An experimental study to evaluate *Gunasankarya* (combination of properties)

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Abstract

Introduction: Gunasankarya implies the combination or admixture of different Guna (properties) in a Dravya (substance). Every Dravya in this world is composed of different Guna varying in their quantity and potency. The final resultant action of the Dravya depends on the combination of Guna and their interaction with each other. Aim: The present study was aimed to explain the manifestation of Karma (action) of a Dravya through its Guna (properties) and to evaluate the effect of the combination of Sheeta Guna (cool property) with different other Guna on digestion and metabolism in an animal model. Materials and methods: The test drugs Usheera (Vetiveria zizanioides Linn.) and Bala (Sida cordifolia Linn.) were dried and powdered. A total of 18 adult healthy male and female Wistar strain albino rats were grouped randomly into three groups out of which the first was the control group and administered with distilled water. The second and third groups were fed with powder of Usheera and Bala, respectively, in the suspended form in distilled water. Test drugs were administered daily for 12 consecutive days. During the experimental phase, relative food intake, relative water intake, relative urine output, relative stool output, and food conversion ratio were recorded on the 3rd, 6th, 9th, and 12th day among which the 3rd day was considered as the initial day for the assessment, whereas body weight was measured from the 1st day of the study. Throughout the study, the main focus was on how these parameters change in Usheera and Bala-treated rats which are having different combinations of Guna with reference to the control group. Results: Both Usheera and Bala groups showed a nonsignificant increase in relative food and water intake, nonsignificant increase in relative urine output, significant decrease in relative stool output, and significant increase in food conversion ratio. Body weight was nonsignificantly increased in Usheera group, whereas it was nonsignificantly decreased in Bala group. This study aimed to show how the Guna are interacting and resulting in Karma either by dominating or suppressing or giving a combined effect of Guna. This study provides initial data regarding the concept of Gunasankarya (combination of properties). Conclusion: The study concludes that all the Guna in a Dravya interact and exhibit their resultant action, i.e., Karma based on Gunasankarya, with emphasis on example of assessment of the effects of Sheeta Guna Dravya, namely, Usheera and Bala on physiological parameters (digestion and metabolism) have been assessed experimentally in animal models to evaluation of the combination of Guna.

Keywords: Bala, Dravya, Gunasankarya, Karma, Sheeta Guna, Usheera

Introduction

It is a well-known fact that every *Dravya* (substance) contains more than one *Guna* (properties). A *Guru* (heavy) *Dravya* may have *Ruksha* (dry) and *Ushna* (hot) *Guna* in it or may have *Snigdha* (unctuous) and *Sheeta* (cold) *Guna* (property) in it. The final action of the *Dravya* depends on the combined reaction of these *Guna*. It is therefore very important to see how these *Guna* affect each other or how they produce a specific *Karma* (action). This can be considered as *Gunasankarya* even though no classical reference is available on this specific term. The concept of *Guna* and *Karma* has always been an appealing topic for researchers.



The term *Sankara* means mixing or blending^[1] *Gunasankarya* implies the combination or admixture of different *Guna* in a *Dravya*. Each and every *Dravya* in this world is composed of combination of different *Guna* in which they vary in their quantity or potency or substrata. Although *Dravya* is known either by one or two *Guna* which are predominant in nature

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whereas the reality is that a *Dravya* incorporates many other *Guna* too and its final resultant action depends on the reaction among these *Guna*.

Guna within a Dravya behave generally in two ways. The first one is, by supplementing other Guna and thereby expressing a cumulative effect resulting in Karma which is associated with each Guna. The second one is, by suppressing or dominating the other Guna. When two or more Guna are interacting with each other, their Bhautika (configurational) structure decides how they are going to support or get suppressed/dominated by other Guna.[2] For example, in a Dravya such as Ksheera (milk)^[3] Snigdha, Picchila (slimy in nature), and Sheeta Guna are predominantly Jala Mahabhuta Pradhana (predominant), whereas Guru Guna is having a predominance of Prithvi Mahabhuta; the Karma of Ksheera expresses the action of sum aggregate of Guna in it, and therefore, it can be inferred that the resultant action has been complimented by all the Guna present in it. On the other hand, a Dravya like Vasa (Adhatoda vasica Nees.) which has Sheeta, Ruksha Guna in it; out of which Sheeta Guna is predominant of Jala Mahabhuta and Ruksha Guna is predominant of Vayu Mahabhuta^[4] and hence, these Guna does not complement each other and expresses an antagonistic action. Vasa with its Ruksha Guna suppresses Sheeta Guna and thereby acts as Shoshaka (drying up) of the Dravatva (liquidity) and produces an action of Kapha and Pitta Nashana (alleviating). In this study, two Dravya Usheera (Vetiveria zizanioides Linn.) and Bala (Sida cordifolia Linn.) have been selected, out of which both are predominantly Sheeta Guna and distinctively Usheera has Laghu, Ruksha Guna and Bala has Guru, Snigdha Guna associated with it. When these drugs are administered to the Wistar albino rats, the study focuses on the interaction between the Guna and exhibition of resultant Karma (pharmacological action) of Dravya. While going through all the works reviewed at different Institutes, it was observed that none of the work has been yet carried out to explain the concept of Gunasankarya. Hence, this study mainly aimed to explain the manifestation of Karma of a Dravya through its Guna (attributes). To understand the concept well, Sheeta Guna was selected as it can express more evidently and is also considered a Veerya due to its action. To understand the combination of different Guna in a Dravya, the best method is the evaluation of digestion and metabolism in an animal model as the author is focussing on the primary level assessment.

Aim

To explain the manifestation of *Karma* of a *Dravya* through its *Guna* (attributes) and to evaluate the effect of the combination of *Sheeta Guna* with different other *Guna* on digestion and metabolism in an animal model.

Materials and methods

Materials

The test drug Usheera (Vetivera zizanoides Linn.) in root powder form was collected from the Pharmacy of Gujarat Ayurved University, Jamnagar. The test drug *Bala* (*S. cordifolia* Linn.), whole plant was collected locally from Ernakulam district of Kerala, and whole plant was dried and powdered after which both the powders along with root of *Bala* were authenticated from the Pharmacognosy Laboratory of I.P.G.T. and R.A. (Specimen Voucher no. IPGT and RA Phm. 6342/2020-21 and IPGT and RA Phm. 6343/2020-21 for *Usheera* and *Bala*, respectively).

Experimental animals

A total of 18 healthy male and female (9 male + 9 female) adult Wistar strain Albino rats (12–16 weeks old) were selected with body weight in the range of 225 ± 25 g from the animal house attached to the Pharmacology laboratory, I.P.G.T and R.A. The animals were exposed to 12-h light and 12-h dark cycle with the relative humidity of 50%–70% and the ambient temperature during the period of experimentation was $22^{\circ} \pm 03^{\circ}$ C. They were provided with feed and purified water *ad libitum* in polypropylene bottles with stainless steel sipper tube. The experimental study and protocols described in the study were approved by IAEC (Approval number: IAEC/26/2020/01) and in accordance with CPCSEA guidelines.

Methods

Experimental protocol

Animals were kept on acclimatization for 15 days and thereafter they were grouped randomly into three groups and each rat was kept in a separate metabolic cage. The first group served as normal control group and was administered with distilled water. The second and third groups were fed with *Usheera* and *Bala Churna* respectively in a suspended form with distilled water and administered orally through an oral feeding cannula. A test diet was provided daily for 12 consecutive days.

The therapeutic dose for the test drugs, i.e., both *Usheera* and *Bala Churna* for rats were fixed at 1080 mg/kg body weight of rat per day (as per the table of Paget's and Barnes) and control group with distilled water in a dose of 10 ml/kg for the duration of 12 days.

Assessment of study

Body weight, relative food intake, relative water intake, relative urine output, relative stool output, and food conversion ratio were recorded on the 3rd, 6th, 9th, and 12th day among which the 3rd day was considered as initial day and the baseline data for assessment of these parameters of the experiment except body weight for which 1st day was considered as the initial day and baseline for the data. These parameters were selected on the ground to understand the physiological action of the combination of *Guna* in body, i.e., domination of *Snigdha Guna* over *Sheeta Guna* may show an increased food intake, domination of *Sheeta Guna* may show a decreased water intake relatively. In a similar way, other parameters have also been used as an initiative to find out how the results may be contributing and therefore no other biochemical tests were performed on the animals.

Statistical analysis

The obtained data have been presented as mean \pm standard error of the mean, the difference between the groups was statistically determined by Student's *t*-test for paired and unpaired data with the level of significance set at P < 0.05.

Results

Out of the three groups with six animals in each group, one female from *Usheera* treated group was excluded (female rat found pregnant and had to be excluded) accounting to 17 animals in total on which the study was conducted successfully. The health status of the animals including weight, behavior was assessed before the experiment.

The *Dravya* selected, i.e., *Usheera* and *Bala* have both shown similar changes regarding the parameters assessed when they have been compared to the control group which finally leads to the inference that both the *Sheeta Guna Dravya* have presented data which non-significant decrease in Bala group and non-significant increase in Usheera group in body weight parameters. [Table 1] nonsignificant increase in relative food and water intake, [Table 2 and Table 3] nonsignificant increase in relative stool output, [Table 4] significant (P < 0.05) decrease in relative stool output [Table 5] and significant (P < 0.05) increase in food conversion ratio. [Table 6] was observed for Usheera and Bala group.

Discussion

The concept of Gunasankarya can be employed in many facets of treatment modalities such as in making herbal formulations, and compound medicines with different Dravya of various Guna. Paradi Guna like Samskara is mainly intended to modify its Bhautika structure (physical property) to make alterations in their Guna and thereby in Karma too.^[5] In this aspect, Gunasankarya can be understood more efficiently. Various techniques mentioned under Samskara such as Bhavana (trituration), Vasana (flavoring), and Jala-Agni Sannikarsha (contact with water or fire) are some of the modalities which illustrate the concept of Gunasankarya more efficiently.^[5] Even though many pharmacological and clinical studies have been conducted on both the drugs Usheera and Bala concerning various aspects such as anti-stress, antioxidant, anti-hyperglycemic, and hepatoprotective activities, no study results have been found discussed concerning the effect of these drugs on physiological parameters of the body. This study has been an explorative one to find the results on physiological parameters and is being discussed in this study.

Body weight

Body weight indicates the health status of living beings. Change in body weight is an important indicator of the health of an animal. The present study shows nonsignificant increase in body weight in *Usheera* group and decrease in *Bala* group. *Usheera* and *Bala* both are *Sheeta Dravya* which have *Jala Mahabhuta* (water element) in predominance having an

Table 1: Effect of test drugs on body weight (g) of Wistar strain Albino rats

Group	$Mean \pm SEM$	t	<i>P</i> (CI)
Control			
1 st day	$258.833{\pm}5.540$		-
12 th day	$273.167{\pm}5.492$		-
Usheera			
1st day (baseline)	265.7±8.6	-0.695	0.504 (-29.201-15.468)
12 th day	275±14.94	-0.124	0.904 (-35.283-31.617)
Bala			
1st day (baseline)	251.5±18.2	0.386	0.707 (-34.982-49.649)
12 th day	257.67±22.6	0.667	0.520 (-36.281-67.281)

P value: Significance when compared with control group (unpaired *t*-test), CI: CI for the *P* value. CI: Confidence interval, SEM: Standard error of the mean

Table 2: Effect of test drugs on relative food intake (g%) of Wistar strain Albino rats

Group	$Mean \pm SEM$	t	<i>P</i> (CI)
Control			
1 st day	5.871 ± 0.52	-	-
12 th day	6.01±0.22	-	-
Usheera			
3rd day (baseline)	6.71±0.5	-1.188	0.265 (-2.439-0.759)
12 th day	6.22±0.13	-0.808	0.440 (-0.827-0.392)
Bala			
3rd day (baseline)	6.95 ± 0.4	-1.727	0.115 (-2.472-0.314)
12 th day	6.27±0.18	-0.913	0.383 (-0.897-0.376)
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Data: When compared with control group (unpaired *t*-test). CI: CI for the *P* value. CI: Confidence interval, SEM: Standard error of the mean

Table	e 3:	Eff	ect	of	test	drugs	on	relative	water	intake
(mL	%)	of \	Nisi	tar	stra	in Alb	ino	rats		

Group	$Mean \pm SEM$	t	<i>P</i> (CI)
Control			
1 st day	$6.183{\pm}0.72$	-	-
12 th day	5.83 ± 0.57	-	-
Usheera			
3rd day (baseline)	6.512 ± 0.651	-0.332	0.747 (-2.569-1.911)
12 th day	8.31 ± 1.08	-2.138	0.061 (-5.116-0.144)
Bala			
3rd (baseline)	6.080 ± 0.3	0.135	0.896 (-1.603-1.809)
12 th day	6.21±0.68	-1.426	0.672 (-2.367-1.592)

When compared with control group (unpaired *t*-test). CI: CI for the *P* value. CI: Confidence interval, SEM: Standard error of the mean

anabolic activity which should increase the body weight. However, the drug *Usheera* also has *Guna* like *Ruksha* and *Laghu* associated with *Sheeta* which assures the presence of *Vayu Mahabhuta* (air element), probably antagonizing the *Jala Mahabhuta* as observed in the present study. *Vayu* as discussed earlier is *Yogavahi* (nature of imbibing the quality of associated ones), hence when combined with *Jala* will exhibit *Sheeta Guna. Vayu Mahabhuta* can tend to increase the metabolic and digestive activities due to which *Usheera*-treated animals

171

Table 4: Effect of test drugs on relative urine output (mL %) of Wistar strain Albino rats

Group	$Mean \pm SEM$	t	<i>P</i> (CI)
Control			
1 st day	1.813 ± 0.23	-	-
12 th day	1.33 ± 0.17	-	-
Usheera			
3rd day (baseline)	2.09 ± 0.45	-0.584	0.574 (-1.364-0.805)
12 th day	1.86 ± 0.55	-0.984	0.351 (-1.730-0.681)
Bala			
3rd day (baseline)	2.71±0.5	-1.656	0.129 (-2.093-0.308)
12 th day	1.98 ± 0.32	-1.772	0.107 (-1.455-0.166)

Data: When compared with control group (unpaired *t*-test). CI: CI for the *P* value. CI: Confidence interval, SEM: Standard error of the mean

Table 5: Effect of test drugs on relative stool output (g%) of Wistar strain Albino rats

Group	$Mean \pm SEM$	t	<i>P</i> (CI)
Control			
1 st day	0.851 ± 0.1	-	-
12 th day	0.943 ± 0.02	-	-
Usheera			
3rd day (baseline)	$0.81{\pm}0.1$	0.330	0.749 (-0.243-0.326)
12 th day	$0.809 \pm 0.06*$	2.333	0.044* (0.00408-0.263)
Bala			
3rd day (baseline)	$0.83 {\pm} 0.05$	1.518	0.831 (-0.203-0.248)
12 th day	$0.811 \pm 0.05*$	2.606	0.026* (0.0191-0.245)
1			

**P*<0.05, when compared with control group (unpaired *t*-test). *P* value: Significance, CI: CI for the *P* value. CI: Confidence interval, SEM: Standard error of the mean

Table 6: Effect of test drugs on food conversion ratio (g%) of Wistar strain Albino rats

Group	$Mean \pm SEM$	t	Р
Control			
1 st day	$6.97 {\pm} 0.4$	-	-
12 th day	6.37±0.2	-	-
Usheera			
3rd day (baseline)	8.61±0.9	-1.792	0.107 (-3.707-0.429)
12 th day	7.9±0.63*	-2.474	0.035* (-2.8750.129)
Bala			
3rd day (baseline)	8.6 ± 0.71	-1.989	0.075 (-3.420-0.194)
12 th day	7.84±0.44*	-3.071	0.012* (-2.5390.404)

**P*<0.05, when compared with control group (unpaired *t*-test). *P* value: Significance, CI: CI for the *P* value. CI: Confidence interval,

SEM: Standard error of the mean

had shown a nonsignificant increase in body weight, whereas *Bala*-treated animals had shown a nonsignificant decrease in body weight when compared to control group. *Gurutva* and *Sheetatva* of *Bala* is contrary to *Agni* (digestive fire); hence, it is expected to slow down the process of digestion. *Bala* is also known to be *Grahi* (drying up of liquid in stool) according to Ayurveda classics. If the *Agni* is capable to digest, in spite of having anabolic qualities, drug will reduce body weight.

Even though in classical texts, *Bala* is mentioned to increase strength but no direct reference related to body weight has been mentioned.^[6] Various research studies have been conducted on *Bala* (*S. cordifolia* Linn.) regarding anti-diabetic activity in which Narasimha Rao *et al.* have cited that usage of *Bala* has reduced blood sugar as well as body weight which goes hand in hand with the results obtained in the present study.^[7]

Effect on relative food intake, relative water intake

Usheera metabolizes the food much faster than control group with its *Karma* of *Pachana* and *Laghu*, *Ruksha Guna*,^[8] as well as dominating over the *Sheeta Guna*. Similarly, *Bala* is mainly of *Snigdha Guna* and along with *Sheeta Guna*, it directs the anabolic activity thereby reducing the *Agni* which could have led to the nonsignificant increase in relative food intake which was found in *Bala* group as compared to the control group. As discussed earlier, *Vayu Mahabhuta* can tend to increase the metabolic rate, thereby increasing the relative food intake in *Usheera* group.

According to the Ayurveda principles, the human body always repulses the qualities and properties which get increased or aggravated in the body. The drug *Usheera* is *Sheeta* in *Guna* and is made up of *Jala Mahabhuta*, these qualities get increased in the body due to which the body develops reduced thirst resulting in nonsignificant decrease in relative water intake. Our body is designed in such a way that it itself tries to maintain homeostasis thereby keeping the water balance of the body and decrease the water intake till some threshold but with constant use, this may vary.^[9] Here, *Dravya Usheera* which is known to have even although *Trushnaghna Karma* (action of thirst reduction), the body starts to get used to the condition and finally results in nonsignificant increase of relative water intake when compared to the control group.

When the relative water intake of *Bala*-treated group was observed from the beginning, it has shown a nonsignificant decrease which denotes that the *Dravya* was self-nutritive with *Snigdha*, *Sheeta Guna*, and predominantly made of *Jala Mahabhuta*, and hence, did not induce thirst due to which there was a withholding effect on water intake in the beginning which later on increased due to the habitual consumption of the *Dravya*.

Effect on relative urine output, relative stool output

Both Usheera and Bala as Sheeta Guna Dravya are expected to have anabolic action. The role of Gunasankarya can be seen playing its role here. Bala having all Guna of the same group, i.e., Saumya which is predominant with Jala Mahabhuta is the cause of nonsignificant increase of relative urine output whereas Usheera having Laghu, Ruksha Guna from other groups, i.e., Agneya interacting with Sheeta Guna (Saumya) and their ultimate combination of Guna results here in Pachana Karma (digestive process) by dominating the Sheeta Guna but the Sheeta Guna is known to produce more urine with the principle of homeostasis,^[10] i.e., when the Jala Mahabhuta inside the body gets increased, correspondingly body will be having a tendency to expel out the excess of Jala Mahabhuta inside the body reflecting in the increase in relative urine output in both the drug-treated groups when compared to control group. *Bala* has shown a higher percentage of increase when compared to *Usheera* group because *Bala* has *Sheeta Guna* associated with *Snigdha Guna*.

Although Sheeta Guna is known to have Karma of Stambhana (obstruction)[11] but being associated with Ruksha Guna which is composed of Vayu and Agni Mahabhuta (fire element), it does the Pachana Karma (digestion process)[12] and hence with increased absorption, Usheera might have made the fecal matter more formed and dried leading to a decreased stool output. Bala is known to have Sheeta Guna with Guru and Snigdha Guna associated with it. Even though Guru and Snigdha Guna induce Saratva of Mala i.e., produce more fecal output due to Jala Mahabhoota predominance^[13] but here, the Sheeta Guna which is meant to have Stambhana action has surpassed the other Guna while interaction by obstructing or slowing down the process of digestion and metabolism. Furthermore, Bala is known to have Graahi Guna (absorbent property) according to Ayurvedic classics as discussed earlier which provides a similar result of decreased stool output when compared to control group. Graahi Guna of Bala is evident from this research, and it could be due to the dominancy of Sheeta Guna in comparison to Snigdha Guna.

Effect on food conversion ratio in rats

Food conversion ratio is an indicator of the rate of metabolism or the digestion which also shows the rate of food ingested and converted into fecal matter.

Both Usheera and Bala-treated animals have shown increased food conversion ratio when compared to the control group. It could be manifested due to the associated Sheeta Guna. Usheera has Ruksha and Laghu Guna too which has an antagonistic action on Sheeta and dominates the process of digestion and metabolism, leading to a significantly increased Food conversion ratio whereas Bala has Snigdha Guna associated with the Sheeta Guna of which both are Saumya in nature; however, here the Snigdha Guna is helping in digestion even though Sheeta Guna tries to slow down the process which resulted in significantly increased food conversion ratio even though comparatively lesser to Usheera treated animals.

The primary outcome of this study was to see the changes in physiological parameters from the purview of *Guna*. Secondarily, the study was designed to show how the combination of *Guna* in a *Dravya* makes changes in the resultant action.

The dominant *Guna* acted in both the *Dravya* can be concluded. [Table 7].

The present study was conducted aimed to find out how the combinations of *Guna* in various *Dravya* lead to different resultant actions or how it varies. The results obtained have shown that when *Ruksha* (*Agneya Guna*) and *Sheeta Guna* (*Saumya Guna*) which are opposing in nature react with other *Guna* within the same group like *Saumya* or *Agneya* in

Table 7: Dominant Guna in assessed parameters				
Action	Dominant Guna			
	Usheera	Bala		
Body weight	Laghu	Graahi		
Relative food intake	Laghu	Sheeta		
Relative water intake	Sheeta	Sheeta		
Relative urine output	Sheeta	Sheeta		
Relative stool output	Ruksha	Sheeta, Graahi		
Food conversion ratio	Ruksha, Laghu	Snigdha		

nature react differently or in less/higher intensity of its action which leads to suppression or elimination of the action of another Guna. The study was conducted as an experiment on animals which clears out the confounding factors which may be generated due to different food habits in people when done on a clinical aspect. The subjects enrolled in a clinical study may not be able to follow a stable and consistent diet to assess the changes in the interaction of Guna. However, at the same time, there are certain limitations of study when it comes to minute interaction of Guna in a Dravya resulting in different actions. Psychological assessment of rats are not done in a physiological study but the chances of a different action can be associated with any mental afflictions too, which are not available with the data. Even though the whole results have been interpreted with possible assumptions, there are chances that the resultant action could have been in a different direction too as these minute reactions on the level of Mahabhutas are not predictable. Various interaction studies with a greater number of Guna and Dravya with increased duration of the period can be further elaborately studied to get much more precise results. Gunasankarya depends on Dravya itself for Usheera as Sheeta Guna is seen as dominant for certain actions but for Bala Snigdha Guna turns out to be dominant for the same action.

Conclusion

The action or Karma (action) of any Dravya (substance) is based on the Guna present in it. The present study confirms that the combination of different Guna in a particular Dravya will lead to a distinct resultant action due to Gunasankarya. The present experimental study has also indicated that Sheeta Guna Dravya (cool property substasnce), i.e., Usheera (Vetiveria zizanioides Linn.) and Bala (S. cordifolia Linn.) in Wistar strain Albino rats have specific effects on physiological parameters like body weight, relative food intake, relative water intake, relative urine output, relative stool output, and food conversion ratio. Guna interactions are also depending on the area of action, for example, for digestion Ruksha Guna is seen to be attaining more potency in comparison to Sheeta Guna and while for stool formation Sheeta Guna is working predominant in comparison to Ruksha Guna. The combination of Ruksha, Sheeta, and Laghu Guna in Usheera and Snigdha, Sheeta Guna in Bala have shown distinct results in various parameters with varying degrees.

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Conflicts of interest

There are no conflicts of interest.

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