

**Original Research Article****HEALING ROLE OF *GUDUCHI* [*Tinospora cordifolia* (Willd.) Miers] AND *AMALAKI* (*Emblica officinalis* Gaertn.) CAPSUL IN PREMATURE AGING DUE TO STRESS: A COMPARATIVE OPEN CLINICAL TRIAL****ABSTRACT**

**Aims:** To find out the comparative effects of the capsules prepared from the drugs *Guduchi* [*Tinospora cordifolia* (Willd.) Miers] and *Amalaki* (*Emblica officinalis* Gaertn.) in cases of premature aging due to stress.

**Study design:** Comparative open clinical trial

**Place and Duration of Study:** IPD and OPD of National Institute of Ayurveda and Seth Soorajmal Bombawala Hospital, Jaipur, Rajasthan, India, between June 2016 and April 2017.

**Methodology:** We included 30 patients (14 men, 16 women; age range 25-60 years) with premature aging due to stress, without any other acute or serious systemic disorders. They were randomly divided in three groups with 10 patients in each. Group A- was treated with capsule of *Guduchi* 1 gm/day, Group B- capsule of *Amalaki* 1 gm/day and Group C- was treated with 500mg capsules of *Guduchi* and *Amalaki* separately once daily for 3 months. Visual Analogue Scale and Stress were assessed by Hamilton Anxiety Rating Scale (HARS). Objective parameters like CBC, CRP, FBS, Blood Urea, creatinine, SGOT, SGPT, Serum cholesterol were also analysed.

**Results:** Marked effect of therapy was observed in group C, where the significant changes ( $p < 0.05$ ) were observed on subjective parameters like Dizziness Constipation, Aching Muscles, Sleep abnormality, Loss of appetite, Fatigue, Generalized Weakness. In HARS scale significant changes ( $p < 0.01$ ) were found on Anxious Mood and Intellectual power, along with on Tension, Fears, Insomnia Depressed Mood and Gastrointestinal Symptoms statistically significant ( $p < 0.05$ ) changes were also observed. Similarly on objective parameters like Hb%, Eosinophils and TPLC significant effect ( $p < 0.05$ ) of therapy was observed. Along with marked significant ( $p < 0.01$ ) effect was observed in improvement of Neutrophil, Lymphocyte, TRBC, TPLC, PCV. But In ANOVA Test for Intergroup Comparison no significant changes ( $p > 0.05$ ) were found except on the parameters like abnormality in sleep, FBS and TLC where  $p$  value was found to be statistically significant ( $p < 0.05$ ).

**Conclusion:** Both the test drug, stem of *Guduchi* and dried fruit rinds of *Amalaki* were found to be significantly effective in premature ageing due to stress, but the effect was quantitatively better in Group C (*Guduchi* and *Amalaki*). However these predictors need further work to validate reliability.

**Keywords:** Premature aging, Stress, Amalaki, Guduchi, Herbal, Ayurveda

**1. INTRODUCTION**

*Ayurveda* is not only a system of medicine rather a manner of life. According to *Acharya charaka* life is the blend of body, senses, mind and reincarnating soul<sup>[1]</sup>. The age is a factor dependent on *kala pramana vishesha* i.e. quantum of time duration<sup>[2]</sup>. *Acharya sushruta* has mentioned a group of naturally occurring diseases named *svabhava bala roga*, which includes *kshudha* (hunger), *pipasa* (thirst), *nidra* (sleep), *jara* (aging) and *mrityu* (death). The last phase of life span has been referred as *jara* (aging) which is described as a natural and inevitable processes as well as natural disease.<sup>[3]</sup> It is a continuous process which begins with conception and end with death. In ancient *Ayurveda* classics, *Jara* has been categorized in two heading-*kalaja* (irreversible) and *akalaja* (reversible). *Kalaja* is a natural phenomenon,

20 which stems from inherited potential, but *akalaja* (reversible) is premature ageing may be triggered due to  
21 physical and mental stress.<sup>[4]</sup> According to latest health report stress is now becoming more accepted as  
22 being crucially related to our total physical, mental and spiritual health. Various stresses lead to  
23 disturbance in the homeostasis of both the body and mind by vitiating *manasdosha* (mental humor),  
24 *shariradosha* (body humor) and *agni* (conflagration of heartiness)<sup>[5]</sup>. Thus stressful environment and  
25 disturbance in *manasika bhava* has adversely affected the healthy life style and that gives rise to the  
26 symptoms of aging before the time<sup>[6]</sup>. A revolution of life style, changing family structure, economic crises  
27 and social problems, are major stress inducer thrashing developing countries. Various stressors leads to  
28 disturbance in *manasabhava* (emotions), described in *Ayurveda*, are root cause of many diseases  
29 including premature ageing<sup>[7]</sup>. The *kama* (lust), *krodha* (anger), *lobha* (greed), *moha* (delusion), *irsha*  
30 (jealousy), *shoka* (grief), *chinta* (anxiety), fear (*bhaya*) etc. are different *manasabhava*<sup>[8]</sup>. The nerve-  
31 racking atmosphere and annoyance of these *manasabhava* adversely affects the health which ultimately  
32 contributes to premature ageing.

33 The drug *Guduchi* botanically identified as *Tinospora cordifolia* (Willd.) Miers belongs to Menispermaceae  
34 family and the drug *Amalaki* botanically identified as *Emblica officinalis* Gaertn. belongs to family  
35 Euphobiaceae are widely available throughout tropical and sub-tropical region of India<sup>[9]</sup>, also frequently  
36 used by the *Ayurveda* physicians in the condition of premature ageing. Both the drugs are mentioned as  
37 *vrishtya* (aphrodisiac), *medhya* (promote intellectual properties of brain), *valya* (enhance vitality), *rasayana*  
38 (rejuvenative and immunomodulator) and *vayahsthapana* (anti aging) in ancient *Ayurveda* texts.<sup>[10,11]</sup>  
39 Various pharmacological studies also finds out their significant effects in different areas like  
40 immunomodulator, adaptogenic, anti oxidant, anti stress, anti microbial and anti allergic etc. activities<sup>[12,13]</sup>.  
41 On this background the present study was under taken to find out comparative effects of the capsules  
42 prepared from the drug *Guduchi* and *Amalaki* in the premature aging due to stress.

## 43 2. MATERIAL AND METHODS

44 **2.1. Collection of Drugs:** Stem of *Guduchi* [*Tinospora cordifolia* (Willd.) Miers] and Fruits of *Amalaki*  
45 (*Emblica officinalis* Gaertn.) were collected from the natural sources and the materials were identified and  
46 authenticated by the Department of Botany, Rajasthan University, with voucher specimen no.  
47 RUBL211612 (*Guduchi*) and RUBL211616 (*Amalaki*).

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49 **2.2. Method of Preparation of Trial Drugs:** *Ghana* (concentrated decoction) were prepared separately  
50 from both the selected drug i.e stem of *Guduchi* and fruits of *Amalaki*, as per the method mentioned in  
51 *Sharangadhara Samhita*<sup>[14]</sup>. Then both the decoctions were again boiled until water content was  
52 evaporated to make in powdered form. Then the 500 mg of powder were filled in each gelatine capsule  
53 (with the help of Automatic Capsule filling instrument) separately for both the samples.

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55 **2.3. Ethical Clearance:** Present clinical trial was done after getting the ethical approval from Institutional  
56 Ethical Committee of National Institute of Ayurveda, Jaipur with approval no. IEA/ACA/2015/29.

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58 **2.4. Selection of study participants:** 30 Patients having the sign and symptoms of premature aging due  
59 to stress were selected from OPD and IPD of National Institute of Ayurveda and Seth Soorajmal  
60 Bombawala Hospital, Jaipur, Rajasthan, India. They were randomly divided in three groups with 10  
61 patients in each group.

### 62 2.4.1. Inclusion criteria:

- 63 • Patients having the signs and symptoms of premature aging due to stress were selected.
- 64 • Patients of either sex with the age group between 25- 60 years were included.

### 65 2.4.2. Exclusion criteria:

- 66 • Patients suffering from chronic diseases like severe Hypertension, IHD, COPD, DM, Cancer,  
67 hepatic and renal insufficiency and psychotic disorders like depression, schizophrenia.
- 68 • Patients suffering from any other acute or serious illness.

### 69 2.4.3. Withdrawal criteria:

- 70 • Patients who may develop any adverse drug reaction due to the trial drugs.
- 71 • Non complaints of the patients.

- 73 • Any Serious Intercurrent Illness  
74

75 **2.5. Method of study:** The study was carried out as per International conference of Harmonization-Good  
76 Clinical Practices Guidelines (ICH-GCP). Written informed consent was taken on prescribed Proforma  
77 from each patient willing to participate before the start of study. They were briefed about merits and  
78 demerits of research plan before taking the consent. Patients were free to withdrawal from the study at  
79 any time without giving any reason. A detailed Proforma was prepared incorporating *Ayurveda* as well as  
80 modern points. Observations were made according to the standard *Ayurveda* parameters selected and  
81 findings were recorded in well-designed Proforma.  
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83 **2.5.1. Grouping and administration of drug:** 30 clinically diagnosed and registered patients of  
84 premature aging due to stress were selected based on inclusion and exclusion criteria. They were divided  
85 randomly into 3 groups, each group was 10 patients.

86 **Group A-** 500 mg soft Gelatine Capsules of *Guduchi* were given for 3 months, 1 gm/ day in two divided  
87 doses.

88 **Group B-**500 mg soft Gelatine Capsules of *Amalaki* were given for 3 months, 1 gm/ day in two divided  
89 doses.

90 **Group C-** 500 mg/day capsule of *Guduchi* and 500 mg/day capsule *Amalaki* were given for 3 month,  
91 once daily in the form of soft Gelatine capsule.  
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93 **2.5.2 Criteria for assessment:** The assessments of the patients were done based on subjective as well  
94 as objective criteria during the course of trial treatment. The final assessment was done on the basis of all  
95 the parameters and by comparing the laboratorial investigation before and after the treatment.

96 **2.5.3. Subjective criteria:** Visual Analogue Scale and Stress were assessed by Hamilton Anxiety Rating  
97 Scale (HARS) <sup>[15]</sup>

98 **2.5.4. Objective criteria:** CBC, CRP, F.B.S, RFT- Blood Urea, Serum creatinine, SGOT, SGPT, Serum  
99 cholesterol.  
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101 **2.5.5. Statistical analysis:** Graph Pad prism-7 software was used for analysis of the data obtained from  
102 the study. For Non parametric Data Wilcoxon matched-pairs signed ranks test and for intergroup  
103 comparison Kruskal- Wallis multiple comparison tests (Dunn's multiple comparison) was used. While for  
104 Parametric Data Paired 't' Test and for intergroup comparison ANOVA test was used. Subjective  
105 parameters were assessed by the research team as per established grading system.  
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### 107 3. RESULTS AND DISCUSSION

#### 108 3.1. Effect on Subjective parameters:

109 Statistically significant results were observed in parameters like Dizziness, Constipation, Aching Muscles,  
110 Sleep abnormality, Loss of appetite, Fatigue and Generalized Weakness observed in group A (Table-1),  
111 group B (Table-4) and group C (Table-7). Maximum percentage of relief was observed in group C in all  
112 these parameters (Table-1, 4, 7 and Graph-1).  
113

#### 114 3.2. Effect of Therapy on HARS:

115 In HARS scale statistically significant results were observed on the parameters like Anxious Mood,  
116 Tension, Fears, Intellectual and Gastrointestinal Symptoms: in group A (Table-2), group B (Table-5) and  
117 group C (Table-8) where as in parameters like Respiratory Symptoms, Behaviour at interview,  
118 Genitourinary symptoms, Autonomic Symptoms, Somatic (Sensory) no statistically significant changes  
119 were observed. Here also maximum response was observed in group C in comparison to group A and B.  
120 In the parameters like Insomnia and Depressed Mood Maximum percentage of relief was observed in  
121 group B and group C and in Somatic (Muscular) maximum percentage of relief was observed in group B  
122 (Table-2, 5, 8 and Graph-2).  
123

#### 124 3.3. Effect on Objective parameters:

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126 On objective parameters like Hb%, FBS statistically significant results were observed in group A (Table-  
 127 3), group B (Table-6) and group C (Table-9). Maximum percentage of relief was observed in group A. On  
 128 others parameters like Blood Urea and serum SGOT maximum percentage of relief was observed in  
 129 group B. Whereas in group A shows maximum percentage of relief on the parameters SGPT and serum  
 130 cholesterol and on Serum Creatinine maximum percentage of relief was observed in group C. Although  
 131 all these changes were found to be statistically insignificant ( $p>0.05$ ) (Table-3, 6, 9 and Graph-3).

132 But In ANOVA Test for Intergroup Comparison no significant changes ( $p>0.05$ ) were found  
 133 except on the parameters like abnormality in sleep, FBS and TLC where  $p$  value was found to be  
 134 statistically significant ( $p<0.05$ ) (Table-10, 11, 12 and Graph-1, 2, 3).

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### 136 3.4. Discussion on probable mode of action of drug:

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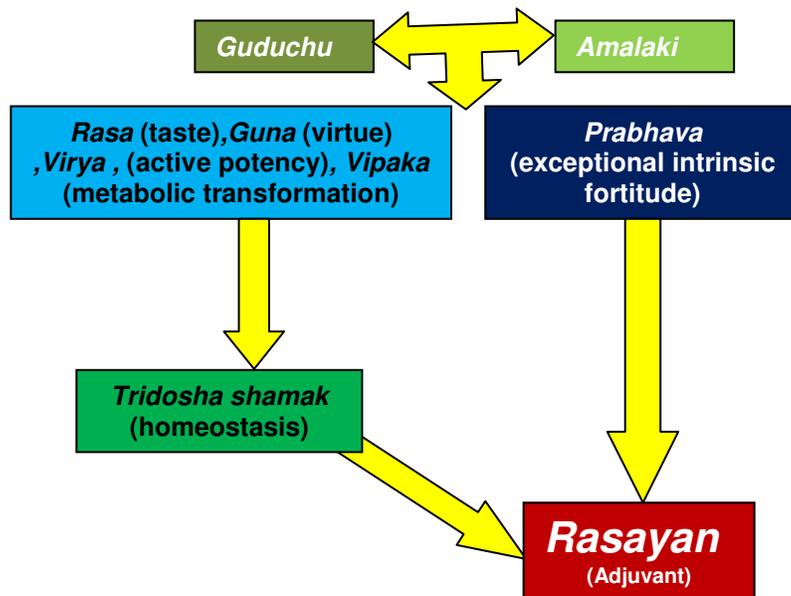
138 As per the fundamental principle of *Ayurveda* pharmacology a *dravya* (drug) performs *karma* (certain  
 139 actions) in the body by virtue of its properties i.e. *guna* (virtue), *rasa* (taste), *virya* (active potency), *vipaka*  
 140 (metabolic transformation) and *prabhava* (exceptional intrinsic fortitude) which exist in it in a state of co-  
 141 inherence<sup>[16]</sup>. The uniformity of proto-elements of the drug on one hand and the proto-elemental  
 142 constituents of the body on the other hand forms the basis of the principles of *samanya vishesha*  
 143 (egalitarianism and discrepancy)<sup>[17]</sup>. These principles imply that the predominant proto-elements of the  
 144 drug will increase similar proto-elements in the body and the dissimilar will decrease the proto-elements.  
 145 The actions of a drug are intimately related to its chemical structure in the form of preponderance of one  
 146 or two proto-elements in them. Ancient scholars have advised to use *rasayana* for longevity of life  
 147 (*ayuanuvruti, vayasthapana*) and for achieving the optimum effect of normal function of *tridosha*  
 148 (physiological humors)<sup>[18]</sup>. The drug *Guduchi* possess *guru, snigdha guna* (heavy and unctuous virtue),  
 149 *tikta, kashaya rasa* (bitter and astringent taste) *madhura vipaka* (sweet metabolic transformation), *ushna*  
 150 *virya* (hot active potency)<sup>[19]</sup>. The qualities of *guru* and *snigdha* are nutritive in nature, being similar in  
 151 quality to *rasadhatu* it enhances and strengthens *rasadhatu* establishing solid grounding for the six  
 152 remaining *dhatu* (basic physiological structure of body). Being *bitter taste* it pacifies *pittadosha*, while  
 153 *astringent* as a secondary taste it also balances *kapha*. *Vata* is balanced by the *Guru* and *snigdha*  
 154 qualities that counter the dry and light qualities of *Vata*. It's hot potency not only stimulates but also  
 155 correct digestive fire, and digest *amadosh*a (undigested food materials)<sup>[20]</sup>. The presence of the two  
 156 *Guna, guru* and *snigdha* and the post-digestive action as *madhura vipaka* indicates that the action of the  
 157 drug is more anabolic rather than catabolic in nature and from an energetic perspective it counters the  
 158 catabolic nature of aging.

159 According to *Ayurveda*, *Amalaki* balances all three *dosha*. While *Amalaki* is unusual in that it contains five  
 160 out of the six tastes recognized by ancient *Ayurveda* sages, it is most important to recognize the effects of  
 161 the "*virya*", or potency, and "*Vipaka*", or post-digestive effect. Considered in this light, *Amalaki* is  
 162 particularly helpful in reducing *Pitta* due to its *shita guna* (arctic virtue). It also balances both *Pitta* and  
 163 *Vata* by virtue of its sweet taste. The *kapha* is balanced primarily due to its *ruksh guna* (seared virtue)<sup>[21]</sup>.  
 164 It act as a *rasayana* (rejuvenate) to promote longevity and traditionally to increase *dipanapachana*  
 165 (digestion and metabolism), *raktaprasadana* (purify the blood), *romasanjana* (stimulate hair growth),  
 166 *jivaniya* (enliven the body) and *medhya* (enhance intellect)<sup>[22]</sup>. It ultimately brings out best quality of *dhatu*  
 167 and slows down the ageing process by generating new cells, antioxidant, anti-atherosclerotic,  
 168 immunomodulation, free radical scavenging activity, anti-hepatotoxic, adrenergic potentiating, etc<sup>[23]</sup>.

169 Mental health also plays a vital role in health, disease and premature ageing. *Acharya Charaka* has  
 170 mentioned that psychological factors cause bodily disorders and vice versa. Again *Charaka* mentioned  
 171 that keeping body and mind under control, following moral code of conducts and living spiritual life would  
 172 itself bring the *rasayan* effects and prevent ageing<sup>[24]</sup>. Both the test drug *Guduchi* and *Amalaki* have effect  
 173 on subjective parameters and on parameters of HARS either given single or in combination. While  
 174 normalising the physiology of the body by pacifying *tridosha*, correcting digestion and metabolism and  
 175 nourishing the *sapta dhatu* in proper way, both drugs release *medhya* effect which correct mental health  
 176 and stress condition of patient. In an experimental study on *Guduchi* it is proved to have antistress and  
 177 adaptogenic activity<sup>[25]</sup>. An ethanolic extract of the roots of *Tinospora cordifolia* normalized stress-induced  
 178 biochemical changes in norepinephrine Anti-stress activity<sup>[26,27]</sup>. *Guduchi* has been claimed to possess  
 179 anti-stress activity<sup>[28,29]</sup>. Ethanolic extract of *Embellica officinalis* has significant antistress and adaptogenic

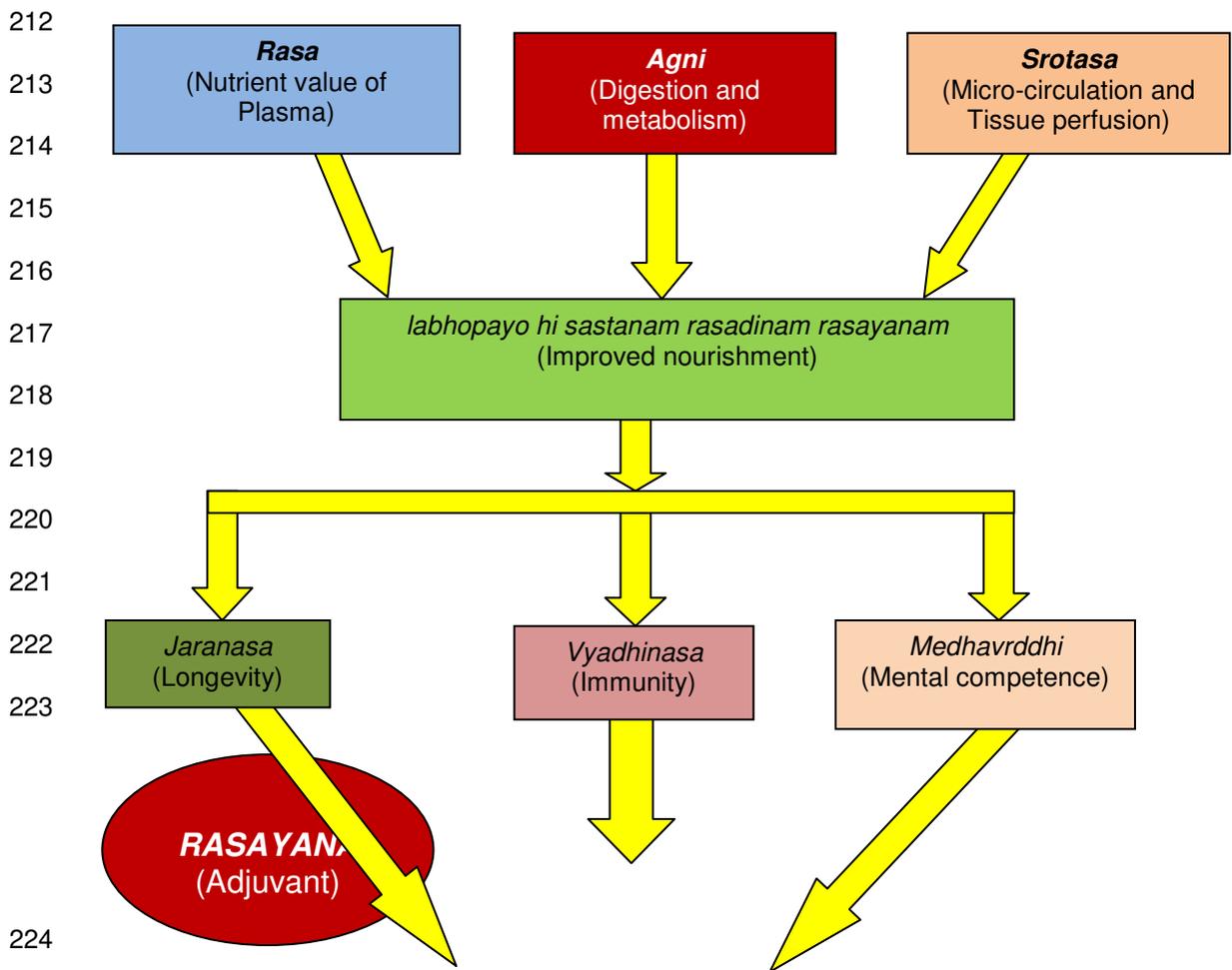
180 activity against variety of biochemical and physiological perturbations<sup>[30]</sup>. It is also claimed as memory  
 181 enhancing, antioxidant and anti-cholinesterase activity. It may be useful for the treatment of cognitive  
 182 impairments induced by cholinergic dysfunction<sup>[31]</sup>. In other similar clinical study on *Amlakyadi rasayana*,  
 183 significant improvement was reported in *medha* and anxiety scale<sup>[32]</sup>. On the basis of above facts it can be  
 184 inferred that *Guduchi* and *Amalaki* both drugs having having *medhya*, *rasayana* and *vayahsthapana*  
 185 property. These may be the reasons both drugs provide satisfactory results either single use or in  
 186 combination (Flow chart-1, 2).

187 **Flow chart-1, Rasayana effect of Guduchi and Amalaki:**



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211 **Flow chart-2 Rasayana effect of Guduchi and Amalaki to prevent premature aging:**



225 **Table 1. effect of therapy on subjective parameters in group A:**

Group A	BT	AT	Diff	% of Relief	SD	SEM	(-w)	P value	Sig
Dizziness	1.2	0.3	0.9	75	0.994	0.314	21	0.0313	S
Constipation	0.5	0.3	0.2	40	0.421	0.1333	3	0.5	NS
Aching Muscles	1.3	0.7	0.6	46.15	0.516	0.163	21	0.0313	S
Joint Pain	1.2	0.7	0.5	41.66	0.527	0.166	15	0.0625	NS
Joint Stiffness	1.1	0.6	0.5	45.45	0.527	0.166	15	0.0625	NS
Sleep Abnormality	1.6	1.1	0.5	31.25	0.527	0.166	15	0.0625	NS
Loss of appetite	2.2	1.2	1	45.45	0.471	0.1490	45	0.0039	S
Fatigue	2.7	1.6	1.1	40.74	0.737	0.233	36	0.0078	S

Gen. Weakness	2.5	1.1	1.4	56	0.516	0.163	55	0.0029	S
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226 \* BT-mean of before treatment, AT-mean of after treatment, Diff.-mean difference, SD-standard deviation,  
 227 SEM-Standard Error of Mean, Sig.-significance level.

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**Table 2. effect of therapy on HARS subjective parameters in group A:**

GROUP A	BT	AT	Diff	% of Relief	SD	SEM	(-W)	P value	Sig
Anxious Mood	2.2	1.3	0.9	40.90	0.56	0.17	36	0.0078	S
Tension	2.8	1.6	1.2	42.85	0.42	0.13	55	0.002	S
Fears	2	1.1	0.9	45	0.87	0.27	21	0.0313	S
Insomnia	1.7	1.3	0.4	23.52	0.51	0.16	10	0.213	NS
Intellectual	1.7	0.8	0.9	52.94	0.31	0.1	45	0.0039	S
Depressed Mood	1.4	0.8	0.6	42.85	0.51	0.16	21	0.0313	S
Somatic muscular	1.2	0.9	0.3	25	0.48	0.15	6	0.2500	NS
Somatic (Sensory)	0.7	0.3	0.4	57.14	0.69	0.22	10	0.125	NS
CVS	0	0	0	0	0	0	0		
Respiratory Symptoms	0.5	0.4	0.1	20	0.31	0.1	1	>0.9999	NS
Gastrointestinal Symptoms	0.6	0.3	0.3	50	0.48	0.15	1	>0.9999	NS
Behaviour at interview	0.6	0.3	0.3	50	0.48	0.15	15	0.0625	NS
Genitourinary symptoms	0.6	0.4	0.2	33.33	0.42	0.13	1	>0.9999	NS

230 \* BT-mean of before treatment, AT-mean of after treatment, Diff.-mean difference, SD-standard deviation,  
 231 SEM-Standard Error of Mean, Sig.-significance level.

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**Table 3. effect of therapy on objective parameters in Group A:**

Group A	BT	AT	Diff	% of Relief	SD	SEM	P value	Sig
HB	13.2	13.64	0.74	5.61	3.708	1.173	0.0207	S
TLC	5540	5760	220	3.97	293.636	92.856	0.0420	S
Neutrophil	55	57.1	2.1	3.82	3.604	1.140	0.0985	NS
Lymphocytes	35.6	38.9	3.3	9.27	2.830	0.895	0.0050	S

Eosinophils	4.2	2.8	1.4	33.33	1.647	0.521	0.0248	S
Monocytes	4.9	5.3	0.4	8.16	1.075	0.340	0.2695	NS
Basophils	0	0	0	0.00	0.000	0.000	0.0000	NS
TRBC	4.83	5.075	0.24	5.07	0.495	0.156	0.1517	NS
TPLC	2.013	2.074	0.06	3.03	0.449	0.142	0.6773	NS
PCV	43.5	42.44	1.05	2.43	2.025	0.640	0.1339	NS
MCV	91.5	90.15	1.38	1.51	2.907	0.919	0.1662	NS
MCH	27.54	26.809	0.731	2.65	1.121	0.355	0.0693	NS
MCHC	30.46	30.117	0.343	1.13	0.556	0.176	0.0830	NS
CRP	0	0	0	0.00	0.000	0.000	0.0000	NS
FBS	96	87.4	8.6	8.96	11.443	3.618	0.0415	S
Blood Urea	32.3	31.4	0.9	2.79	1.912	0.605	0.1708	NS
SR. Creatinine	0.93	0.9	0.03	3.23	0.106	0.033	0.3938	NS
SGOT	41	38.5	2.5	6.10	3.100	0.980	0.0312	NS
SGPT	28.1	27	1.1	3.91	2.283	0.722	0.1619	NS
Sr. Cholesterol	176.5	168.3	8.2	4.65	14.793	4.678	0.1135	NS

235 \* BT-mean of before treatment, AT-mean of after treatment, Diff.-mean difference, SD-standard deviation,  
 236 SEM-Standard Error of Mean, Sig.-significance level.  
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238 **Table 4. effect of therapy on subjective parameters in group B**  
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Group B	BT	AT	Diff	% of Relief	SD	SEM	(-W)	P value	Sig
Dizziness	0.5	0.1	0.4	80	0.516	0.163	21	0.031	S
Constipation	0.7	0.3	0.4	57.14	0.516	0.163	10	0.125	NS
Aching Muscles	3.3	0	2.1	63.63	2.024	0.640	21	0.0313	S
Joint Pain	4.5	3.9	0.6	13.33	0.699	0.221	15	0.0625	NS
Joint Stiffness	2.1	1.8	0.3	14.28	0.483	0.152	6	0.25	NS
sleep abnormality	2.3	1.9	0.4	17.39	1.264	0.4	1	>0.9999	NS
Loss of appetite	2.7	1.5	1.2	44.44	1.229	0.388	21	0.0313	S

Fatigue	2.8	1.3	1.5	53.57	1.840	0.582	21	0.0313	S
Gen. Weakness	4.2	3.5	0.7	16.66	0.674	0.213	21	0.0313	S

240 \* BT-mean of before treatment, AT-mean of after treatment, Diff.-mean difference, SD-standard deviation,  
 241 SEM-Standard Error of Mean, Sig.-significance level.  
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244 **Table 5. effect of therapy on HARS subjective parameters in Group B**

Group B	BT	AT	Diff	% of Relief	SD	SEM	(-W)	P value	Sig
Anxious Mood	1.6	1	0.6	37.5	0.51	0.16	21	0.0313	S
Tension	1.4	0.7	0.7	50	0.67	0.21	21	0.0313	S
Fears	1.3	1	0.3	23.07	0.48	0.15	6	0.25	NS
Insomnia	1.4	0.7	0.7	50	0.67	0.21	21	0.0313	S
Intellectual	1.5	0.7	0.8	53.33	0.78	0.24	21	0.0313	S
Depressed Mood	1.3	0.8	0.5	38.46	0.52	0.16	15	0.0625	NS
Somatic (Muscular)	1.2	0.8	0.4	33.33	0.51	0.16	10	0.125	NS
Respiratory Symptoms	0.6	0.3	0.3	50	0.48	0.15	6	0.25	NS
Gastrointestinal Symptoms	0.8	0.4	0.4	50	0.51	0.16	3	0.5	S

245 \* BT-mean of before treatment, AT-mean of after treatment, Diff.-mean difference, SD-standard deviation,  
 246 SEM-Standard Error of Mean, Sig.-significance level.  
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**Table 6. effect of therapy on Objective parameters in Group B**

Group B	BT	AT	Diff	% of Relief	SD	SEM	P value	Sig
HB	12.595	12.792	0.197	1.56	0.208	0.066	0.0152	S
TLC	6220	5960	260	4.18	411.501	130.128	0.0768	NS
Neutrophil	62.3	60.7	1.6	2.57	2.914	0.921	0.1165	NS
Lymphocytes	29	26.8	2.2	7.59	3.155	0.998	0.0549	NS
Eosinophils	4.2	3.4	0.8	19.05	1.317	0.416	0.0868	NS
Monocytes	4.5	4	0.5	11.11	0.850	0.269	0.0957	NS
Basophils	0	0	0	0.00	0.000	0.000	0.0000	NS

TRBC	4.463	4.535	0.072	1.61	0.412	0.130	0.5938	NS
TPLC	1.911	1.93	0.019	0.99	0.149	0.047	0.6962	NS
PCV	41.49	41.414	0.076	0.18	0.521	0.165	0.6556	NS
MCV	93.17	92.97	0.2	0.21	0.320	0.101	0.0793	NS
MCH	28.49	28.113	0.377	1.32	0.642	0.203	0.0964	NS
MCHC	30.49	29.925	0.565	1.85	0.865	0.274	0.0690	NS
CRP	0	0	0	0.00	0.000	0.000	0.0000	NS
FBS	98.7	96.7	2	2.03	3.162	1.000	0.0766	NS
Blood Urea	30.7	28.6	2.1	6.84	3.107	0.983	0.0613	NS
SR. Creatinine	0.98	0.84	0.14	14.29	0.201	0.064	0.0552	NS
SGOT	44.9	39.8	5.1	11.36	7.781	2.461	0.0681	NS
SGPT	32.1	32.9	0.8	2.49	4.077	1.289	0.5503	NS
Sr.Cholesterol	177.1	169	8.1	4.57	9.171	2.900	0.0209	NS

251 \* BT-mean of before treatment, AT-mean of after treatment, Diff.-mean difference, SD-standard deviation,  
 252 SEM-Standard Error of Mean, Sig.-significance level.  
 253

254 **Table 7.effect of therapy on subjective parameters in Group C**

<b>Group C</b>	<b>BT</b>	<b>AT</b>	<b>Diff</b>	<b>% of Relief</b>	<b>SD</b>	<b>SEM</b>	<b>(-w)</b>	<b>P value</b>	<b>Sig</b>
Dizziness	1.6	0.3	1.3	81.25	1.337	0.422	21	0.0313	S
Constipation	1.3	0.3	1	76.92	1.054	0.333	21	0.0313	S
Aching Muscles	2.5	1	1.5	60	1.649	0.521	21	0.0313	S
Joint Pain	2.5	2.1	0.4	16	0.516	0.163	10	0.125	NS
Joint Stiffness	2.2	1.9	0.3	13.63	0.483	0.152	6	0.25	NS
Sleep abnormality	2.7	0.7	2	74.07	1.943	0.614	21	0.0313	S
Loss of appetite	2.9	0.9	2	68.96	2.160	0.683	21	0.0313	S
Fatigue	2.3	1	1.3	56.52	1.159	0.366	21	0.0313	S
Gen. Weakness	2.3	1.1	1.2	52.17	1.135	0.359	21	0.0313	S

256 **Table 8. effect of therapy on HARS subjective parameters in group C**

Group C	BT	AT	Diff	% of Relief	SD	SEM	(-W)	P value	Sig
Anxious Mood	1.4	0.6	0.8	57.14	0.42	0.13	36	0.0078	S
Tension	1.1	0.4	0.7	63.63	0.48	0.15	28	0.0156	S
Fears	1.2	0.6	0.6	50	0.51	0.16	21	0.0313	S
Insomnia	1.6	0.8	0.8	50	0.91	0.29	21	0.0313	S
Intellectual	1	0.2	0.8	80	0.42	0.13	36	0.0078	S
Depressed Mood	1.6	0.9	0.7	43.75	0.67	0.21	21	0.0313	S
Respiratory Symptoms	1.2	0.9	0.3	25	0.48	0.15	6	0.2500	NS
Gastrointestinal Symptoms	1.4	0.7	0.7	50	0.48	0.15	28	0.0156	S
Autonomic Symptoms	1	0.8	0.2	20	0.42	0.13	3	0.5000	NS

257 \* BT-mean of before treatment, AT-mean of after treatment, Diff.-mean difference, SD-standard deviation,  
 258 SEM-Standard Error of Mean, Sig.-significance level.  
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**Table 9. effect of therapy on objective parameters in Group C**

Group C	BT	AT	Diff	% of Relief	SD	SEM	P value	Sig
HB	12.15	12.71	0.56	4.61	0.631	0.200	0.0205	S
TLC	6100	6150	50	0.82	271.825	85.959	0.5751	NS
Neutrophil	57.4	54.7	2.7	4.70	1.567	0.496	0.0004	S
Lymphocytes	35.3	33.6	1.7	4.82	1.160	0.367	0.0012	S
Eosinophils	2.6	2.2	0.4	15.38	0.516	0.163	0.0368	S
Monocytes	4.7	4.2	0.5	10.64	0.850	0.269	0.0957	NS
Basophils	0	0	0	0.00	0.000	0.000	0.0000	NS
TRBC	4.679	4.479	0.2	4.27	0.166	0.053	0.0042	S
TPLC	2.304	2.16	0.144	6.25	0.196	0.062	0.0449	S
PCV	41.31	40.974	0.336	0.81	0.235	0.074	0.0014	S

MCV	88.28	87.98	0.3	0.34	0.226	0.071	0.0023	S
MCH	26.85	26.76	0.09	0.34	0.197	0.062	0.1823	NS
MCHC	30.37	30.36	0.01	0.03	0.396	0.125	0.9380	NS
CRP	0	0	0	0.00	0.000	0.000	0.0000	NS
FBS	93.8	93.3	0.5	0.53	1.269	0.401	0.2443	NS
Blood Urea	32.5	32	0.5	1.54	1.509	0.477	0.3221	NS
SR. Creatinine	1	0.85	0.15	15.00	0.222	0.070	0.0617	NS
SGOT	39.7	38.4	1.3	3.27	3.368	1.065	0.2533	NS
SGPT	28.2	27.7	0.5	1.77	1.179	0.373	0.2126	NS
Sr. Cholesterol	166.3	164.3	2	1.20	4.163	1.317	0.1631	NS

262 \* BT-mean of before treatment, AT-mean of after treatment, Diff.-mean difference, SD-standard deviation,  
 263 SEM-Standard Error of Mean, Sig.-significance level.  
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265 **Table 10.analysis of variance (ANOVA) Test on subjective parameters for Intergroup Comparison:**  
 266

Inter Group Comparison	Group A	Group B	Group C	P value	Sig
Dizziness	0.4	0.4	1.3	0.2301	NS
Constipation	0.4	0.4	1	0.1050	NS
Aching Muscles	2.1	2.1	1.5	0.3352	NS
Joint Pain	0.6	0.6	0.4	0.8170	NS
Joint Stiffness	0.3	0.3	0.3	0.5741	NS
Sleep abnormality	0.4	0.4	2	0.0481	S
Loss of appetite	1.2	1.2	2	0.7768	NS
Fatigue	1.5	1.5	1.3	0.9497	NS
Gen. Weakness	0.7	0.7	1.2	0.1478	NS

267 \* BT-mean of before treatment, AT-mean of after treatment, Diff.-mean difference, SD-standard deviation,  
 268 SEM-Standard Error of Mean, Sig.-significance level.  
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 271 **Table 11 - Analysis of variance (ANOVA) Test on HARS subjective parameters for Intergroup**  
 272 **Comparison:**  
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Inter Group Comparison	Group A	Group B	Group C	P value	Sig
Anxious Mood	0.9	0.6	0.8	0.4221	NS
Tension	1.2	0.7	0.7	0.0749	NS
Fears	0.9	0.3	0.6	0.1975	NS
Insomnia	0.4	0.7	0.8	0.4969	NS
Intellectual	0.9	0.8	0.8	0.8235	NS
Depressed Mood	0.6	0.5	0.7	0.7967	NS
Somatic (Muscular)	0.3	0.4	-	0.0962	NS
Somatic (Sensory)	0.4	-	-	-	-
Respiratory Symptoms	0.1	0.3	0.3	0.4865	NS
Gastrointestinal Symptoms	0.3	0.4	0.7	0.1858	NS
Autonomic Symptoms	-	-	0.2	-	-
Behaviour at interview	0.3	-	-	-	-
Genitourinary symptoms	0.2	-	-	-	-

274 \* BT-mean of before treatment, AT-mean of after treatment, Diff.-mean difference, SD-standard deviation,  
 275 SEM-Standard Error of Mean, Sig.-significance level.  
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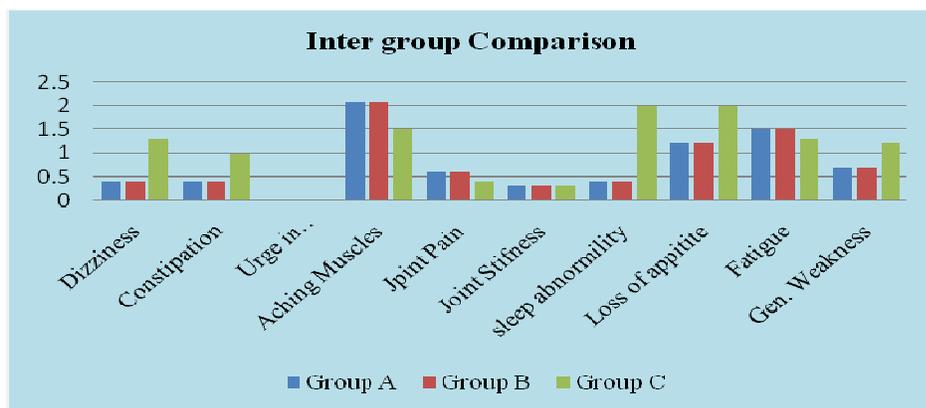
**Table 12. analysis of variance (ANOVA) Test on objective parameters for intergroup comparison**

Intergroup Comparison	Group A	Group B	Group C	P value	Sig
HB	0.74	0.197	0.56	0.3989	NS
TLC	220	260	50	0.0107	S
Neutrophil	2.1	1.6	2.7	0.9583	NS
Lymphocytes	3.3	2.2	1.7	0.4756	NS
Eosinophils	1.4	0.8	0.4	0.4134	NS
Monocytes	0.4	0.5	0.5	0.0856	NS
Basophils	0	0	0	-	NS
TRBC	0.245	0.072	0.2	0.7483	NS
TPLC	0.061	0.019	0.144	0.6732	NS

PCV	1.055	0.076	0.336	0.1943	NS
MCV	1.385	0.2	0.3	0.2399	NS
MCH	0.731	0.377	0.09	0.1830	NS
MCHC	0.343	0.565	0.01	0.456	NS
CRP	0	0	0	-	NS
FBS	8.6	2	0.5	0.0323	S
Blood Urea	0.9	2.1	0.5	0.0743	NS
SR. Creatinine	0.03	0.14	0.15	0.2850	NS
SGOT	2.5	5.1	1.3	0.1581	NS
SGPT	1.1	0.8	0.5	0.3114	NS
Sr. Cholesterol	8.2	8.1	2	0.3223	NS

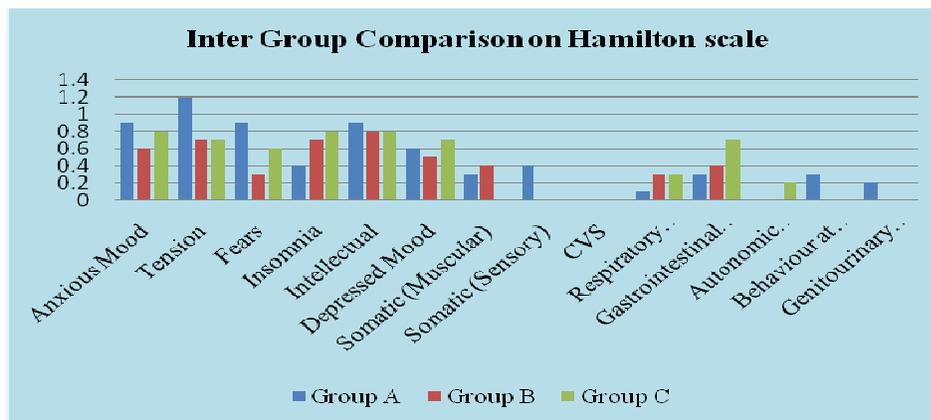
278 \* BT-mean of before treatment, AT-mean of after treatment, Diff.-mean difference, SD-standard deviation,  
 279 SEM-Standard Error of Mean, Sig.-significance level.  
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310 **Graph 1. Analysis of variance (ANOVA) Test for Intergroup Comparison on subjective parameters:**  
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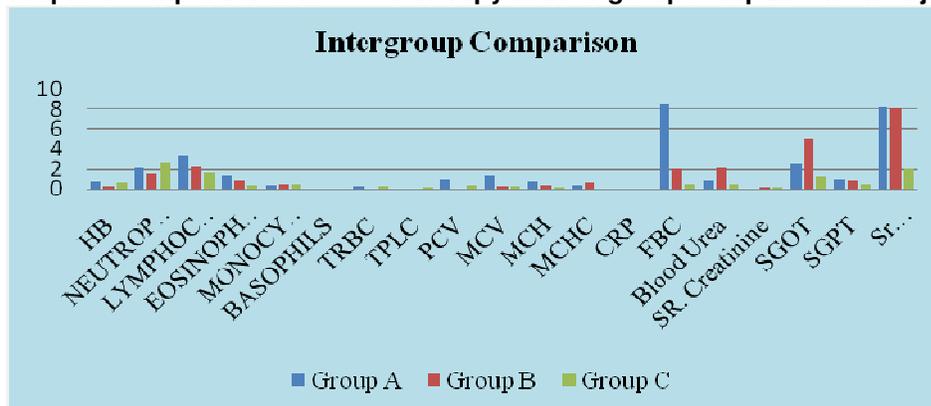
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**Graph 2.comperative effects of therapy in inter group comparison on Hamilton scale:**



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**Graph 3. comperative effects of therapy in inter group comparison on objective parameters:**



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**4. CONCLUSION**

Both the test drug, stem of *Guduchi* and Dried fruit rinds of *Amalaki* were found to have significant effect in delaying premature ageing due to stress. But in comparison to other groups the effect was better in Group C (*Guduchi* and *Amalaki*). The trial drugs were tolerated well in the study population as no ADR was observed during the trial duration. The study team suggests that since *Guduchi* and *Amalaki* were

328 found to be effective in delaying premature ageing due to stress, but considering the small size of the trial  
329 population, the trial should further be extended to larger sample size and for longer trial duration to draw  
330 more conclusive evidence.

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332 **CONSENT**  
333 All authors declare that 'written informed consent was obtained from the patient (or other approved  
334 parties) for publication of this manuscript. A copy of the written consent is available for review by the  
335 Editorial office/Chief Editor/Editorial Board members of this journal.

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338 **ETHICAL APPROVAL**  
339 All authors hereby declare that all experiments have been examined and approved by the appropriate  
340 ethics committee and have therefore been performed in accordance with the ethical standards laid down  
341 in the 1964 Declaration of Helsinki.

- 342  
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## 414 ABBREVIATIONS

415  
416 HB-Haemoglobin; TLC-Total Leukocyte Count; TRBC-Total Red Blood Cell Count; TPLC-Total Platelet  
417 Count; PCV- Packed Cell Volume; MCV- Mean corpuscular volume; MCH- Mean Cell Hemoglobin;  
418 MCHC- Mean Corpuscular Haemoglobin Concentration; CRP- C-Reactive Protein (CRP); FBS- Fasting  
419 Blood Sugar; SGOT- Serum Glutamic-Oxaloacetic Transaminase, It is also known as AST, or Aspartate  
420 Aminotransferase; SGPT- Serum Glutamic Pyruvic Transaminase (SGPT), It is also known as Alanine  
421 Aminotransferase (ALT).

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## 423 APPENDIX

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