done for a period of 1 month. Patients were advised to stop other ongoing medications which were prescribed for the treatment of iron deficiency anemia and informed to reduce intake of food which are sour, salty, *Vidahi* (which causes burning sensation), *Vishthambhi* (which causes constipation), and *Abhishyandikara Ahara* (which blocks the channels). Assessment of clinical signs and symptoms was done before, during, and after treatment, and after follow-up period of 2 months. Hematological, biochemical, and routine urine examinations were repeated after the completion of 2 months' intervention.

Diagnostic criteria

Patients were diagnosed based on clinical signs and symptoms of *Pandu Roga* and levels of hematological parameters like Hb% (<12 g% in females and <13 g% in males), TRBC (<4.2 mill/c. mm), serum iron (<37 μg/dL), serum ferritin (>73 μg/dL), and total iron-binding capacity (TIBC) (>250 μg/dL).

Inclusion criteria

Patients between the age of 18–60 years, irrespective of gender having Hb% below 12 g (for females) and below 13 g (for males) or decreased RBC count (<4.2 mill/c. mm), serum iron (<37 μg/dL), serum ferritin (>73 μg/dL), and TIBC (>250 μg/dL) were included for the present study.

Exclusion criteria

Patients with age <18 years and more than 60 years, other types of anemia like hemolytic anemia, aplastic anemia, thalassemia, pregnant or lactating women, severe renal, hepatic and cardiac disease, cancer and other life-threatening diseases, any continuing blood loss, for example, hematemesis, melena, bleeding piles, etc., were excluded from the present study.

Posology

Juice of *O elatior (Nagaphani Swarasa)* preserved using sodium benzoate (1 g/kg) was given in a dose of 20 ml with lukewarm water to group A and group B received *Punarnava Mandura* [Table 1]*,* two tablets of 500 mg each, with *Takra* (butter milk)*.* Both the drugs were administered twice daily, before meal for a duration of 2 months.

Investigations

Blood parameters included total WBC, neutrophil percentage, lymphocyte percentage, eosinophil percentage, monocyte percentage, hemoglobin content, packed cell volume (PCV), TRBC, platelet count, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and MCH concentration (MCHC).

Parameters like serum ferritin, serum iron, total iron-binding capacity, serum urea, serum creatinine, serum glutamic oxaloacetic transaminase and serum glutamic pyruvic transaminase were also estimated. The tests were carried out before and after treatment. Among these, serum ferritin, serum iron, total iron-binding capacity were assessed to monitor the iron stores in the body. Other tests were carried out to rule out the adverse effects on vital organs, as the drug is new.

Collection and preparation of drug

Fruits of *Opuntia elatior* were collected from its natural habitat in its fully ripened state, from the surrounding area of Jamnagar, Gujarat, following good collection practices guidelines. Freshly collected fruits were thoroughly washed with the adequate amount of tap water and the bunch of thorns over the fruits were neatly plucked using forceps. Following this, the outer skin of fruits was removed and the pulp was macerated and the resultant juice obtained was passed through the sieve and filtered. The filtered juice was then preserved by the addition of 1 g sodium benzoate per one

Table 1: Composition and ingredients of *Punarnava Mandura*

Name of drug	Latin name	Part used	Proportion
Punarnava	*Boerhaavia diffusa* Linn.	Root	1 part
Trivrut	*O. turpethum* Linn.	Root	1 part
Shunthi	*Z. officinale* Rosc.	Rhizome	1 part
Pippali	*P. longum* Linn.	Fruit	1 part
Maricha	*P. nigrum* Linn.	Fruit	1 part
Vidanga	*E. ribes* Burm. f.	Fruit	1 part
Devadaru	*C. deodara (roxb.)* Loud.	Wood	1 part
Chitraka	*P. zeylanica* Linn.	Root	1 part
Kushtha	*S. lappa* C.B. Clarke	Root	1 part
Haridra	*C. longa* Linn.	Rhizome	1 part
Daruharidra	*B. aristata* DC	Root	1 part
Amalaki	*E. officinalis* Gaertn.	Fruit	1 part
Haritaki	*T. chebula* Retz.	Fruit	1 part
Vibhitaki	*T. belerica* Roxb.	Fruit	1 part
Danti	*Baliospurmum montanum* Muell.	Root	1 part
Chavya	*P. retrofractum* Vahl.	Fruit	1 part
Indrayava	*Holarrena antidysentrica* Wall.	Seed	1 part
Pippali	*P. longum* Linn.	Fruit	1 part
Pippalimula	*P. longum* Linn.	Root	1 part
Musta	*C. rotundus* Linn.	Rhizome	1 part
Mandura Bhasma	Incinerated red oxide of iron	-	40 parts
Gomutra	Cow's urine	-	128 parts

P. longum: *Piper longum*, *C. rotundus*: *Cyperus rotundus*, *P. retrofractum*: *Piper retrofractum*, *T. belerica*: *Terminalia belerica*, *T. chebula*: *Terminalia chebula*, *E. officinalis*: *Emblica officinalis*, *B. aristata*: *Berberis aristata*, *C. longa*: *Curcuma longa*, *S. lappa*: *Saussurea lappa*, *P. zeylanica*: *Plumbago zeylanica*, *C. deodara*: *Cedrus deodara*, *P. nigrum*: *Piper nigrum*, *E. ribes*: *Embelia ribes*, *Z. officinale*: *Zingiber officinale*, *O. turpethum*: *Operculina turpethum*, *B. diffusa*: *Boerhaavia diffusa*

liter of juice and stored in properly sealed food-grade plastic bottles of 500 ml capacity [Figures 1-8]. *Punarnava Mandura*, the standard control drug, containing many herbo-mineral ingredients [Table 1], was prepared by the Pharmacy of GAU (Gujarat Ayurved University), following recommended procedures mentioned in AFI[17] and was dispensed to the patients in airtight food grade plastic boxes.

Criteria for assessment

The assessment was done in accordance with the Clinical Research Pro forma of *Pandu Roga* developed by CCRAS,[18] with slight modifications considering the study requirement. Cardinal symptoms of *Pandu Roga* like *Panduta* (pallor)*, Daurbalya* (general weakness)*, Akshikutashotha* (periorbital edema), *Hritspandana* (palpitation), *Bhrama* (vertigo), *Shiroruja* and *Shotha* (headache and edema), *Anna-Aruchi* (loss of appetite), *Pindikodweshthana* (calf muscle cramp), *Shrama* (fatigue), *Ayasena Shwasa* (exertional dyspnoea) were graded from 0 to 4, Hematological parameters such as Hb%, total RBC serum iron, and serum ferritine were also considered before and after treatment.

Statistical analysis

The data obtained from the study were subjected to statistical tests and analyzed for the percentage of improvement in each parameter in both the trial group and standard control group. Wilcoxon signed-Rank Test was applied to evaluate the effect of therapy in the individual group for subjective criteria.

Figure 1: Photograph showing collection of fruit

Paired "*t*" test was applied to evaluate the effect of therapy on hematological and biochemical investigations. Tests were performed using Sigma Stat 3.1 software 2005 by Jandel scientific software, San Jose, California).[19] The results were interpreted as; for "*t*" test ("*P*" value) Insignificant >0.05, Significant ≤0.05 ≤0.01 ≤0.001. For Wilcoxon Signed Rank ("*P*" value) Insignificant >0.05, Significant ≤0.05 ≤0.02 ≤0.001. Coefficient of variation (CV) was applied to the statistical data for evaluating the differences of effect in between groups for clinical, hematological and biochemical parameters.
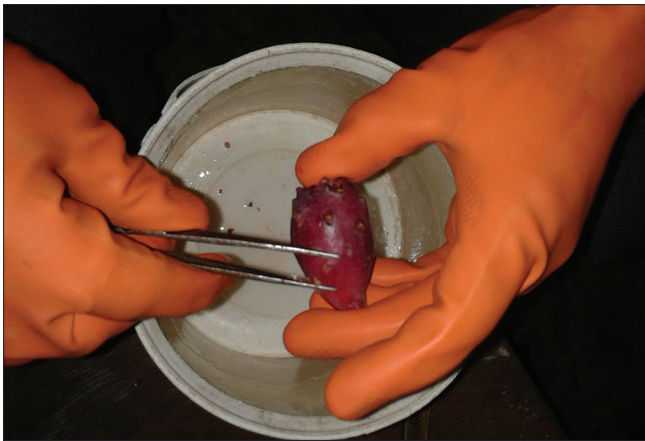
Figure 2: Bunch of thorns over the fruits neatly plucked by using forceps

Figure 3: After remove the outer skin fruits macerated

Figure 4: Resultant juice obtained, passed through sieve and filtered
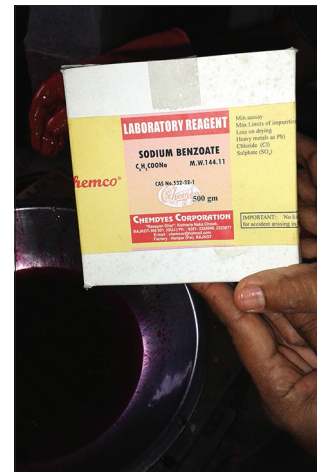
Figure 5: Photograph showing preservative i.e. Sodium benzoate

Figure 6: Sodium benzoate well mixed in the juice

Figure 7: Packing into plastic container of food grade quality

Observations

Registered 105 patients were randomly divided into two groups for evaluation of the efficacy of the *Opuntia elatior* fruit juice (*Nagaphani Swarasa)* and *Punarnava Mandura* tablet. Total 96 patients completed the intervention and the rest 9 patients (8.57%) dropped out due to various personal reasons. [Figure 9] Demographic data show, among the registered patients, maximum (62.58%) belonged to the age group of 31–50 years, in both groups. [Table 2] The scrutiny of gender revealed the incidence of *Pandu* was higher in females (78.90%), in comparison to males in both groups [Table 2]. About 33.33% of patients were housewives

Figure 8: Final product i.e. *Nagaphani swarasa*

Table 2: Age, gender wise distribution of patients (*n*=105)

Group	Age (years)			Sex	
	18-30	31-50	51-60	Male	Female
Group-A	13	33	07	09	44
Group-B	09	33	10	14	38
Total	22	66	17	23	82
Percentage	20.95	62.85	16.19	21.90	78.90

n: sample size

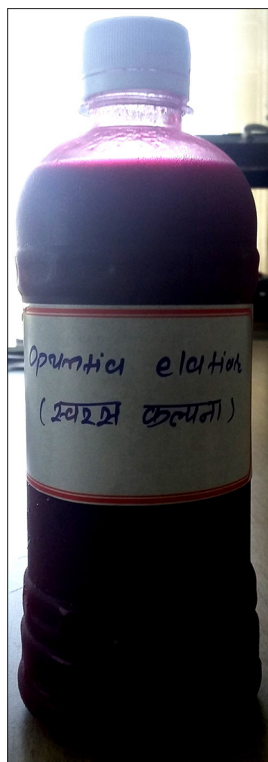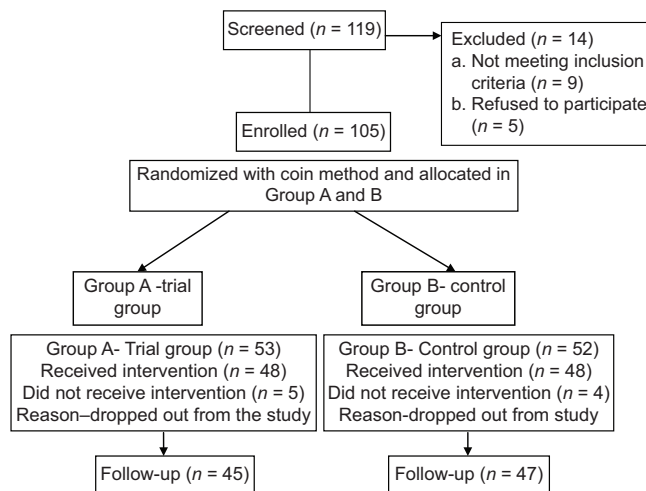as the female population was more in the study conducted, followed by 36.19% having office job as occupation and 30.47% of the registered population were from labor class [Table 3]. About 35.23% of registered patients belonged to the middle class followed by 24.76% in the lower middle class, 8.57% in upper-middle-class and 30.47% in the poor category [Table 4].

At baseline, 97.14% of patients had *Panduta* (pallor)*,* while 99.04% had *Daurbalya* (general weakness). *Akshikutashotha* (periorbital edema) was found in 36.19%. Other complains were *Shrama* (fatigue) (98.09%), *Pindikodwesthana* (calf muscle cramp), (92.38%) and *Shiroruja* (headache) (74.28%) [Table 5]. At baseline, Hb% was noted to be 9.24 ± 0.18 and 9.44 ± 0.17 in trial and standard group, respectively. TRBC was reported to be 4.30 ± 0.13 in the trial group and 4.40 ± 0.13 in the standard group. Serum iron was 50.56 ± 7.04 in the trial group and 49.80 ± 5.40 in the standard group. Serum ferritin was 21.08 ± 5.81 in trial group and 13.98 ± 3.62 in the standard group. TIBC was found to be

Figure 9: Consort flow-chart

369.13 ± 7.82 and 380.87 ± 9.40 in trial group and standard group, respectively. Maximum patients were taking *Pittaprakopaka Ahara* (food which vitiates *Pitta*) like; *Atiushna Ahara* (food which is very hot in potency) (85.71%) *Katurasayukta Ahara* (spicy food) (80.95%), *Atitikshana Ahara* (food articles which are sharp in nature) (77.14%)*, Ati Amla Ahara* (sour food) (60.95%), *Vidahibhojana* (food which causes burning sensation) (89.52%), and *Pramitabhojana* (eating less) (67.71%). *Vegavidharana* (suppression of urges of urination, defecation or hunger) was found in 90.47% and *Ratrijagarana* (being awake in the night) in 84.76% of patients while indulging in day sleep was reported in 50.47% of the patients. *Chinta* (worries) was found in the majority of patients (62.85%). *Krodha* (anger) and *Shoka* (grief) were found in 54.28% and 37.14% of patients, respectively.

Results

The effectiveness of the treatment was assessed on the basis of improvement in clinical signs and symptoms and various hematological factors pertaining to anemia, based on before and after completion of the treatment.

Both trial and standard drugs provided significant results ($P < 0.001$) in pacifying the cardinal symptoms of *Pandu Roga* [Table 6]. Both trial and standard drugs produced a statistically significant increase in Hb% (<0.001), total red blood cell (TRBC) count (<0.05). The effect of both the drugs was not significant on total whit blood cell count, MCV, MCH, MCHC, and platelet count. On PCV, the standard drug produced a statistically significant increase ($P<0.001$) while the effect of the trial drug was statistically insignificant ($P>0.05$) [Table 7]. There was a statistically significant increase in serum iron level in both the groups but the increase level was high in standard (26.10%) than trial group (17.85%). Both provided significant increase in serum ferritin level, but the level of increase was much higher in standard (82.68%) than trial drug (14.32%. In TIBC level, statistically significant decrease ($P < 0.05$) in trial drug (1.14%) and statistically insignificant

decrease ($P > 0.05$) in standard drug (6.01%) was observed. [Table 8] Both the drugs provided insignificant ($P > 0.05$) changes in serum creatinine, blood urea, SGPT, SGOT, and blood sugar level [Table 9]. There was no significant difference ($P > 0.05$) in the comparative effect of therapies between trial and standard drug in clinical signs and symptoms, Daurbalya (general weakness), Panduta (pallor), Akshikutashotha (periorbital edema), Hritspandana (palpitation), Bhrama (dizziness), Aruchi (tastelessness), Pindikodweshthana (calf muscle cramps), Shrama (fatigue), Ayasaja Shwasa (dyspnea on exersion) [Table 10] as well as hematological [Table 11] and biochemical parameters [Table 12].

Discussion

The study drug *Opuntia elatior* has been reported to be safe[12] and a potent hematinic, through various animal experiments.[13,14] Clinical efficacy of the drug in anemia, has been reported on the geriatric population.[15] *Punarnava*

Mandura, was considered as the standard control, based on its classical claims and being re-established for its hematinic activity, through various animal and clinical experiments. In this study, an attempt had been made to evaluate the clinical efficacy of *Nagaphani (O. elatior)* fruit juice on the general population of anemia.

According to the demographic data, most of the registered patients belonged to the middle class, lower middle class and poor class. More incidence of anemia was seen in the female population, aged between 31 and 50 [Tables 2 and 3]. This may be due to a lack of nutrition owing to their economic status and nature of work.

The better percentage of improvement and significant results in both groups with regards to increase in hemoglobin might be due to the direct availability of iron element in its most acceptable form to the human body, which aided in quicker response in hematological parameters.

The presence of iron and ascorbic acid (Vitamin C) in trial drug[20] which reduces ferric iron to ferrous iron and remains soluble even at neutral pH and is better absorbed. Even when the diet is poor in iron, Vitamin C supplements with each meal enhances iron absorption. Vitamin C taken in divided doses with each meal increase iron absorption to a greater extent.[21] Thus, *Opuntia elatior* (trial drug) possesses a promising role in the management of anemia, especially iron-deficiency anemia. Its mode of action in modern parlance correlates with the *Raktavardhaka* (hematinic) property in Ayurveda probably by enhancing the normal physiological functions of *Pitta Dosha* and promoting the normal formation of *Raktadhatu* by *Ranjana* (coloration) of *Rasadhatu*.

There was no significant difference ($P > 0.05$) in the comparative effect of therapies between trial and standard drug on Hb% and RBC count. Both the drugs contain iron and Vitamin C, so it can produce improvement in Hb%.

Both trial and standard drugs were significant in increasing serum iron and ferritin. However, the standard drug was more significant in increasing serum iron (<0.001) and serum ferritin (>0.05) when compared to the trial drug.

Table 3: Occupation wise distributions of patients (*n*=105)

Group	Occupation		
	Labor	House wife	Service
Group-A	14	20	19
Group-B	18	15	19
Total	32	35	38
Percentage	30.47	33.33	36.19

n: sample size

Table 4: Socioeconomic wise status of patients (*n*=105)

Group	Socioeconomic status				
	Poor	Lower middle	Middle	Upper middle	Rich
Group-A	15	14	19	4	1
Group-B	17	12	18	5	-
Total	32	26	37	9	1
Percentage	30.47	24.76	35.23	8.57	0.95

n: sample size

Table 5: Chief complaints presented by study subjects (*n*=105)

Chief complaints (*n*=105)	Number of patients		Total	Percentage of patients
	Group-A	Group-B		
Pandu Rogata (pallor)	52	50	102	97.14
Daurbalya (general weakness)	53	51	104	99.04
Akshikutashotha (periorbital oedema)	20	18	38	36.19
Hridspandana (palpitation)	30	23	53	50.47
Bhrama (vertigo/giddiness)	36	26	62	59.04
Shiroruja and *Shotha* (headache and oedema)	40	38	78	74.28
Anna-aruchi (loss of appetite)	30	32	62	59.04
Pindikodweshtana (calf muscle cramp)	46	51	97	92.38
Shrama (fatigue)	52	51	103	98.09
Shwasa (exertional dyspnoea)	42	30	72	68.57

n: sample size

Table 6: Effect of therapies on cardinal symptoms (48 patients in each group)

Symptoms (*n*=96)	Group	Mean±SEM		Relief		Rank (W)	T+	T−	P	S
		BT	AT	Mean±SEM	Percentage					
Pandu Rogata	A	3.00±0.00	1.56±0.13	1.44±0.13	48↓	−70	0	−70	<0.001	SI
	B	2.66±0.09	1.59±0.10	1.07±0.01	40.22↓	−59	0	−59	<0.001	SI
Daurbalya	A	3.00±0.00	1.10±0.12	1.90±0.12	63.33↓	−1128	0	−1128	<0.001	SI
	B	2.93±0.03	1.12±0.12	1.81±0.09	61.77↓	−1035	0	−1035	<0.001	SI
Akshikuta	A	1.45±0.18	0.85±0.10	0.60±0.08	41.37↓	−66	0	−66	<0.001	SI
Shotha	B	1.72±0.13	1.00±0.14	0.72±0.01	41.86↓	−78	0	−78	<0.001	SI
Hrida	A	1.61±0.15	1.00±0.09	0.61±0.06	37.88↓	−105	0	−105	<0.001	SI
Spandana	B	1.86±0.11	1.04±0.10	0.82±0.01	44.08↓	−136	0	−136	<0.001	SI
Bhrama	A	1.78±0.08	0.96±0.11	0.82±0.03	46.06↓	−231	0	−231	<0.001	SI
	B	1.81±0.10	0.92±0.12	0.89±0.02	49.17↓	−190	0	−190	<0.001	SI
Shiroruja	A	1.86±0.08	0.81±0.09	1.05±0.01	56.45↓	−595	0	−595	<0.001	SI
	B	1.89±0.07	0.73±0.08	1.16±0.01	61.37↓	−595	0	−595	<0.001	SI
Aruchi	A	1.93±0.13	0.96±0.08	0.97±0.05	50.25↓	−351	0	−351	<0.001	SI
	B	2.15±0.11	0.90±0.09	1.18±0.01	54.88↓	−406	0	−406	<0.001	SI
Pindiko-dwestana	A	3.15±0.11	0.97±0.10	2.18±0.01	69.20↓	−0.99	0	−0.99	<0.001	SI
	B	3.14±0.06	1.23±0.10	1.91±0.04	60.82↓	−1081	0	−1081	<0.001	SI
Shrama	A	2.87±0.04	1.08±0.09	1.79±0.05	62.36↓	−112	0	−112	<0.001	SI
	B	3.14±0.07	1.29±0.11	1.85±0.04	58.91↓	−1128	0	−1128	<0.001	SI

n: sample size, SEM: Standard error of the mean, BT: Before treatment, AT: After treatment, Rank (W): Wilcoxon rank-sign test, *P*: Level of significance, SI: Statistically insignificant

Table 7: Effect of therapies on hematological parameters (48 patients in each group)

Parameters (*n*=96)	Group	Mean±SEM		Difference	SD (±)	Percentage	't'	P
		BT	AT					
Hemoglobin (g %)	A	9.24±0.18	10.35±0.26	−1.10	1.88	11.90↑	−4.08	<0.001
	B	9.44±0.17	10.46±0.24	−1.02	1.81	10.88↑	−3.91	<0.001
TRBC (mil/cumm)	A	4.30±0.13	4.57±0.11	−0.265	0.67	6.16↑	−2.715	<0.05
	B	4.40±0.13	4.72±0.11	−0.321	0.78	7.29↑	−2.822	<0.05
TWBC (/Cumm)	A	6337.50±274.38	6706.25±274.35	−368.75	1911.12	30.15↑	−1.337	>0.05
	B	6354.16±241.21	6668.75±245.94	−314.58	2074.87	4.95↑	−1.050	>0.05
MCV (fl)	A	71.69±2.20	72.34±1.83	−0.65	7.87	0.90↑	−0.572	>0.05
	B	70.83±1.84	69.45±2.13	1.37	12.45	1.93↓	0.766	>0.05
MCH (Pg)	A	22.49±0.81	22.91±0.68	−0.42	3.27	1.86↑	−0.903	>0.05
	B	22.11±0.61	22.57±0.57	−0.46	2.61	2.08↑	−1.230	>0.05
MCHC (g/dl)	A	31.20±0.27	31.57±0.26	−0.37	1.47	1.18↑	−1.747	>0.05
	B	31.26±0.26	31.52±0.27	−0.26	1.17	0.83↑	−1.539	>0.05
PCV (%)	A	92.61±62.67	32.65±0.76	59.96	434.47	64.74↓	0.956	>0.05
	B	30.40±0.56	33.36±0.63	−2.96	5.24	9.73↑	−3.909	<0.001
Platelet count (10³/ul)	A	2080.18±1743.02	328.39±12.60	1751.79	12,074.64	84.21↓	1.005	>0.05
	B	331.52±15.74	316.54±14.05	14.79	87.15	4.46↓	1.191	>0.05

n: sample size, SEM: Standard error of the mean, BT: Before treatment, AT: After treatment, SD: Standard deviation, *P*: Level of significance, PCV: Packed cell volume, MCV: Mean corpuscular volume, MCH: Mean corpuscular hemoglobin, MCHC: Mean corpuscular hemoglobin concentration, total red blood cell (TRBC), total white blood cell (TWBC)

In the management of *Panduta*, the results in both groups were statistically significant ($P < 0.001$). There was no significant difference ($P > 0.05$) in the comparative effect of therapies between trial drugs and standard drugs. *Madhura Rasa* (sweet taste) and *Snigdhaguna* (unctuous nature)[22] of trial drug may have worked in the pacification of *Rukshata* of *Vata* (dryness produced by *Vata*) and *Tikshnata* of *Pitta* (sharp nature of *Pitta*). This might have helped in the correction of the physiological function of *Ranjaka Pitta* which would have normalized *Raktadhatu* formation.

Trial drug was found to be statistically more significant ($P < 0.001$) in relieving *Daurbalya* (63.33%). This may be due to *Madhura Rasa* (sweet taste) which reduces *Pitta* and replenishes *Rasadhatu*, hence does *Poshana* of *Uttarottara Dhatu* (progressive nourishment of body tissues), in turn, results in relief of *Daurbalya* (general weakness). This can

Table 8: Effect of therapy on anemia profile (48 patients in each group)

Parameters (n=96)	Group	Mean±SEM BT	Mean±SEM AT	Difference	SD (±)	Percentage	't'	P
Serum iron (µg/dl)	A	50.56±7.04	59.59±7.33	−9.03	24.41	17.85↑	−2.562	<0.01
	B	49.80±5.40	62.80±5.11	−13	22.59	26.10↑	−3.984	<0.001
Serum ferritin	A	21.08±5.81	24.10±6.38	−3.02	21.50	14.32↑	−0.973	>0.05
	B	13.98±3.62	25.54±9.24	−11.56	65.86	82.68↑	−1.216	>0.05
TIBC (mg/dl)	A	369.13±7.82	364.90±7.33	4.23	48.55	1.14↓	0.604	<0.05
	B	380.87±9.40	358.68±6.26	22.19	67.62	6.01↓	2.273	>0.05

n: sample size, SEM: Standard error of the mean, BT: Before treatment, AT: After treatment, SD: Standard deviation, *P*: Level of significance, TIBC: Total iron-binding capacity

Table 9: Effect of trial drugs on biological parameters (48 patients in each group)

Group	n	Mean±SEM BT	Mean±SEM AT	Difference	SD(±)	Percentage	't'	P
Serum creatinine								
A	48	0.79±0.02	0.83±0.03	−0.03	0.21	3.79↑	−1.235	>0.05
B	48	1.12±0.19	0.97±0.13	0.14	0.91	12.5↓	1.085	>0.05
Blood urea								
A	48	22.16±0.89	22.27±1.13	−0.10	6.18	0.45↑	−0.117	>0.05
B	48	23.60±1.99	26.98±4.38	−3.38	33.39	14.32↑	−0.702	>0.05
SGPT								
A	48	18.02±2.29	17.10±1.30	0.91	11.76	5.04↓	0.540	>0.05
B	48	15.87±1.70	16.60±1.18	−0.72	13.01	4.53↑	−0.388	>0.05
SGOT								
A	48	26.70±1.97	25.56±1.74	1.14	9.706	4.26↓	0.818	>0.05
B	48	26.16±2.07	23.41±0.90	2.75	15.09	10.51↓	1.262	>0.05
Blood sugar								
A	48	86.10±2.47	83.75±2.52	2.35	17.34	2.72↓	0.940	>0.05
B	48	88.12±3.65	85.89±4.52	2.22	20.31	2.51↓	0.760	>0.05

n: sample size, SEM: Standard error of the mean, BT: Before treatment, AT: After treatment, SD: Standard deviation, *P*: Level of significance, SGPT: Serum glutamic pyruvic transaminase, SGOT: Serum glutamic oxaloacetic transaminase

Table 10: Comparative effect of trial drugs on chief complaints

Chief complaints	N A	N B	Mean A	Mean B	SD A	SD B	CV A	CV B	Better group
Panduta	48	47	1.42	1.06	1.01	0.84	71.12	79.24	Group A
Daurbalya	48	47	1.85	1.80	0.87	0.85	47.02	47.22	Group A
Akshikutashotha	19	17	0.68	0.76	0.67	0.56	98.52	73.68	Group B
Hridspandana	24	21	0.66	0.90	0.63	0.53	95.45	58.88	Group B
Bhrama	32	26	0.81	0.96	0.69	0.66	85.18	68.75	Group B
Shiroruja and shotha	37	36	1.08	1.19	0.49	0.52	45.37	43.69	Group B
Anna-Aruchi	28	31	1.03	1.29	0.42	0.78	40.77	60.46	Group A
Pindikodveshthana	48	47	2.22	1.91	0.80	0.83	36.03	43.45	Group A
Shrama	48	48	1.79	1.85	0.65	0.82	36.31	44.32	Group A
Shwasa	37	26	0.67	0.69	0.58	0.54	86.56	78.26	Group B

n: Sample size, SD: Standard deviation, CV: Coefficient of variance

also be attributed to macronutrients like; protein (1.98%w/w), carbohydrate (7.54%w/w), total sugar (13.73% w/w), Vitamin C (1.29% w/w), and fat (0.25%w/w).[23] present in *O. elatior* fruit (trial drug). *Punarnava Mandura* which is a herbo-mineral compound is having herbal drugs like *Shunthi, Pippali,*

Maricha, Chitraka which are having appetizer, digestive, and carminative properties.[24] Hence, it improves digestive power and ultimately absorption of nutrients and drug.

In relieving *Hritspandana* (palpitation) and *Bhrama* (giddiness), both trial and standard drug provided significant (*P* < 0.001)

Table 11: Comparative effect of test drugs on hematological parameters

Parameters	N		Mean		SD		CV		Better group
	A	B	A	B	A	B	A	B	
Hemoglobin	48	48	1.10	1.02	1.88	1.81	170.90	177.45	Group A
TWBC	48	48	368.75	314.58	1911.12	2074.87	518.26	659.56	Group A
Neutrophils	48	48	1.47	0.37	10.37	8.77	705.44	2370.27	Group A
Lymphocytes	48	48	0.39	0.20	9.95	8.42	2551.28	4210	Group A
Eosinophils	48	48	0.02	0.72	1.39	2.39	6950	331.19	Group B
Monocytes	48	48	0.04	0.06	0.77	0.81	1925	1350	Group A
T RBC	48	48	0.26	0.32	0.67	0.78	257.69	243.75	Group B
ESR	48	48	8.75	3.05	27.80	31.33	317.71	1027.21	Group A
MCV	48	48	0.65	1.37	7.87	12.45	1210.76	908.75	Group B
MCH	48	48	0.42	0.46	3.27	2.61	778.57	567.39	Group B
MCHC	48	48	0.37	0.26	1.47	1.17	397.29	450	Group A
PCV	48	48	59.96	2.96	434.47	5.24	724.59	117.02	Group B
Platelet counts	48	48	1751.79	14.97	12074.64	87.15	689.27	582.16	Group B

n: Sample size, SD: Standard deviation, CV: Coefficient of variance, PCV: Packed cell volume, MCV: Mean corpuscular volume, MCH: Mean corpuscular hemoglobin, MCHC: Mean corpuscular hemoglobin concentration, total red blood cell (T RBC), erythrocyte sedimentation rate (ESR) , total white blood cell (TWBC)

Table 12: Comparative effect of test drugs on biochemical parameters

Parameters	N		Mean		SD		CV		Better group
	A	B	A	B	A	B	A	B	
Serum ferritin	48	48	3.02	11.56	21.50	65.86	711.92	569.72	Group B
Serum iron	48	48	9.02	12.99	24.41	22.59	270.62	173.90	Group B
TIBC	48	48	4.23	22.18	48.55	67.62	1147.75	304.86	Group B
Creatinine	48	48	0.03	0.14	0.21	0.91	700	650	Group B
Blood urea	48	48	0.10	3.38	6.18	33.39	6180	987.86	Group B
SGPT	48	48	0.91	0.72	11.76	13.01	1292.30	1806.94	Group A
SGOT	48	48	1.14	2.75	9.70	15.09	850.87	548.72	Group B
Blood sugar	48	48	2.35	2.22	17.34	20.31	737.87	914.86	Group A

n: Sample size, SD: Standard deviation, CV: Coefficient of variance, TIBC: Total iron binding capacity, SGPT: Serum glutamic pyruvic transaminase, SGOT: Serum glutamic oxaloacetic transaminase

results. This can be substantiated through the pacification of *Vata* and *Pitta* because of *Madhura Rasa* and *Snigdha Guṇa* of trial drug. Standard drug contains *Mandura* which is *Raktavardhaka* (hematinic) so helps to increase oxygen-carrying capacity thus workload on the heart decreases. Hence, it might be the reason in decreasing *Hridspandana*.

Vatanulomaka (facilitating the downward movement of *Vata*) and *Srotoshodhaka* (clearing the channels) properties of standard drug helps in proper circulation in the brain, so relief was found in *Bhrama* (giddiness).

Vatapittahara (pacifying *Vata* and *Pitta*) property of the trial drug, may have improved digestion and thus, managed to treat *Aruchi* (tastelessness). Standard drug has *Kapha Shamaka* (pacifying *Kapha*), *Dipana* (carminant), *Amapachana* effect, which might have facilitated to decrease *Aruchi*.

Vatahara (pacifying *Vata*) property of the trial drug, may have contributed to relieve *Pindikodweshtana* (calf muscle pain). In standard drugs, this can be attributed to its *Vata-Kapha Shamaka* property.

Increase in *Rasa* and *Uttarottara Dhatu* (subsequent tissues) by trial drug gives proper nutrition to muscles and tissues which might be the cause of relief in *Shrama* (fatigue). Standard drug, due to its *Dipana* (carminant), *Pachana* (digestive) and *Rasayana* (rejuvenating) properties may provide nourishment and strength to tissues and thus may relieve fatigue.

Trial drug improves the quality of *Rasa* and *Rakta* as explained earlier might reduce the load on the heart and improves the circulation, thus relief of *Ayasena Shwasa* (dyspnea on exertion). *Mandura Bhasma* in *Punarnava Mandura* increases oxygen-carrying capacity of RBCs may facilitate the heart in its functions, which in turn helps reducing *Shwasa*. By virtue of *Rasa* (taste) and *Guna* (properties), pacifies aggravated *Pitta* (bile) and maintains normalcy, improves digestion and metabolism. The ferric and ferrous fractions of *Mandura* provide sufficient amount of iron, which is needed for normal erythropoiesis.

Other drugs present in *Punarnava Mandura*[25] like *Haridra (Curcuma longa* Linn.*)*, *Amalaki (Phylanthus embelica* Linn.*), Pippali (Piper longum* Linn.*),*

Punarnava (Boerhavia diffusa Linn.*),* and *Trivrut (Operculina terpethum* Linn.*)* are *Pandughna* in nature.[24] Drugs like *Amalaki (Phylanthus embelica* Linn.*)*,[26], *Danti (Baliospermum momtanum* Muell. Arg.*)*,[27] *Pippali (Piper longum* Linn.*)*,[28] *Punarnava (Boerhavia diffusa* Linn.*)*,[29] *Kushtha (Sausurea lappa)*[30] and *Daruharidra (Berberis aristate* DC.*)*[31] are documented as drugs which are having immunomodulator and antioxidant properties. Hence, they may have the potential to confer beneficial health effects due to their antioxidant activity and thus useful in anemia. *Amalaki* possesses antioxidant activity and could be an important dietary source of Vitamin C, which is a powerful water-soluble anti-oxidant[32] and helps in increasing iron absorption from the gut. In addition, *Pippali* is said to be bioavailability enhancer of the drug,[33] which further helps in easy assimilation of the drug components. Cow urine is reported to have anti-anemic action due to its erythropoietin stimulating factor.[34] Buttermilk used as *Anupana* (vehicle) has *Dipana-Pachana* (carminative and digestive), *Pandughna*[35] properties and rich source of minerals and Vitamin B$_{12}$.[36]

The trial drug showed equally significant results in many of the factors analyzed, as compared to the standard group. Although the study revealed that the standard drug is more effective when compared with the trial drug, considering few factors regarding the trial drug like, it is used as a single drug, it is mineral-free, nontoxic, easily available, palatable, nutritive, cost-effective, it can be a drug of choice in iron deficiency anaemia.

Conclusion

Nagaphani (*Opuntia elatior* Mill.) fruit juice and *Punarnava Mandura* were found effective in the management of *Pandu Roga* by significantly relieving the cardinal symptoms of *Pandu Roga*. Both the drugs produced a statistically significant increase in Hb%, TRBC and serum iron. Trial drug showed a statistically significant reduction of total iron-binding capacity, while *Punarnava Mandura* produced a statistically insignificant reduction. When the effect was compared in between the groups, standard drug, *Punarnava Mandura* was seen to be more effective than the trial drug, *Nagaphani* (*O. elatior*) fruit juice. No adverse reactions were reported during the clinical trial indicating the safety of *Nagaphani* fruit juice. These results support the folklore use of *O. elatior* fruits in the treatment of anemia. Further large-scale studies may be carried out to establish *O. elatior* as the choice of drug for the management of *Pandu Roga* (iron deficiency anaemia).

Conflicts of interest

There are no conflicts of interest.

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