

# Comparative therapeutic response of instant and incremental oleation (*Sadya* and *Aarohi Snehapana*) over hematological and biochemical parameters in healthy volunteers: a pilot study

Punam G. Sawarkar<sup>1</sup>, Gaurav R. Sawarkar

Departments of Panchakarma and <sup>1</sup>Rachana Sharir, Mahatma Gandhi Ayurved College, Hospital and Research Centre, DMIMS (DU), Wardha, Maharashtra, India

## Abstract

**Introduction:** *Snehapana* (oleation) is the prerequisite treatment for the purification in Panchakarma. Internal oleation therapy is essential for the *Dosha* migration in the body. So, to identify the effectiveness of instant oleation with incremental as well as safety nature of intervention, the pilot study was planned. **Materials and Methods:** The study was randomized, parallel group, controlled trial. Participants eligible for internal oleation were included in the study, having age in-between 20 and 40 years. Plain cow ghee was the intervention for both groups. The only difference was in dose pattern. In Group A, 150 mL cow ghee with rock salt as used instant administration, whereas in Group B, administration was carried out in incremental dose from 30 mL to maximum 210 mL (up to the appearance of proper oleation features). Fifteen participants were recruited in each group. Randomization was carried out with simple block method. **Objective:** The pilot study was intended to measure the hematological and biochemical changes appearing after *Snehapana* (internal oleation). **Outcome:** The primary trial objectives of the study were to evaluate therapeutic response of internal oleation in instant and incremental manner and to find out effective modality in vogue. Secondary outcome was to assess safety for the consumption of cow ghee in large amount, which is not good as pretended by contemporary science. **Conclusion:** Intended primary outcome was not possible to attain because of small sample size. But significant results were obtained in some parameters such as blood urea, serum bilirubin, total bilirubin, serum triglyceride, serum glutamic-pyruvic transaminase, and very low-density lipoprotein ( $P < 0.05$ ) but within normal range, and the intervention in both were safe in nature, none of the participants had developed any harm in the study. Further clinical trial is suggested for large sample size.

**Keywords:** Biochemical, hematological, oleation, *Sadya* and *Aarohi Snehapana*

## INTRODUCTION

*Snehapana* (oleation) is one of the preparatory therapies in Ayurveda, a system of medicine in India. *Sneha* means fat, either oil or ghee. Oleation is a prerequisite treatment in Panchakarma.<sup>[1]</sup> The ghee is made from clarified butter (obtained from cow's milk) and it is boiled with various herbs to make a specific disease-oriented medicine of *Sneha*. Oleation is carried out in the morning after attending to the calls of nature, on an empty stomach for *Shodhana* (purification) purpose.<sup>[2]</sup> The health, age, illness condition, and digestive capacity of the patient needs to be taken into consideration when determining the dosage of *Sneha*

for unction. The treatment lasts between minimum 3 and maximum 7 days depending on the condition of the patient and the illness being treated.<sup>[3]</sup> Many times, the oleation is used as preparative phase, which makes it an ideal suitable environment for elimination of the *Dosha*. But the changes that are introduced in the blood circulation due to this modality are not yet authentically established. Thereby, it

**Address for correspondence:** Dr. Gaurav R. Sawarkar,  
Department of Rachana Sharir, Mahatma Gandhi Ayurved College,  
Hospital and Research Centre, DMIMS (DU), Salod (Hirapur),  
Wardha 442001, Maharashtra, India.  
E-mail: drsawarkar.gaurav@gmail.com

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is a need-based study to evaluate the hematological and biochemical changes appearing after *Snehapana*.

**Aims and objective:** The primary aim of the study was to evaluate the therapeutic response in instant and incremental oleation over hematological and biochemical changes in the body; in addition, which of the modality is effective in vogue, and the generation of collateral evidence with safety profile for the consumption of cow ghee in large amount, which is considered as unhealthy as misconception of contemporary science.

## MATERIALS AND METHODS

### Trial Design and Participants, Sample Size, Inclusion, and Exclusion

The study was randomized, parallel group, controlled trial. Between the age of 20 and 40 years, all healthy participants (both males and females) who were fit for oleation<sup>[4]</sup> were included in the study and who had aversion to the fat were excluded from the study. The study was conducted in two groups, Group A with instant oleation and Group B with incremental oleation. Total 30 participants were enrolled in the study and equally distributed in both the groups.

$$n = \frac{Z^2 P x (1 - P)}{d^2}; n = \frac{1.96^2 \times 0.07 \times 0.93}{0.05^2}$$

With confidence interval 5% level of significance and 95% power of test using aforementioned formula.

### Study Center

This study was carried out at the Mahatma Gandhi Ayurved College, Hospital and Research Centre, Salod (Hirapur), Wardha, Maharashtra, India.

### Intervention

The cow ghee was the intervention drug in the study, which would be administered in two-way modality; instant and incremental. In instant oleation, cow ghee was administered at once in the quantity of 150 mL stat with rock salt, whereas in incremental, it was administered in 30, 60, 90, 120, 150, 180, and 210 mL dose (i.e., fixed 30 mL increase daily) up to the appearance of proper oleation features for not more than 7 days.<sup>[5]</sup> Total time taken for completion of experimental study was 6 months.

### Methodology

Heading	Group A	Group B
Sample size	15	15
Intervention	Instant oleation	Incremental oleation
Selection of materials	Standard material (cow ghee) for oleation	
Composition of material	Cow ghee + rock salt	Cow ghee
Dosage	150 g cow ghee + 10 g rock salt	30 g of cow ghee in progression/day

Heading	Group A		Group B	
Duration of treatment	Day 1–3	<i>Trikatu</i> powder 1 gm with lukewarm water twice daily before meal for 3 days	Day 1–3	<i>Trikatu</i> powder 1 gm with lukewarm water twice daily before meal for 3 days
	4th day	Pretest sampling	4th day	Pretest sampling
	4th day	150 g cow ghee + 10 g rock salt at 7 A.M. on empty stomach	4th–10th day	30 g of cow ghee in progression/daily (with 30 g increase daily) at 7 A.M. on empty stomach (dose was increased equal to the test dose daily till the appearance of proper oleation features or maximum 7 days which ever was earlier)
	5th day	Posttest sampling (after 24 h of ghee consumption)	Next day of last <i>Snehapana</i>	Posttest sampling (after 24 h of last dose ghee consumption)
<i>Anupana</i>	Lukewarm water			
Follow-up	7th day	After 2 consecutive days	-	After 3 consecutive days
Total duration	6 days		9–13 days	

### Outcomes

The primary outcome of the study was to evaluate hematological and biochemical changes in the body in both modalities as well as which one of the modalities effective in vogue and to develop safety profile for the consumption of cow ghee in such a large amount. To assess the hematological and biochemical parameters, in Group A, the first blood sample was drawn on the very first day before starting oleation in both the groups and the second was drawn after 24 h of consumption of 150 mL, whereas in Group B, it was obtained after the appearance of proper oleation features as per subjective parameters quoted in context.<sup>[6]</sup>

### Randomization

Block method was used to generate the random allocation sequence of the participants. Random allocation was made in blocks to keep the sizes of treatment groups similar. The randomization was carried out by allocating random permutations of treatments within each block.

### Statistical Methods

Paired and unpaired *t* tests for mean values of normally distributed continuous data, and nonparametric tests by Wilcoxon rank sum test and Mann–Whitney rank sum test for skewed continuous variables. A *P* value < 0.05 was considered statistically significant. SigmaStat software for

Windows, version 3.1 (Copyright 2004, SYSTAT software, USA) was used for all analyses.

## RESULTS

The paired *t* test was used to compare hematological and biochemical parameters before and after the intervention, whereas unpaired *t* test was used for the comparison between Groups A and B. Where, normality test failed nonparametric Wilcoxon rank sum test for comparison within the group and Mann–Whitney rank sum test for comparison in between the groups were used for the purpose of statistic.

In Tables 1 and 2, data pertaining to hematological and biochemical parameters in Group A are depicted. The

findings represent that all parameters showed changes after intervention but these changes were not statistically significant except bleeding time (BT), neutrophil count, serum albumin (Sr. Alb), and serum very low-density lipoprotein (VLDL). BT and neutrophil count were found to be significantly decreased, whereas serum VLDL and Sr. Alb values were found to be significantly increased.

In Tables 3 and 4, data pertaining to hematological and biochemical parameters in Group B are depicted. The findings represent that all parameters showed changes after intervention but these changes were not statistically significant except serum blood urea and serum direct bilirubin, in which values significantly decreased.

**Table 1: Observations of hematological parameters in Group A (paired *t* test, instant oleation)**

Parameter	Treatment name	N	Mean	Standard deviation	SEM	<i>t</i> value	<i>P</i> value
HB	BI	15	12.567	1.607	0.415	0.786	0.445
	AI	15	12.387	1.603	0.414		
	Difference	15	0.18	0.887	0.229		
TLC	BI	15	6226.667	1084.611	280.045	−0.579	0.572
	AI	15	6440	1501.808	387.765		
	Difference	15	−213.333	1427.719	368.635		
N	BI	15	56.8	6.603	1.705	−2.219	0.043
	AI	15	59.867	6.278	1.621		
	Difference	15	3.067	5.351	1.382		
L	BI	15	37.6	6.045	1.561	0.935	0.366
	AI	15	36.267	5.97	1.541		
	Difference	15	1.333	5.525	1.427		
Total RBC	BI	15	4.753	0.691	0.178	−0.783	0.447
	AI	15	4.803	0.672	0.174		
	Difference	15	−0.0507	0.251	0.0647		
T. platelet	BI	15	260733.3	96703.87	24968.83	−0.217	0.831
	AI	15	262466.7	90590.02	23390.24		
	Difference	15	−1733.33	30918.02	7982.998		
ESR	BI	15	29	22.656	5.85	2.098	0.055
	AI	15	24.067	19.998	5.163		
	Difference	15	4.933	9.106	2.351		
MCH	BI	15	25.9	5.487	1.417	0.301	0.768
	AI	15	25.8	5.343	1.38		
	Difference	15	0.1	1.286	0.332		
BSL-F	BI	15	95.933	14.582	3.765	−0.276	0.787
	AI	15	96.667	13.663	3.528		
	Difference	15	−0.733	10.306	2.661		
BSL-PP	BI	15	108.933	14.28	3.687	0.285	0.780
	AI	15	108	19.745	5.098		
	Difference	15	0.933	12.697	3.278		
Sr. Cr.	BI	15	0.87	0.216	0.0557	0.470	0.645
	AI	15	0.86	0.167	0.0432		
	Difference	15	0.01	0.0824	0.0213		
Sr. Na	BI	15	139.533	2.924	0.755	1.054	0.310
	AI	15	139	3.185	0.822		
	Difference	15	0.533	1.959	0.506		
Sr. K	BI	15	4.159	0.408	0.105	−0.136	0.894
	AI	15	4.173	0.363	0.0938		
	Difference	15	−0.014	0.4	0.103		

**Table 1: Continued**

Parameter	Treatment name	N	Mean	Standard deviation	SEM	t value	P value
Sr. T Bil	BI	15	0.877	0.442	0.114	0.555	0.588
	AI	15	0.821	0.481	0.124		
	Difference	15	0.056	0.391	0.101		
Sr. D Bil	BI	15	0.299	0.123	0.0318	-0.520	0.611
	AI	15	0.314	0.168	0.0435		
	Difference	15	-0.0147	0.109	0.0282		
Sr. ID Bil	BI	15	0.615	0.427	0.11	0.0565	0.956
	AI	15	0.611	0.491	0.127		
	Difference	15	0.004	0.274	0.0708		
SGOT	BI	15	20.4	3.942	1.018	-0.846	0.412
	AI	15	21.733	7.545	1.948		
	Difference	15	-1.333	6.102	1.576		
Sr. Alk Phos	BI	15	181	85.858	22.168	-1.126	0.279
	AI	15	190	68.403	17.662		
	Difference	15	-9	30.958	7.993		
Sr. Pro T	BI	15	7.3	0.568	0.147	-1.062	0.306
	AI	15	7.467	0.628	0.162		
	Difference	15	-0.167	0.608	0.157		
Sr. Glob	BI	15	2.913	0.513	0.132	-0.462	0.651
	AI	15	2.973	0.543	0.14		
	Difference	15	-0.06	0.503	0.13		
Sr. T Chol	BI	15	185.867	56.591	14.612	-1.473	0.163
	AI	15	192.667	49.348	12.742		
	Difference	15	-6.8	17.873	4.615		
Sr. HDL	BI	15	35.133	12.094	3.123	-2.112	0.053
	AI	15	38.467	14.237	3.676		
	Difference	15	-3.333	6.114	1.579		
Sr. LDL	BI	15	134.2	41.679	10.762	-0.316	0.757
	AI	15	135.667	40.489	10.454		
	Difference	15	-1.467	17.964	4.638		
BT	BI	15	84.533	29.321	7.571	2.507	0.025
	AI	15	72	23.664	6.11		
	Difference	15	12.533	19.361	4.999		
CT	BI	15	253.533	53.38	13.783	-1.642	0.123
	AI	15	288.067	84.86	21.911		
	Difference	15	-34.533	81.47	21.036		

SEM = standard error of mean, HB = hemoglobin, RBC = red blood cell, ESR = erythrocyte sedimentation rate, MCH = mean corpuscular hemoglobin, BSL-F = blood sugar level (fasting), BSL-PP = blood sugar level (postprandial), Sr. Cr. = serum creatinine, Sr. Na = serum sodium, Sr. K = serum potassium, Sr. TBil = serum total bilirubin, Sr. DBil = serum direct bilirubin, Sr. ID Bil = serum indirect bilirubin, SGOT = serum glutamic-oxaloacetic transaminase, Sr. AlkPhos = serum alkaline phosphate, Sr. Pro T = serum protein, Sr. Glob = serum globulin, Sr. T Chol = total cholesterol, Sr. HDL = serum high-density lipoproteins, Sr. LDL = serum low-density lipoproteins, BT = bleeding time, CT = clotting time, BI = before intervention, AI = after intervention, TLC = total leukocyte count, N = neutrophil, L = leucocyte

In Tables 5 and 6, data pertaining to hematological and biochemical parameters in Group B are depicted. The findings represent that all parameters showed changes after intervention but these changes were not statistically significant except serum protein, Sr. Alb, basophils, mean corpuscular volume (MCV), serum total bilirubin (Sr. TBil), serum glutamic-oxaloacetic transaminase (SGOT), and SGPT.

In Tables 7 and 8, data pertaining to hematological and biochemical parameters in Group A and Group B are depicted. The findings represent that all parameters showed changes after intervention but these changes were not statistically significant except MCV, fasting blood sugar, clotting time, basophils, Sr. TBil, serum indirect bilirubin, serum glutamic-pyruvic transaminase (SGPT), Sr. Tri, and serum VLDL.

**Table 2: Observations of hematological parameters in Group A (Wilcoxon rank-sum test, instant oleation)**

Parameter	Group	N	Median	25%	75%	P value
M	BI	15	2	2	3.75	0.313
	AI	15	2	2	3	
B	BI	15	0	0	0	1.000
	AI	15	0	0	0	
E	BI	15	2	2	2	1.000
	AI	15	2	2	2	
MCV	BI	15	80	72.5	86	0.083
	AI	15	80	71.75	85.5	
MCHC	BI	15	33.3	30.425	34.3	0.583
	AI	15	33.1	30.75	34.875	
Bl. Ur.	BI	15	18	18	21	0.119
	AI	15	20	18.25	22	
SGPT	BI	15	17	14.25	25	0.188
	AI	15	18	10	22	
Sr. Alb	BI	15	4.4	4.3	4.5	0.034
	AI	15	4.5	4.4	4.675	
Sr. Tri	BI	15	88	62	111.25	0.169
	AI	15	90	64	113.25	
Sr. VLDL	BI	15	17	12	21	0.049
	AI	15	19	12.25	23	

MCV = mean corpuscular volume, MCHC = mean corpuscular hemoglobin concentration, Bl Ur. = blood urea, SGPT = serum glutamic-pyruvic transaminase, Sr. Alb = serum albumin, Sr. Tri = serum triglyceride, Sr. VLDL = serum very low-density lipoproteins, BI = before intervention, AI = after intervention, M = monocyte, B = basophil, E = erythrocytes

**Table 3: Observations of hematological parameters in Group B (paired t test, incremental oleation)**

Parameter	Treatment name	N	Mean	Standard deviation	SEM	t value	P value
HB	BI	15	13.313	2.14	0.552	-0.289	= 0.777
	AI	15	13.373	1.921	0.496		
	Difference	15	-0.06	0.805	0.208		
TLC	BI	15	6800	1424.781	367.877	0.466	0.648
	AI	15	6633.333	1468.073	379.055		
	Difference	15	166.667	1384.437	357.46		
N	BI	15	58.533	6.046	1.561	-1.208	0.247
	AI	15	61.4	8.007	2.067		
	Difference	15	-2.867	9.187	2.372		
L	BI	15	34.467	5.317	1.373	0.787	0.444
	AI	15	32.8	7.58	1.957		
	Difference	15	1.667	8.2	2.117		
B	BI	15	3.4	2.501	0.646	0.000	1.000
	AI	15	3.4	1.882	0.486		
	Difference	15	0	2.591	0.669		
E	BI	15	3.267	2.282	0.589	0.837	0.417
	AI	15	2.933	1.668	0.431		
	Difference	15	0.333	1.543	0.398		
Total RBC	BI	15	4.611	0.676	0.175	-0.235	0.817
	AI	15	4.625	0.688	0.178		
	Difference	15	-0.014	0.23	0.0595		
ESR	BI	15	27	20.231	5.224	0.137	0.893
	AI	15	26.533	22.627	5.842		
	Difference	15	0.467	13.228	3.415		
MVC	BI	15	89.047	8.641	2.231	-1.237	0.236
	AI	15	90	7.819	2.019		
	Difference	15	-0.953	2.984	0.77		

**Table 3: Continued**

Parameter	Treatment name	N	Mean	Standard deviation	SEM	t value	P value
MCHC	BI	15	32.567	1.375	0.355	0.216	0.832
	AI	15	32.459	2.074	0.536		
	Difference	15	0.107	1.925	0.497		
BSL-F	BI	15	93.133	19.361	4.999	1.738	0.104
	AI	15	84.133	19.331	4.991		
	Difference	15	9	20.061	5.18		
Bl. Ur.	BI	15	22.533	8.798	2.272	2.484	0.026
	AI	15	18.333	5.551	1.433		
	Difference	15	4.2	6.549	1.691		
Sr. Cr.	BI	15	0.851	0.222	0.0573	-0.141	0.104
	AI	15	0.859	0.198	0.051		
	Difference	15	-0.00733	0.201	0.0519		
Sr. Na.	BI	15	139.533	2.031	0.524	-1.759	0.100
	AI	15	140.8	2.513	0.649		
	Difference	15	-1.267	2.789	0.72		
Sr. K	BI	15	4.14	0.25	0.0646	1.523	0.150
	AI	15	3.973	0.369	0.0954		
	Difference	15	0.167	0.424	0.109		
Sr. T Bil	BI	15	1.072	0.327	0.0845	0.642	0.531
	AI	15	1.02	0.489	0.126		
	Difference	15	0.052	0.314	0.081		
Sr. D Bil	BI	15	0.327	0.1	0.0258	2.943	0.011
	AI	15	0.244	0.0476	0.0123		
	Difference	15	0.0827	0.109	0.0281		
Sr. ID Bil	BI	15	0.763	0.303	0.0783	-0.163	0.873
	AI	15	0.776	0.473	0.122		
	Difference	15	-0.0133	0.317	0.0818		
SGOT	BI	15	26.8	7.163	1.85	0.595	0.561
	AI	15	25.667	8.845	2.284		
	Difference	15	1.133	7.376	1.905		
SGPT	BI	15	34.207	19.498	5.034	-0.227	0.823
	AI	15	35.067	17.862	4.612		
	Difference	15	-0.86	14.647	3.782		
Sr. Alk Phos.	BI	15	193.067	49.034	12.661	1.009	0.330
	AI	15	180.4	57.201	14.769		
	Difference	15	12.667	48.643	12.559		
Sr. Alb	BI	15	4.733	0.274	0.0708	0.871	0.398
	AI	15	4.673	0.281	0.0727		
	Difference	15	0.06	0.267	0.0689		
Sr. Glob	BI	15	3.06	0.374	0.0965	1.246	0.233
	AI	15	2.947	0.311	0.0804		
	Difference	15	0.113	0.352	0.091		
Sr. T Col	BI	15	184	31.332	8.09	-0.791	0.442
	AI	15	189.667	22.611	5.838		
	Difference	15	-5.667	27.748	7.165		
Sr. HDL	BI	15	39.133	9.628	2.486	1.903	0.078
	AI	15	33.533	5.449	1.407		
	Difference	15	5.6	11.394	2.942		
Sr. LDL	BI	15	120.267	33.824	8.733	-0.444	0.664
	AI	15	124.667	20.141	5.2		
	Difference	15	-4.4	38.344	9.9		
Sr. Tri	BI	15	122.6	57.152	14.757	-1.614	0.129
	AI	15	139.8	63.959	16.514		
	Difference	15	-17.2	41.271	10.656		



**Table 3: Continued**

Parameter	Treatment name	N	Mean	Standard deviation	SEM	t value	P value
Sr. VLDL	BI	15	22.267	10.872	2.807	-1.247	0.233
	AI	15	27.533	12.889	3.328		
	Difference	15	-5.267	16.36	4.224		
CT	BI	15	254	60.775	15.692	1.988	0.067
	AI	15	236.133	44.487	11.487		
	Difference	15	17.867	34.809	8.988		

SEM = standard error of mean, HB = hemoglobin, RBC = red blood cell, ESR = erythrocyte sedimentation rate, MCV = mean corpuscular volume, MCHC = mean corpuscular hemoglobin concentration, BSL-F = blood sugar level (fasting), BI Ur. = blood urea, Sr. Cr. = serum creatinine, Sr. Na = serum sodium, Sr. K = serum potassium, Sr. T Bil = serum total bilirubin, Sr. D Bil = serum direct bilirubin, Sr. ID Bil = serum indirect bilirubin, SGOT = serum glutamic-oxaloacetic transaminase, SGPT = serum glutamic-pyruvic transaminase, Sr. Alk Phos = serum alkaline phosphate, Sr. Alb = serum albumin, Sr. Glob = serum globulin, Sr. T Chol = total cholesterol, Sr. HDL = serum high-density lipoproteins, LDL = low-density lipoproteins, Sr. Tri = serum triglyceride, Sr. VLDL = serum very low-density lipoproteins, CT = clotting time, BI = before intervention, AI = after intervention N = neutrophil, L = leucocyte, M = monocyte, B = basophil, E = erythrocytes, TLC = total leukocyte count, MCV = mean corpuscular volume

**Table 4: Observations of hematological parameters in Group B (Wilcoxon rank sum test, incremental oleation)**

Parameter	Group	N	Median	25%	75%	t value	P value
M	BI	15	2	2	3	-29.000	0.148
	AI	15	2	1.25	2		
T. platelet	BI	15	257000	226000	292500	-45.000	0.421
	AI	15	270000	220000	348000		
MCH	BI	15	29.1	27.225	31.175		0.626
	AI	15	29	26.95	30.75		
BSL-PP	BI	15	110	102.25	131.75		0.121
	AI	15	110	100.25	122.25		
Sr. Pro T	BI	15	7.8	7.425	8.2		0.414
	AI	15	7.6	7.425	7.8		
BT	BI	15	60	56.25	120		0.055
	AI	15	55	48.25	87.5		

MCH = mean corpuscular hemoglobin, BSL-PP = blood sugar level (postprandial), Sr. Pro T = serum protein, BT = bleeding time, BI = before intervention, AI = after intervention, M = monocyte

**Table 5: Observations of hematological parameters in Group A (instant) and Group B (incremental) before oleation (unpaired t test)**

Parameter	Treatment name	N	Mean	Standard deviation	SEM	t value	P value
HB%	Group A	15	12.567	1.607	0.415	-1.081	0.289
	Group B	15	13.313	2.14	0.552		
TLC	Group A	15	6226.667	1084.611	280.045	-1.240	0.225
	Group B	15	6800	1424.781	367.877		
N	Group A	15	6226.667	1084.611	280.045	-0.750	0.460
	Group B	15	6800	1424.781	367.877		
L	Group A	15	37.6	6.045	1.561	1.507	0.143
	Group B	15	34.467	5.317	1.373		
Total RBC	Group A	15	4.753	0.691	0.178	0.569	0.574
	Group B	15	4.611	0.676	0.175		
MCH	Group A	15	25.9	5.487	1.417	-1.869	0.072
	Group B	15	29.013	3.396	0.877		
MCHC	Group A	15	32.44	2.224	0.574	-0.188	0.853
	Group B	15	32.567	1.375	0.355		
Sr. Na	Group A	15	139.533	2.924	0.755	0.000	1.000
	Group B	15	139.533	2.031	0.524		
Sr. Pro T	Group A	15	7.3	0.568	0.147	-2.546	0.017
	Group B	15	7.78	0.459	0.118		

**Table 5: Continued**

Parameter	Treatment name	N	Mean	Standard deviation	SEM	t value	P value
Sr. Alb	Group A	15	4.387	0.207	0.0533	-3.910	0.001
	Group B	15	4.733	0.274	0.0708		
Sr. LDL	Group A	15	134.2	41.679	10.762	1.005	0.323
	Group B	15	120.267	33.824	8.733		
Sr. Tri	Group A	15	92.6	33.655	8.69	-1.752	0.091
	Group B	15	122.6	57.152	14.757		
CT	Group A	15	253.533	53.38	13.783	-0.0223	0.982
	Group B	15	254	60.775	15.692		

SEM = standard error of mean, HB = hemoglobin, RBC = red blood cell, MCH = mean corpuscular hemoglobin, MCHC = mean corpuscular hemoglobin concentration, Sr. Na = serum sodium, Sr. Pro T = serum protein, Sr. Alb = serum albumin, Sr. LDL = serum low-density lipoproteins, Sr. Tri = serum triglyceride, CT = clotting time, N = neutrophil, L = leucocyte

**Table 6: Observations of hematological parameters in Group A (instant) and Group B (incremental) before oleation (Mann-Whitney rank sum test)**

Parameter	Group	N	Median	25%	75%	t value	P value
M	A	15	2	2	3.75	259.500	0.270
	B	15	2	2	3		
B	A	15	0	0	0	146.000	0.001
	B	15	3	2	5		
E	A	15	2	2	2	196.000	0.134
	B	15	2	2	4		
Total RBC	A	15	4.753	0.691	0.178	0.569	0.574
	B	15	4.611	0.676	0.175		
T. platelet	A	15	237000	199250	343750	225.000	0.772
	B	15	257000	226000	292500		
ESR	A	15	21	15	41.25	235.500	0.917
	B	15	28	9.25	32.25		
MCV	A	15	80	72.5	86	167.000	0.007
	B	15	89	85.75	91		
BSL-F	A	15	93	90.25	103.5	239.500	0.787
	B	15	93	79.5	105.75		
BSL-PP	A	15	106	103	111.5	209.000	0.340
	B	15	110	102.25	131.75		
Bl. Ur	A	15	18	18	21	195.500	0.130
	B	15	22	16.75	27.25		
Sr. Cr	A	15	0.86	0.713	0.915	240.000	0.772
	B	15	0.83	0.683	1		
Sr. K	A	15	4.03	3.925	4.35	219.500	0.604
	B	15	4.2	4	4.2		
Sr. T Bil	A	15	0.72	0.647	0.79	170.000	0.010
	B	15	1.06	0.843	1.21		
Sr. D Bil	A	15	0.26	0.203	0.362	207.500	0.309
	B	15	0.3	0.242	0.4		
Sr. ID Bil	A	15	0.48	0.425	0.665	179.500	0.029
	B	15	0.63	0.59	0.932		
SGOT	A	15	19	18.25	21.5	162.500	0.004
	B	15	25	22.25	32		
SGPT	A	15	17	14.25	25	178.000	0.025
	B	15	25	18.5	49.75		



**Table 6: Continued**

Parameter	Group	N	Median	25%	75%	t value	P value
Sr. AlkPhos	A	15	170	126.25	194.75	200.500	0.191
	B	15	189	161.25	225.75		
Sr. Glob	A	15	2.7	2.6	3.35	202.000	0.213
	B	15	3	2.725	3.45		
Sr. T Cholesterol	A	15	195	128.25	223.5	238.500	0.820
	B	15	191	171	208.5		
Sr. HDL	A	15	31	28	40.5	194.000	0.115
	B	15	37	32.25	41.5		
Sr. VLDL	A	15	17	12	21	204.500	0.254
	B	15	18	14.25	29.75		
BT	A	15	75	60	119.5	239.000	0.803
	B	15	60	56.25	120		

RBC = red blood cell, ESR = erythrocyte sedimentation rate, MCV = mean corpuscular volume, BSL-F = blood sugar level (fasting), BSL-PP = blood sugar level (postprandial), BI Ur. = blood urea, Sr. Cr. = serum creatinine, Sr. K = serum potassium, Sr. T Bil = serum total bilirubin, Sr. D Bil = serum direct bilirubin, Sr. ID Bil = serum indirect bilirubin, SGOT = serum glutamic-oxaloacetic transaminase, SGPT = serum glutamic-pyruvic transaminase, Sr. Alk Phos = serum alkaline phosphate, Sr. Glob = serum globulin, Sr. T Cholesterol = total cholesterol, Sr. HDL = serum high-density lipoproteins, Sr. VLDL = serum very low-density lipoproteins, BT = bleeding time, M = monocyte, B = basophil, E = erythrocytes

**Table 7: Observations of hematological parameters in Group A (instant) and Group B (incremental) after oleation (unpaired t test)**

Parameter	Treatment name	N	Mean	Standard deviation	SEM	t value	P value
HB%	Group A	15	12.387	1.603	0.414	-1.527	0.138
	Group B	15	13.313	2.14	0.552		
N	Group A	15	59.867	6.278	1.621	-0.584	0.564
	Group B	15	61.4	8.007	2.067		
L	Group A	15	36.267	5.97	1.541	1.392	0.175
	Group B	15	32.8	7.58	1.957		
Total RBC	Group A	15	4.803	0.672	0.174	0.719	0.478
	Group B	15	4.625	0.688	0.178		
MCV	Group A	15	77.067	11.56	2.985	-3.589	0.001
	Group B	15	90	7.819	2.019		
MCH	Group A	15	25.8	5.343	1.38	-2.151	0.040
	Group B	15	29.167	2.86	0.739		
MCHC	Group A	15	33.113	2.658	0.686	0.751	0.459
	Group B	15	32.459	2.074	0.536		
BSL-F	Group A	15	96.667	13.663	3.528	2.051	0.050
	Group B	15	84.133	19.331	4.991		
Sr. Cr.	Group A	15	0.86	0.167	0.0432	0.0199	0.984
	Group B	15	0.859	0.198	0.051		
Sr. Na	Group A	15	139	3.185	0.822	-1.718	0.097
	Group B	15	140.8	2.513	0.649		
Sr. K	Group A	15	4.173	0.363	0.0938	1.495	0.146
	Group B	15	3.973	0.369	0.0954		
Sr. Pro. T	Group A	15	7.467	0.628	0.162	-0.691	0.495
	Group B	15	7.607	0.471	0.122		
Sr. Alb	Group A	15	4.493	0.255	0.0658	-1.836	0.077
	Group B	15	4.673	0.281	0.0727		
Sr. Glob	Group A	15	2.973	0.543	0.14	0.165	0.870
	Group B	15	2.947	0.311	0.0804		
CT	Group A	15	288.067	84.86	21.911	2.099	0.045
	Group B	15	236.133	44.487	11.487		

SEM = standard error of mean, HB = hemoglobin, RBC = red blood cell, MCV = mean corpuscular volume, MCH = mean corpuscular hemoglobin, MCHC = mean corpuscular hemoglobin concentration, BSL-F = blood sugar level (fasting), Sr. Cr. = serum creatinine, Sr. Na = serum sodium, Sr. K = serum potassium, Sr. Pro T = serum protein, Sr. Alb = serum albumin, Sr. Glob = serum globulin, CT = clotting time

**Table 8: Observations of hematological parameters in Group A (instant) and Group B (incremental) after oleation (Mann-Whitney rank-sum test)**

Parameter	Group	N	Median	25%	75%	t value	P value
TLC	A	15	6100	5725	6400	219.000	0.590
	B	15	6200	5750	7425		
M	A	15	2	2	3	270.500	0.118
	B	15	2	1.25	2		
B	A	15	0	0	0	139.000	<0.001
	B	15	4	2.25	5		
E	A	15	2	2	2	199.500	0.176
	B	15	2	2	3.75		
T platelet	A	15	234000	210500	342000	211.000	0.384
	B	15	270000	220000	348000		
ESR	A	15	18	9	45	226.500	0.820
	B	15	25	8.25	33.75		
BSL-PP	A	15	102	98.5	112	204.000	0.245
	B	15	110	100.25	122.25		
Bl. Ur.	A	15	20	18.25	22	260.500	0.254
	B	15	18	15	21.5		
Sr. T Bil	A	15	0.72	0.597	0.875	179.500	0.029
	B	15	0.9	0.748	0.995		
Sr. D Bil	A	15	0.25	0.212	0.3	257.000	0.319
	B	15	0.25	0.203	0.29		
Sr. ID Bil	A	15	0.5	0.345	0.642	183.000	0.042
	B	15	0.63	0.485	0.777		
SGOT	A	15	19	19	24.5	204.500	0.254
	B	15	24	17.75	31.5		
SGPT	A	15	18	10	22	162.000	0.004
	B	15	30	19.25	49		
Sr. Alk Phos	A	15	178	138.5	208.25	237.000	0.868
	B	15	175	137.25	192.25		
Sr. T Chol	A	15	197	145	236.25	232.500	0.983
	B	15	190	174	208.5		
Sr. HDL	A	15	37	25	47.5	248.500	0.520
	B	15	32	30	36		
Sr. LDL	A	15	135	97	157	250.500	0.468
	B	15	125	110.25	132		
Sr. Tri	A	15	90	64	113.25	174.000	0.016
	B	15	116	96.25	179		
Sr. VLDL	A	15	19	12.25	23	182.000	0.038
	B	15	22	19.5	35.75		
BT	A	15	65	56.25	78.75	248.500	0.520
	B	15	55	48.25	87.5		

ESR = erythrocyte sedimentation rate, BSL-PP = blood sugar level (postprandial), Bl Ur. = blood urea, Sr. T Bil = serum total bilirubin, Sr. D Bil = serum direct bilirubin, Sr. ID Bil = serum indirect bilirubin, SGOT = serum glutamic-oxaloacetic transaminase, SGPT = serum glutamic-pyruvic transaminase, Sr. AlkPhos = serum alkaline phosphate, Sr. T Chol = total cholesterol, Sr. HDL = serum high-density lipoproteins, Sr. LDL = serum low-density lipoproteins, Sr. Tri = serum triglyceride, Sr. VLDL = serum very low-density lipoproteins, BT = bleeding time, M = monocyte, B = basophil, E = erythrocytes

## DISCUSSION

BT in Group A reduced significantly by 14.82% because saturated fat and cholesterol-rich diet impact hemodynamic parameters, including plasma and blood viscosity, plasma triglycerides, and red blood cells deformability, which are associated with increasing the risk of circulatory disorders.<sup>[7]</sup> Fat increases blood viscosity due to a rise in hematocrit.<sup>[8]</sup> As well, Sr. Alb

also increases in within range; Thalacker-Mercer *et al.*<sup>[9]</sup> mentioned that hepatic albumin synthesis is suppressed during an extended fasting period and is stimulated with nutrient ingestion and in Group A, bolus dose of 150 mL cow ghee was highly nutrient sufficient to increase value of albumin.

Serum VLDL and LDL are well known as “bad” cholesterols because they can contribute to the formation

of plaque in the arteries. But, after so much of fat consumption at once in Group A, VLDL increased by 11.76% within normal range ( $P < 0.05$ ) and that was might be not increased or decreased in Group B because of incremental dose pattern as well because of confounding physiological factors.<sup>[10]</sup>

In Group B, blood urea was significantly reduced by 17.75%, this might be because of high fat administered on empty stomach early in the morning, which converts amino acids into glucose. The process of deamination removes the amino groups from amino acids and urea is formed, which is passed through the blood to the kidney for excretion from the body.<sup>[11]</sup>

Serum direct bilirubin was significantly reduced by 25% in Group B ( $P < 0.05$ ). Bilirubin is the end product of heme catabolism and lipid-soluble waste product that needs to be excreted. Byoung<sup>[12]</sup> reported that a large number of healthy populations confirmed the inverse relationship between serum bilirubin level and the occurrence of nonalcoholic fatty liver disease.

While doing intergroup comparison after intervention, it was observed SGPT (Group A = 5.88% and Group B = 20%), Patell *et al.*<sup>[13]</sup> reported in study of non-alcoholic fatty liver disease that the mean levels of SGOT and SGPT in the two groups was found to be statistically significantly increased. So, this can happen because of intake of fat in large scale,<sup>[13]</sup> and serum VLDL (Group A = 11.76% and Group B = 22.22%) significantly increased within normal limit. VLDL particles are produced by the liver with triglyceride. They can vary depending on the quantity of triglyceride carried in the particle. When triglyceride production in the liver is increased, VLDL particles are larger in size. Whether, Sr. Tri (Group A = 2.17% and Group B = 5.38%) significantly decreased within normal range ( $P < 0.05$ ). Because lipids, such as cholesterol and triglycerides, are basically insoluble in water, these must be transported in association with proteins in the circulation.<sup>[14]</sup> The reason behind this physiologic variation is not understandable and it needs more investigation and analysis.

### Limitations

This study had some limitations, as larger scale study needs to be carried out to get more accurate picture of instant and incremental oleation effect on hematological and biochemical parameters. It is not clear that what type of confounding factors affecting hematological and biochemical changes in the body and responsible for physiological changes due to administration of cow ghee in large amount. On the basis of this pilot study result, further clinical trial studies need to be conducted on large sample size with controlled physiological conditions, if possible, will be required in future.

### Interpretation and Conclusion

In this pilot study, it was expected to find the effect of oleation (instant and incremental) on hematological and biochemical parameters. The effectiveness of both modalities not likely to be assessed in small sample size. There were some significant results found in BT, neutrophil, Sr. Alb, and VLDL in Group A and blood urea and serum bilirubin in Group B. Whereas in both group, total bilirubin, Sr. Tri, SGPT, and VLDL, significant changes ( $P < 0.05$ ) were observed within the normal limit. Though, it was concluded that both instant and incremental oleation modalities had some effect on hematological and biochemical parameters, but extension study will be needed for more practical clarification. All the changes increased or decreased in normal limit; hence, the said intervention is safe for the treatment purpose in the field of Ayurveda. This study aimed to develop and evaluate the feasibility and acceptability of treatment method of Ayurveda.

### Ethical clearance

Ethical clearance for this study was obtained (Ref. no. DMIMS (DU)/IEC/2017–18/6357).

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Nil.

### Conflicts of interest

There are no conflicts of interest.

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