Evaluation of Anthelmintic Efficacy of Ethanolic Extract of *Punica granatum* in Goats

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ABSTRACT

As per the 19th animal census of India, total population of goat is around 135.17 million contributing 26.40% of total animal population. Maharashtra shares about 6.24% of the total goat population. Helminthic infections are one of the constraints reducing the productivity of these small ruminants and manifested by symptoms like anaemia, emaciation, enteritis, dehydration and death in kids. Chemotherapeutic agents mainly from benzimidazole group, imidathiazole and avermectins are used for the treatment of helminth infections. However, the cost effectiveness and increasing resistance to these chemotherapeutics is the largest field problem. Hence the present study was undertaken in order to evaluate the anthelmintic efficacy of *Punica granatum* in goats. Total 30 infected goats were divided into three groups comprising 10 animals in each. Pre-treatment EPG counts (Mean ±SE) of faecal samples from the groups A, B, and C were recorded as 665±177.81, 790±210.79 and 720±136.87 respectively. Group A consisted of ten helminthic infested goats (Mean EPG of 665). Group B animals (Mean EPG of 790) received treatment with fenbendazole @ 5 mg/kg b.wt orally. Group C animals (Mean EPG of 720) received treatment with ethanolic extract of *P. granatum* peel @ 200 mg/kg b.wt orally. Treatment group (B and C) showed significant (p≤0.01) increase in Hb, TEC, PCV and while the EPG of faeces and TLC were found to be decreased significantly (p≤0.01) at 15th day and 28th day in gastrointestinal nematodes infested goats

**Keywords:** *P. granatum, Benzimidazole, Anthelmintic, Goats*
INTRODUCTION
Livestock is an integral part of the farming system. In the traditional agricultural economy, livestock plays a crucial role in providing the most needed manure, valuable protein and food like meat and milk. India occupies second position in terms of goat population, fifth position in meat production, growth rate of meat production is 5.21% and situated at first position in milk production. Chevon (goat meat) is most preferred and widely consumed meat in the country contributing 14.25% to total meat production of India. Goats have earned a special place in the lives of poor and landless farmer and therefore it is also known as "poor man’s cow”. Population of goats is about 135.17 millions, which is 26.40 % of total livestock population in India out of these Maharashtra share 6.24 % involving about 79.71 lakh as per 19th census of India (FAOSTAT, 2014).

Helmintic infection of Gastrointestinal (GI) tract is one of the major causes of decreased productivity in goat worldwide, particularly under grazing conditions. GI parasitic infection in goat is characterized clinically by anaemia, emaciation, enteritis, dehydration and death in kids. Parasitic infection in goats also has an adverse effect on blood enzyme levels, which decreases goat production in different biological ways.

Punica granatum (P. granatum) a member of family Punicaceae, is a favorite table fruit of the tropical and subtropical regions of the world. It is native from Iran to the Himalayas in India and has been cultivated over the whole Mediterranean region since ancient times (Holland et al., 2009). Pomegranate can be grown throughout India but due to its better adaptability towards arid climate, the commercial cultivation is being done in Maharashtra, Karnataka and Rajasthan. Maharashtra is the leading producer of pomegranate in the country and accounts for 66.2% of the total production of pomegranate in the country. Maharashtra state is producing 0.49 million ton of pomegranate from an area of 0.08 million hector, which is the highest in the country.

Control of gastrointestinal nematode in goat is a big challenge due to the emergence of drug resistance, environmental pollution and food residues. Therefore, there is urgent need for alternate methods of control and to reduce worm burden, which should be less toxic, cheaper with wide margin of safety, easily available, and eco-friendly (Raje & Jangde, 2003). Hence the anthelmintic activity of P. granatum was investigated against GI roundworms in goats.

MATERIALS AND METHODS
The present investigation was carried out at Punyashlok Ahilyadevi Sheep & Goat Development Farm, Mahud, Tal- Sangola, Dist- Solapur to access in vivo evaluation of anthelmintic activity of A. indica in caprines by the egg per gram (EPG) count method before and after treatment.

Collection and Extraction of plant material: P. granatum plant was identified botanically and authenticated from the Department of Botany Shripatarao Kadam Mahavidyalaya, Shirwal. P. granatum peels were selected for the ethanolic extraction and extraction was done at Department of Pharmacology and Toxicology, Krantisinh Nana Patil College of Veterinary Science Shirwal Dist Satara.

Experimental animals: Osmanabadi goats (n=30) of 8 to 12 months age were randomly selected and divided in three groups comprising 10 animals in each. The experimental protocol was approved by the Institutional Animal Ethics Committee (IAEC) as per the guidelines of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA). The helmintic infection was confirmed before beginning of the study by fecal examination of each animal, as per the standard procedure described by Soulsby (1982). The animals having more than 600 eggs per gram (EPG) count were included in this experiment. All groups of experimental goats were housed in separate pens at Punyashlok Ahilyadevi Sheep Development Farm.
and Goat Development Farm Mahud, Tal-
Sangola, Dist- Solapur. The goats were
allowed free grazing daily early in the
morning. Then fed with fresh grass and
concentrate diet and ad-libitum potable
drinking.

Experimental groups and treatments:
Total 30 goats were selected on the basis of
EPG count and divided in three groups as A, B
and C. The experimental groups and schedule
of treatment are detailed in Table 1.

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Group</th>
<th>No of Animals</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>A</td>
<td>10</td>
<td>Control –untreated</td>
</tr>
<tr>
<td>2</td>
<td>B</td>
<td>10</td>
<td>Fenbendazole @ 5 mg/kg b.wt dosed orally once at day ‘0’.</td>
</tr>
<tr>
<td>3</td>
<td>C</td>
<td>10</td>
<td>P. granatum peels extract @ 200 mg/kg b.wt for first five days and on 16th day</td>
</tr>
</tbody>
</table>

Sample collection and Processing:
The egg per gram (EPG) count of faeces of
each fecal sample was determined by modified
McMaster Technique (Holland et al., 2009)
and calculated as follows:
Egg per gram (EPG) = Number of egg in the
chamber x 50

Anthelmintic efficacy was calculated by the
faecal egg count reduction (FECR) test (Raje,
& Jangde, 2003) according to the following
formula:

\[
\text{FECR \%} = \frac{\text{Pre-treatment EPG} - \text{Post-treatment EPG}}{\text{Pre-treatment EPG}} \times 100
\]

Statistical analysis
All the values in the test groups are presented
as Means ± SE and the data was analysed by
using Completely Randomized Design (CRD)
by using WASP software ‘P’ value p≤0.01 or
p≤0.05.

RESULTS AND DISCUSSION
General observations: The body weights
were taken on day ‘0’ (before start of
experiment) and day 28th i.e. termination day
of the experiment. All the treatment groups
found to be apparently healthy without any
side effects or behavioural signs throughout
the experimental duration

Phytochemical analysis: Phytochemical
analysis of ethanolic extract of P. granatum
peels had showed the presence of alkaloids,
reducing sugars, tannins and glycoside.

Anthelmintic efficacy: Anthelmintic efficacy
of P. granatum compared with control and
standard group is depicted in Table 2. Pre-
treatment EPG counts (Mean±SE) of fecal
samples from the groups A, B and C were
recorded as 665±177.81, 790±210.79 and
720±136.87 respectively. Pre-treatment EPG
count (Mean±SE) among the groups did not
vary significantly at p≤0.05. Control group A
did not show any significant variation in EPG
count on 15th and 21st day of the study.

Pre-treatment EPG count (Mean±SE) in group B (standard drug) was 790±210.79.
EPG counts on days 15 and 28 post treatment
were 125±39.612 and 100±34.155
respectively. Statistically significant decrease
(p≤0.01) in fecal egg counts were recorded on
day 15 and day 28 post treatment when
compared with pre-treatment EPG count. Post
treatment FEC reduction was 84.2% on day 15
and 87.4% on day 28.

The present findings are further
supported by the previous studies of Singh [9]
who reported that fenbendazole @ 10 mg/kg
b.wt orally achieved 99.52% reduction in fecal
egg count of infected goats on 13th day post-
treatment. The animals treated with
fenbendazole revealed 83% efficacy by fecal
egg count reduction test in goats [8]. Goats

treated with fenbendazole at a dosage rate of 5 mg/kg body weight orally achieved 71.08% reduction in FEC on 14th day post treatment Lalhmingchhuanmawii et al. (2014) reported that fenbendazole exhibited 85 and 100 % efficacy on day 7th and 14th of post treatment.

Pre-treatment EPG count (Mean±SE) in group C was 720±136.87. EPG counts of faeces on days 15 and 28 post treatment were 210±76.666 and 180± 64.637, respectively. Significant reduction in fecal egg counts were recorded on day 15 and day 28 post treatment when compared with pre-treatment count (p≤0.01). This fecal egg count reduction was 69.4% on day 15 and 73.7% on day 28.

The present findings are further supported by the previous studies of Boonmasawai (2013) who reported that alcoholic extract from pomegranate peels reduces worm egg production significantly at day 1, 3 and 7 (45 ± 11%, 57 ± 14% and 64 ± 7%, respectively) against gastrointestinal nematodes of infected goats.

Similarly Lalhmingchhuanmawii et al. (2014) also reported that ethanol extract of *P. granatum* rind achieved maximum significant reduction (97.95 %) in EPG count on day 21 post-treatment with 50 mg/ml concentration of extract against paramphistomes infected sheep.

**CONCLUSION**

*P. granatum* (pomegranate) is proved to be a good anthelmintic activity and further studies should be conducted in order to make a feasible pharmaceutical formulation.

### Table 2: EPG count of different groups on Pre-treatment Day (-7th) and post treatment 15th and 28th day

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatment</th>
<th>Mean EPG count ± (Mean SE)</th>
<th>Day 15th</th>
<th>Day 28th</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Pre-treatment Day (-7th)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>A</td>
<td>Control</td>
<td>EPG count</td>
<td>665± 177.81&lt;sup&gt;a&lt;/sup&gt;</td>
<td>745±186.26&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>B</td>
<td>Fenbendazole</td>
<td>EPG count</td>
<td>790± 210.79&lt;sup&gt;b&lt;/sup&gt;</td>
<td>125± 39.61&lt;sup&gt;bc&lt;/sup&gt;</td>
</tr>
<tr>
<td>C</td>
<td>Ethanol extract of <em>Punica granatum</em></td>
<td>EPG count</td>
<td>720± 136.87&lt;sup&gt;c&lt;/sup&gt;</td>
<td>275± 95.23&lt;sup&gt;bc&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Fig. 1: Groupwise EPG count on Day 7, 15 and 28th of study
Acknowledgement
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REFERENCES


