In vivo study using experimental animal model is the best way of validating and testifying the results in order to determine the efficacy, potency and therapeutic nature of the drugs. A number of screening methods have been used to validate the efficacy of various herbomineral formulations. No study can be accomplished without the help of animal model and animal study for which selection of proper animal model and screening method is important. Although, the ultimate beneficiaries of any in vivo study are human beings, but as it is not possible to use humans for all experimentations. Hence appropriate animal models are designed which closely resemble the human body environment and therefore are used to validate the results.

Relevance of animal experimentation for the present investigation on Shwaskuthar Rasa – Although animal experimentation for any therapeutic formulation is must and without such study particularly no drug can be allowed for human use / consumption, it is more important and necessary in ease of present research topic on Shwaskuthar Rasa – a highly valued and reputed herbomineral formulation of Ayurveda where so-called heavy metals i.e. mercury, arsenic and poisonous herbal ingredient aconite have been used. It was therefore proposed not only to study physical and chemical aspects of the preparation and evaluate its pharmacology for its therapeutic efficacy on one hand but also to study the preparation for its toxicity / safety aspects. With a view to study the impact of milling for particle size reduction on bio-efficacy and accumulation in body organs. Keeping this in mind the in-process withdrawn samples of Shwaskuthar Rasa were subjected to overall pharmacological investigation employing appropriate animal models.

6.1 EXPERIMENTAL ANIMALS

The animals were acclimatized to animal house prior to experimentation, they were divided into different group of six animals per group, kept in colony cages at ambient temperature of 280 ± 2 °C and 45 to 55 % relative humidity with a 12 hrs light/dark cycle and allowed
free access to standard diet and water \textit{add libitum}. The protocol for animal experimentation approved by Institutional animal ethical committee (IAEC) of Dr. H. S. Gour Vishwavidyalaya, Sagar was followed.

\section*{6.2 Screening of In-process Withdrawn Samples of Shwaskuthar Rasa at Scheduled Intervals for Their Antiasthmatic Activity}

The in-process withdrawn \textit{Shwaskuthar Rasa} samples were evaluated for antiasthamtic activity on following models.

\subsection*{6.2.1 Study of histamine induced bronchospasm in guinea pigs}

\begin{tabular}{ll}
\textbf{Animal} & Guinea pigs \\
\textbf{Species} & Duncon hartely \\
\textbf{Age/ Weight} & Adult / 400-600 gm \\
\textbf{Sex} & Either male / female \\
\textbf{No. of animals:} & 36 (Six animals per group) \\
\end{tabular}

\textbf{Preparation of drug solution:} \textit{Shwaskuthar Rasa} (20 mg/kg, p.o.) suspensions were prepared using 2 \% v/v Tween 80. Group-I animals received 2 \% v/v of Tween-80 (10 ml/kg, p.o) Animals of Group-II, III, IV, V and VI were treated with \textit{Shwaskuthar Rasa} samples -SWR 1220, SWR 829, SWR 574, SWR 216 and SWR 92 (20 mg/kg, p.o) respectively.

\textbf{Procedure:} Prior to and after drug (\textit{Shwaskuthar Rasa} suspension) treatment, animals of each group i.e. Group-II, III, IV, V and VI were individually placed in the histamine chamber and exposed to 0.2\% histamine aerosol under constant pressure (40 mm/Hg in an aerosol chamber). The preconvulsive time (PCT) was determined from the time of exposure to the onset of dyspnoea leading to the appearance of preconvulsive dyspnoea in a min. The percentage (\%) protection against bronchospasm offered by in-process withdrawn samples of \textit{Shwaskuthar Rasa} in preconvulsive time (PCT) was calculated according to Gokhale and Saraf, 1996.
Table 6.1 Effect of milling on in-process withdrawn samples of *Shwaskuthar Rasa* on histamine induced bronchospasm in guinea pig

<table>
<thead>
<tr>
<th>S. No</th>
<th>Treatment Group</th>
<th>Particle size of preparation (nm)</th>
<th>Dose mg/kg p.o</th>
<th>PCT (T1)</th>
<th>PCT (T2)</th>
<th>Mean exposition time (T2-T1)</th>
<th>% Protection against bronchospasm</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Control</td>
<td>-</td>
<td>10 ml&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.518 ± 0.24</td>
<td>1.550 ± 0.32</td>
<td>0.032 ± 0.22</td>
<td>2.06</td>
</tr>
<tr>
<td>II</td>
<td>SWR 1220</td>
<td>1220</td>
<td>20 mg</td>
<td>1.442 ± 1.12</td>
<td>2.454 ± 0.66</td>
<td>1.012 ± 0.75*</td>
<td>41.23</td>
</tr>
<tr>
<td>III</td>
<td>SWR 829</td>
<td>829</td>
<td>20 mg</td>
<td>1.536 ± 1.15</td>
<td>2.806 ± 1.36</td>
<td>1.270 ± 1.22*</td>
<td>45.26</td>
</tr>
<tr>
<td>IV</td>
<td>SWR 574</td>
<td>574</td>
<td>20 mg</td>
<td>1.387 ± 0.76</td>
<td>3.211 ± 1.20</td>
<td>1.824 ± 0.78*</td>
<td>56.80</td>
</tr>
<tr>
<td>V</td>
<td>SWR 216</td>
<td>216</td>
<td>20 mg</td>
<td>1.512 ± 0.88</td>
<td>4.872 ± 1.45</td>
<td>3.360 ± 1.44**</td>
<td>68.96</td>
</tr>
<tr>
<td>VI</td>
<td>SWR 92</td>
<td>92</td>
<td>20 mg</td>
<td>1.368 ± 0.45</td>
<td>5.722 ± 0.62</td>
<td>4.354 ± 1.36**</td>
<td>76.09</td>
</tr>
</tbody>
</table>

<sup>a</sup> Tween 80 (10 ml/kg, p.o), PCT (T1) - Pre-convulsive time before drug administration, PCT (T2) - Pre-convulsive time after drug administration. The data expressed are mean ± S.E.M. n=6; *p<0.05 - significant; ** p<0.01 - highly significant (one way ANOVA followed by paired 't'-test)

![Fig 6.1 Effect of milling on in-process withdrawn samples of *Shwaskuthar Rasa* on histamine induced bronchospasm in guinea pigs](attachment:image.png)
6.2.2 Study of mast cell degranulation in mice

**Animal** : Mice  
**Species** : Albino  
**Age/Weight** : Adult / 25-30 gm  
**Sex** : Either male / female  
**No. of animals** : 36 (Six animals per group)

**Preparation of drug solution**: *Shwaskuthar Rasa* (33 mg/kg, p.o.) suspensions were prepared using 2 % v/v Tween 80.

Group-I animals received 2 % v/v of Tween-80 (10 ml/kg, p.o)

Animals of Group-II, III, IV, V and VI were treated with *Shwaskuthar Rasa* samples - SWR 1220, SWR 829, SWR 574, SWR 216 and SWR 92 (33 mg/kg, p.o) respectively.

**Procedure**: Three days drug (*Shwaskuthar Rasa* suspension) treatment schedule was followed. On day fourth each mice was injected with 4 ml/kg, 0.9 % w/v NaCl solution into peritoneal cavity. By gentle massage, peritoneal fluid was collected after 5 min and transferred into siliconised test tube containing 7-10 RPMI-1640 buffer medium (pH 7.2-7.4). The contents of test tubes were centrifuged at 400-500 rpm. Pellets of mast cell were washed with same buffer medium twice by centrifugation, discarding supernatant. The cells were challenged with clonidine (50 μg) and incubated at 37 °C in a water-bath for 10 min. Followed by staining with 1 % toluidine blue, the cells were observed under microscope (45 X). Total 100 cells were counted from different visual area. Percentage protection against degranulation was calculated according to *Lakdawala et al., 1980*. 
Table 6.2 Effect of milling on in-process withdrawn samples of *Shwaskuthar Rasa* on clonidine induced mast cell degranulation in mice

<table>
<thead>
<tr>
<th>S.No</th>
<th>Treatment Group</th>
<th>Particle size of preparation (nm)</th>
<th>Dose mg/kg p.o</th>
<th>No of mast cell degranulated per cubic mm</th>
<th>% Protection of mast cell degranulation</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Control</td>
<td>-</td>
<td>10ml&lt;sup&gt;a&lt;/sup&gt;</td>
<td>345.58 ± 2.34</td>
<td>-</td>
</tr>
<tr>
<td>II</td>
<td>SWR 1220</td>
<td>1220</td>
<td>33mg</td>
<td>228.23 ± 1.22*</td>
<td>33.95</td>
</tr>
<tr>
<td>III</td>
<td>SWR 829</td>
<td>829</td>
<td>33mg</td>
<td>184.15 ± 1.78*</td>
<td>46.71</td>
</tr>
<tr>
<td>IV</td>
<td>SWR 574</td>
<td>574</td>
<td>33mg</td>
<td>165.42 ± 2.32**</td>
<td>52.13</td>
</tr>
<tr>
<td>V</td>
<td>SWR 216</td>
<td>216</td>
<td>33mg</td>
<td>122.56 ± 1.66**</td>
<td>64.53</td>
</tr>
<tr>
<td>VI</td>
<td>SWR 92</td>
<td>92</td>
<td>33mg</td>
<td>95.28 ± 1.44**</td>
<td>72.42</td>
</tr>
</tbody>
</table>

<sup>a</sup> Tween 80 (10 ml/kg, p.o)  
The data expressed are mean ± S.E.M. n=6; *p<0.05 - significant; ** p<0.01 - highly significant (one way ANOVA followed by Dunnett's-test)
6.2.3 Study of clonidine induced catalepsy in mice

Catalepsy is a condition in which the animal maintains imposed posture for long time before regaining normal posture. Catalepsy is a sign of extrapyramidal effect of drug that inhibit dopaminergic transmission or increases histamine release in brain. Clonidine, a $\alpha_2$ adreno receptor agonist induces dose dependent catalepsy in mice, which is inhibited by histamine H$_1$ receptor antagonist. Different stages of catalepsy appear to be directly correlated with brain histamine content (Chopra and Dandiya, 1975).

**Animals**: Mice

**Species**: Albino

**Age/Weight**: Adult / 25-30 gm

**Sex**: Either male / female

**No. of animals**: 36 (Six animals per group)

**Preparation of drug solution**: *Shwaskuthar Rasa* (33 mg/kg, p.o.) suspensions were prepared using 2 % v/v Tween 80.

Group-I animals received 2 % v/v of Tween-80 (10 ml/kg, p.o)

Animals of Group-II, III, IV, V and VI were treated with *Shwaskuthar Rasa* samples - SWR 1220, SWR 829, SWR 574, SWR 216 and SWR 92 (33 mg/kg, p.o) respectively.

**Procedure**: The forepaws of mice were placed on a horizontal bar (1cm in diameter, 3 cm above the table). The time required to remove the paws from bar was noted for each mice. Animals of all the groups received clonidine (1 mg/kg, s.c) one hour after the drug (*Shwaskuthar Rasa* suspension) administration. Duration of catalepsy was measured at 15, 30, 60, 90, 120, 150 and 180 min according to Ferre et al., 1990.
Fig 6.3 Effect of milling on in-process withdrawn samples of *Shwaskuthar Rasa* on clonidine induced catalepsy in mice.
6.3 SCREENING OF IN-PROCESS WITHDRAWN SAMPLES OF SHWASKUTHAR RASA AT SCHEDULED INTERVALS FOR THEIR ANTIALLERGY ACTIVITY

Antiallergic activity of withdrawn samples of *Shwaskuthar Rasa* was performed on following models.

### 6.3.1 Study of passive cutaneous anaphylaxis in rats

**Animal** : Rats  
**Species** : Albino  
**Age/Weight** : Adult / 100-150 gm  
**Sex** : Male  
**No. of animals**: 36 (Six animals per group)

**Preparation of drug solution**: *Shwaskuthar Rasa* (23 mg/kg, p.o.) suspensions were prepared using 2 % v/v Tween 80.  
Group-I animals received 2 % v/v of Tween-80 (10 ml/kg, p.o)  
Animals of Group-II, III, IV, V and VI were treated with *Shwaskuthar Rasa* samples - SWR 1220, SWR 829, SWR 574, SWR 216 and SWR 92 (23 mg/kg, p.o) respectively.

**Procedure**: Male rats were injected subcutaneously with 100 μg egg albumin (EA) coated with 12 mg aluminum hydroxide on the 1st, 3rd and 5th day for both sensitization and homologous antiserum. On day 10th, blood was collected from the retro-orbital plexus of rats and serum containing IgE antibodies separated by centrifugation. Dorsal surface of the male rats was shaved and sensitized with 0.1 ml of rat homologous antiserum injected intradermally on the shaved dorsal surface after 48 hrs of sensitization, so that antibodies become fixed to receptors of the mast cell membranes. Drug (*Shwaskuthar Rasa* suspension) was administered orally to all groups except control, after one hour, 1 % egg albumin along with 0.5 % Evan’s blue (0.25 ml) was administered intravenously. Subsequently after one hour, the blue coloured area of the skin was measured in square mm (mm²) according to Yang et al., 2008.
### Table 6.4 Effect of milling on in-process withdrawn samples of *Shwaskuthar Rasa* on passive cutaneous anaphylaxis in rats

<table>
<thead>
<tr>
<th>S.No</th>
<th>Treatment Group</th>
<th>Particle size of preparation (nm)</th>
<th>Dose mg/kg p.o</th>
<th>Area of blue coloured skin (Square mm)</th>
<th>% Protection against PCA</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Control</td>
<td>-</td>
<td>10 ml&lt;sup&gt;a&lt;/sup&gt;</td>
<td>452.16 ± 1.14</td>
<td>-</td>
</tr>
<tr>
<td>II</td>
<td>SWR 1220</td>
<td>1220</td>
<td>23 mg</td>
<td>295.24 ± 1.70*</td>
<td>34.70</td>
</tr>
<tr>
<td>III</td>
<td>SWR 829</td>
<td>829</td>
<td>23 mg</td>
<td>263.06 ± 2.10*</td>
<td>41.82</td>
</tr>
<tr>
<td>IV</td>
<td>SWR 574</td>
<td>574</td>
<td>23 mg</td>
<td>224.17 ± 1.98*</td>
<td>50.42</td>
</tr>
<tr>
<td>V</td>
<td>SWR 216</td>
<td>216</td>
<td>23 mg</td>
<td>197.43 ± 1.10**</td>
<td>56.33</td>
</tr>
<tr>
<td>VI</td>
<td>SWR 92</td>
<td>92</td>
<td>23 mg</td>
<td>162.22 ± 2.08**</td>
<td>64.12</td>
</tr>
</tbody>
</table>

<sup>a</sup> Tween 80 (10 ml/kg, p.o), PCA - Passive cutaneous anaphylaxis

The data expressed are mean ± S.E.M. n=6; *p*<0.05 - significant; **p**<0.01 - highly significant (one way ANOVA followed by Dunnett’s-test)

![Graph showing % Protection against PCA for different treatment groups](image)

**Fig 6.4** Effect of milling on in-process withdrawn samples of *Shwaskuthar Rasa* on passive cutaneous anaphylaxis in rats
6.3.2 Study of passive paw anaphylaxis in rats

**Animal** : Rats

**Species** : Albino

**Age/Weight** : Adult / 100-150 gm

**Sex** : Male

**No. of animals**: 36 (Six animals per group)

**Preparation of drug solution**: *Shwaskuthar Rasa* (23 mg/kg, p.o.) suspensions were prepared using 2 % v/v Tween 80.

Group-I animals received 2 % v/v of Tween-80 (10 ml/kg p.o).

Group-II, III, IV, V and VI were treated with *Shwaskuthar Rasa* samples - SWR 1220, SWR 829, SWR 574, SWR 216 and SWR 92 (23 mg/kg, p.o) respectively.

**Procedure**: Albino rats were injected subcutaneously the doses of 100 μg of egg albumin on day 1<sup>st</sup>, 3<sup>rd</sup> and 5<sup>th</sup>. On 10<sup>th</sup> day of sensitization, blood was collected by retro-orbital plexus and centrifuged to separate serum containing IgE antibodies. Right foot pad of the male rats was sensitized with 0.1 ml of rat homologous antiserum after 24 hrs and in-process withdrawn samples of *Shwaskuthar Rasa* were administered to all groups except control. Animals were again challenged with 1 % egg albumin and edema inhibition was calculated with Plethysmometer according to **Penna et al., 2003**.
Fig 6.5 Effect of milling on in-process withdrawn samples of *Shwaskuthar Rasa* on passive paw anaphylaxis in rats.
6.4 RESULTS AND DISCUSSION

In vivo study using experimental animal model was performed to evaluate the effect of pulverization of Shwaskuthar Rasa. The in-process samples of Shwaskuthar Rasa from planetary ball mill at schedule time intervals of 0, 6, 12, 18 and 24 hrs, were withdrawn and these samples representing preparation of different particle size were subjected to evaluate their therapeutic value for antiasthmatic and antiallergic activity on experimental animal models.

Histamine induced bronchospasm in guinea pigs

Bronchial asthma is a chronic inflammatory disease characterized by both bronchoconstriction and airway inflammation which leads to bronchial hyper responsiveness to various stimuli, in which many cell types play a role, more important being mast cells, eosinophils and T-lymphocytes. Different agonists like acetylcholine, histamine, 5-hydroxytryptamine and bradykinin are responsible for contractile responses.

Histamine is one of the major inflammatory mediators in the immediate phase of asthma, causing airway hyper responsiveness and bronchial airway inflammation. The data of study (Table 6.1 and Fig 6.1) reveal that although all withdrawn samples of Shwaskuthar Rasa caused the bronchorelaxation at given dose level compared to control. The maximum bronchorelaxation (76.09 %) was observed with sample SWR 92 followed by Shwashuthar Rasa samples - SWR 216, SWR 574, SWR 829 and SWR 1220 with 68.96 %, 56.80 %, 45.26 % and 41.23 % respectively for bronchorelaxant activity. Statistically highly significant (p< 0.01) mean exposition time was shown by SWR 92 as most effective drug, although the values for SWR 216 and SWR 574 were also highly significant (p< 0.01) followed by SWR 829, SWR 1220 as only significant (p< 0.05) compared to control (Table 6.1 and Fig 6.1). This study on one
hand proved the efficacy of *Shwaskuthar Rasa* as antiasthmatic agent, on other hand study clearly indicated the significant of particle size reduction as preparation of smaller particle size (SWR 92) exhibited more bronchorelaxant action compared to *Shwaskuthar Rasa* samples of higher particle size.

**Mast cell degranulation in mice**

Administration of clonidine resulted in significant peritoneal mast cell degranulation in mice. Pretreatment of mice with in-process withdrawn samples of *Shwaskuthar Rasa* resulted in significant reduction in degranulation of mast cell when challenged with clonidine and the percent protection was found to be 33.95 %, 46.71 %, 52.13 %, 64.53 % and 72.42 % for *Shwaskuthar Rasa* samples - SWR 1220, SWR 829, SWR 574, SWR 216 and SWR 92 respectively (*Table 6.2 and Fig 6.2*). Samples SWR 92, SWR 216 and SWR 574 showed statistically highly significant (p<0.01) whereas *Shwaskuthar Rasa* samples - SWR 574 and SWR 829 showed significant (p<0.05) level compared with control group (*Photograph 6.1*) in the reduction of degranulation of mast cells in mice.

Clonidine causes degranulation of mast cells like compound 48/80 and release of inflammatory mediators such as histamine, leukotrienes and prostaglandins etc. The mast cell degranulation and its correlation with the release of histamine after administration of compound 48/ 80, the mast cell degranulating agent was established (*Uvnas, 1969*). Both clonidine and compound 48/80 acts through the dynamic expulsion of granules without causing any damage to the cell wall. Clonidine releases histamine from mast cells in a similar manner to a selective liberator like compound 48/80 (*Lakadawala, 1980*). Different particle size of *Shwaskuthar Rasa* prevented degranulation of mast cells probably raising the cyclic adenosine monophosphate. It has been known that
Shwaskuthar Rasa may increase intracellular levels of Cyclic AMP, relaxes airway smooth muscle and inhibit the release of autacoids from the tissues and basophils.

**Clonidine induced catalepsy in mice**

In the present study, prior treatment with samples of different particle size of Shwaskuthar Rasa significantly inhibited the clonidine induce catalepsy in mice. The effect of Shwaskuthar Rasa on clonidine induced catalepsy may be due to its anti-histaminic activity (H₁ receptor antagonist activity). Clonidine (1mg/kg, s.c) produced catalepsy in mice, which remained for 3 hrs. The vehicle treated group has shown maximum duration of catalepsy at 120 min after the administration of clonidine. However, there was highly significant (p<0.01) inhibition of catalepsy with Shwaskuthar Rasa samples - SWR 92 and SWR 216 at 90 min and SWR 574 at 120 min. Significant (p<0.05) level of inhibition of clonidine induced catalepsy with SWR 829 and SWR 1220 at 120 min *(Table 6.3 and Fig 6.3)* was also noticed. The study indicated that particle size of preparation plays important role in reducing the duration of clonidine induced catalepsy i.e. the smaller the particle size better was the effect. Finally it can be said that the effect was particle size dependent.

**Passive cutaneous anaphylaxis in rats**

The passive cutaneous anaphylaxis test was used to evaluate the antiallergic activity of Shwaskuthar Rasa. The anti-PCA test is the most classic and commonly used experimental study. In passive cutaneous anaphylaxis test, antibody is administered passively by intradermal route in fresh rats and is allowed to fix on the mast cell membrane receptors. After 48 hrs of fixation of antibodies, Shwaskuthar Rasa samples of different particle size were administered orally (23 mg/kg) and again
antigen along with dye was administered intravenously in rats. Although all the samples of *Shwaskuthar Rasa* caused protection from passive cutaneous anaphylaxis it was most prominent 64.12 % with SWR 92 followed by other samples 56.33 %, 50.42 %, 41.82 %, 34.70 % with SWR 216, SWR 574, SWR 829, and SWR 1220 respectively. *Shwaskuthar Rasa* samples - SWR 92 and SWR 216 have shown statistically highly significant (p<0.01) and SWR 574, SWR 829 and SWR 1220 shown significant (p<0.05) level anti-PCA activity (Table 6.4 and Fig 6.4) compared with control group (Photograph 6.2). It suggests that all the withdrawn samples of *Shwaskuthar Rasa* are capable to block the release of mediators from the mast cells. Appearance of a blue wheel shows the extent of liberation of mediators by mast cell. Meaning thereby that larger the area of the wheel, higher will be the amount of mediators released. This study also revealed that reduction in particle size of *Shwaskuthar Rasa* caused increased level of ant-PCA activity.

**Passive paw anaphylaxis in rats**

Allergy is a chronic inflammatory process occurring due to exposure of allergen resulting in the activation of T-lymphocyte with subsequent release of inflammatory mediators. Immunomodulating agents are useful in the treatment of allergy by inhibiting the antigen-antibody (AG-AB) reaction and thereby inhibiting the release of inflammatory mediators. The data of study revealed that the maximum percentage protection of paw edema volume was observed in *Shwaskuthar Rasa* sample - SWR 92 at 4 hrs. As per statistical analysis of paw edema volume highly significant results (p<0.01) were observed from SWR 92, SWR 216 and SWR 574 with percentage protection 67.81 %, 62.06 %, 56.32 % respectively. And significant inhibition (p<0.05) in paw edema was recorded from samples - SWR 829 and SWR 1220 with
percentage protection 43.67 % and 31.03 % respectively (Table 6.5 and Fig 6.5) compared with control group (Photograph 6.3). The overall study revealed that decrease in particle size of Shwaskuthar Rasa enhances the protection against paw edema and the effect was particle size dependent meaning thereby as the size of the preparation is reduced the protective effect against paw edema increased.

*Shwaskuthar Rasa*, a reputed herbomineral formulation of Ayurveda, apart from many therapeutic potential is specially valued for respiratory problems with its specific effect on asthma and allergy. Study of in-process withdrawn samples of *Shwaskuthar Rasa* from planetary ball mill at schedule time intervals of 0, 6, 12, 18 and 24 hrs revealed that progressive milling gradually decreases particle size of the formulation. Subsequently, when these withdrawn samples were pharmacologically evaluated for their antiasthmatic and antiallergic potential using suitable animal models, it was revealed that maximum effect was exhibited by *Shwaskuthar Rasa* sample with smallest particle size i.e. SWR 92 and as the particle size of samples increases the efficacy also reduces nearly in same order. It is therefore concluded particle size reduction is directly proportional to absorption, digestion, assimilation followed by bioavailability and finally for therapeutic efficacy of *Shwaskuthar Rasa* preparation. Effect of antiasthma and antiallergy activities in experimental animals was fineness dependant i.e. smaller the particle size, better the therapeutic effect.

Therefore it can be concluded that efficacy of the *Shwaskuthar Rasa* increases as the particle size reduce which facilitated the drug to cross the biological barriers thus increasing the availability of the drug responsible for its enhanced antiasthma and antiallergic activity.