Clinical Studies on The Efficacy of Different Topical Drugs With Ivermectin Injection in The Treatment of Egyptian Water Buffalo Calves Infested with Psoroptes Spp. Mites

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ABSTRACT
A total of thirty water buffalo calves at smallholder farm suffering from skin lesions and clinical signs compatible with mange were investigated clinically and parasitologically. The efficacy of three topical drugs with ivermectin in the treatment of psoroptes mites was investigated. Infested calves were randomly allocated into four groups (6 calves each) in addition to a positive control group. The four uncontrolled groups were treated with a single subcutaneous injection of ivermectin at the rate of 1mL/50kg body weight. Three groups were treated with different topical agents including benzyl benzoate 25%, crotamiton 10% and permethrin 5% once daily for two consecutive days. The number of live mites in skin scrapings was obtained before and after treatment for eight weeks. Severity of skin lesions was evaluated and scored. The synergistic effect of each topical agent with ivermectin were compared for eliminating mites, aiming to obtain the ideal treatment model. Mite counts of the treated animals with ivermectin plus permethrin were significantly (p < 0.05) lower than those of the ivermectin plus benzyl benzoate at 2 weeks post-treatment. The efficiency of ivermectin plus permethrin was found to be significantly more effective in the treatment of mange even in the first days after application compared with other groups. Topical permethrin with ivermectin injection is a good combination for treatment of psoroptes mites infestation in buffalo calves.

1. INTRODUCTION
Ectoparasites, including mites, are responsible for great economic losses in livestock industry (Jabeen et al., 1998). It is a noticeable disease in most countries including Egypt where environmental and unhygienic conditions are favourable for its growth and transmission especially in small feedlots or barns related to rancher (Yassin, 2011). Most calves in these places do not get suitable prophylactic drugs against ectoparasites and the owners have rare idea about the importance of ectoparasites control. One of the most important ectoparasites is Psoroptes mite that responsible for mange disease in animals (Losson and Lonneux, 1996). Mostly, It can be combated by using anthelmintic.

The anthelmintic activity of the avermectins was first detected in 1979 (Miller et al., 1979). Ivermectin is a pioneering drug, a dihydro derivative of avermectin, originating from a microorganism isolated in Tokyo and Japan. Originally introduced as a veterinary drug for killing a wide range of internal and external parasites in commercial livestock and companion animals (Crump and Ōmura, 2011), Ivermectin is effective against parasites in a wide variety of hosts including sheep, cattle, swine, horses and dogs (Fink and Porras, 1989). Intensive use of ivermectin can lead to resistance which has been documented for some intestinal helminthes (Soutello et al., 2007). Resistance also noted against mites (Clark et al., 1995; Currie et al., 2004). From this point of view, it seems that ivermectin need synergistic product for rapid treatment of domestic animal ectoparasites. Benzyl benzoate, crotamiton 10% (Eurax) and permethrin 5% are common antiscabietic agents that can be used.

Benzyl benzoate lotion is a rapidly acting agent against mites when applied to the skin for 24 hours. Benzyl benzoate has been used effectively in the form of ester of benzoic acid and benzyl alcohol. The main disadvantage of benzyl benzoate is skin
irritation within minutes after its application (Walton et al., 2000; McCarthy et al., 2004). Crotamiton 10% (Eurax) is active antiscabetic agents with low toxicity profile. A satisfactory response need multiple applications (Taplin et al., 1990). Permethrin 5% is a synthetic pyrethroid insecticide that is well tolerated, has low toxicity, rapid metabolism but more expensive than other agents (Strong and Johnstone, 2007).

Water buffaloes are more reared than cattle in rural area of Egypt for it is productivity, thus the objectives of this a prospective study were to investigate Psoroptes spp. mite infestation in buffalo calves that were kept at a private buffalo farm, and to compare the synergistic effect of some topical drugs with ivermectin injection in the treatment.

2. MATERIALS AND METHODS

2.1. Animals

Thirty Egyptian water buffaloes male calves, suffering from skin lesions were included in the study. Animal history includes introduction of two infested calves, nine month age, showing skin lesions into feedlot include 28 male calves aged 8-14 months. The clinical signs were compatible with mange. The barns are closed and made from mud. The calves shared food and water places. After introduction of infested animals, the infestation rate began to increase gradually in animals and within one week all calves showing clinical signs of restlessness and itching.

2.2. Clinical and parasitological examination

On the first visit to the infested barn, all animals showed varied degree of itching, scales, hair loss and dermatitis. The skin lesions were recognized and recorded for each animal. Deep skin scrapings were collected from all animals with precaution according to method given by Tikaram and Ruprah, (1986). Samples were brought to the laboratory and examined microscopically according to Soulsby, (1983). The parasite identified according to the characters showed by Sanders et al., (2000). The severity of infestation was graded from mild to moderate and severe according to the results of clinical examination that focus on pruritus, alopecia, scales, erythema and abrasion degree.

2.3. Grouping of animals for treatment protocols

The infested calves were randomly allocated into 4 groups separated from each other in different barns, each group contained 6 animals plus a positive control group. All barns were treated with butox (Deltamethrin B.P (Vet)) 12.5 mg. All groups were given the same tonics, antihistaminic, and anti-inflammatory therapy. Animals in group B were treated with ivermectin (Ivomec; Merial, France) subcutaneously at dose rate of 0.2 mg/kg body weight. Group C animals were treated with ivermectin and topically sprayed with benzyl benzoate 25% lotion (Pharaonia). Group D were treated with ivermectin and topically sprayed with EURax 10% lotion (crotamiton 10%, Novartis). Group E were treated with ivermectin and topically sprayed with ectomethrin 5% lotion (permethrin 5%, Arab Perfum). All topical drugs were applied once daily for two consecutive days.

2.4. Mite counts and mange lesion scores for treatment evaluation

Before administration of the drugs, on day 0, the numbers of live mites were counted from a total of 10 skin regions of each animal on approximately 3 cmx3 cm of body surface for each region that determined through previously designed frame and marked by black pen. Scrapings collected from the edges of active lesions or, from the apparently recovered one, if it began to be limited after beginning of treatment. A sharp blade dipped in mineral oil was used to collect the sample till blood oozing. Each collected scrape of each animal were collected in separate plate containing mineral oil and closed tightly, secured by tape, labelled and immediately transported to laboratory for counting. Sometimes, sample clearing, if it is necessary, was performed using potassium hydroxide 10%, in this case all mites observed after clearing were recorded as live (Shanks et al., 2000). Parasitological and clinical follow up were performed on day 1, 7, 14, 28, 42 and 56 after treatment following the same steps. Ten different spots on slides were examined from each scraping. Thick scales were avoided as it may hide mites under and any slight movement under the fine scales counted to be active mite. The slides spots were covered by a cover slip and examine microscopically under low magnification power (10x objective). Total mite counts of each animal related to each follow up equal the summing of the counts from each scraping site.

2.5. Statistical analysis

The obtained results analysis performed according to Visser et al., (2013) in which the number of living mites at each time point was transformed to the natural logarithm of (count + 1) for geometric means (GM) calculation. In order to assess the influence of anti-parasitic drug on mite counts on the first examination day were compared among groups using repeated measures ANOVA with time as the within-subjects factor and drug as between-subject factors. Wilks’ Lambda test was used to determine the main effect of drugs and time.
In addition to time and drug interactions. Where Wilks’ Lambda test indicated a statistically significant difference between drug groups, one-way ANOVA with Tukey HSD post-hoc multiple comparison test was used to identify which group was statistically different from the rest. Differences between means at P<0.05 were considered significant. Statistical analysis was carried out through the general linear models (GLM) procedure of the statistical package for social sciences version 22.0 (IBM Corp., Armonk, NY, USA). Efficacy was calculated as 100 [(C − T)/C], where C is the GM for the control group and T is the GM for the treated group. A two-sided test was used at the 0.05 significance level.

3. RESULTS

3.1. Clinical signs

Clinical investigations revealed itching between animals against each other and objects. Itching lead to small abrasion and wound in different body parts, mild to severe scales, skin erythema, and hair loss. The lesion location differs between each animal (Figure 1 and Table 1). Some animals (3 out of 30) showed slight edema in dewlap. Mites infestation was more common on the back, followed by wither, under tail, thigh, rump, head and ear, neck and scrotum in descending order (Table 1). The most prominent lesion among animals were moderate in severity according to the selected categories and parameters.

3.2. Parasitological examination

Skin scrapings revealed presence of Psoroptes spp. mites (Figure 2). The severity of infestation in each animal scored to be mild, moderate and severe depending on the accurate observation and examination of animals. Majority of calves were moderate infested, while only a few calves showed severe infestation (Table 2).

3.3. The efficiency of topical antiscabietic agents

Most of the treated animals showed a good to excellent improvement, with complete healing of cutaneous lesions and partial or total re-growth of hair. The control group remained positive for mange mites throughout the course of study. On clinical level, there was no any adverse effect for any of the used drugs. Improvement was observed rapidly in the groups treated with ivermectin plus permethrin and ivermectin plus benzyl benzoate compared with that of ivermectin injection only or ivermectin plus crotamiton. Ivermectin plus permethrin showed faster effect than ivermectin plus benzyl benzoate at day 14 post treatment (Table 4). The efficiency of ivermectin plus permethrin was the highest among all the treated groups even in the first day after application (Table 3). Treatment outcomes of 24 buffalo calves revealed that permethrin 5% gave the highest synergistic effect followed by benzyl benzoate 25% then crotamiton.

<p>| Table (1): The site of lesions in different body parts of infested animals. |</p>
<table>
<thead>
<tr>
<th>Site of lesion</th>
<th>No. of infested animals</th>
<th>Percentage (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Head and ear</td>
<td>4</td>
<td>13.3</td>
</tr>
<tr>
<td>Neck</td>
<td>3</td>
<td>10</td>
</tr>
<tr>
<td>Dewlap</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Wither</td>
<td>15</td>
<td>50</td>
</tr>
<tr>
<td>Shoulder</td>
<td>10</td>
<td>33.3</td>
</tr>
<tr>
<td>Chine</td>
<td>20</td>
<td>66.6</td>
</tr>
<tr>
<td>Loin</td>
<td>20</td>
<td>66.6</td>
</tr>
<tr>
<td>Rump</td>
<td>5</td>
<td>16.6</td>
</tr>
<tr>
<td>Thigh</td>
<td>5</td>
<td>16.6</td>
</tr>
<tr>
<td>Crops</td>
<td>17</td>
<td>56.6</td>
</tr>
<tr>
<td>Leg</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Under tail</td>
<td>8</td>
<td>26.6</td>
</tr>
<tr>
<td>Scrotum</td>
<td>2</td>
<td>6.6</td>
</tr>
</tbody>
</table>
Table (2): Number of animals suffered from different lesion grades at first clinical examination.

<table>
<thead>
<tr>
<th>Lesion</th>
<th>Mild</th>
<th>Moderate</th>
<th>Severe</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pruritus</td>
<td>Itch itself one time per 10-15 M</td>
<td>Itch itself one time per 5-9 M</td>
<td>Itch itself one time per 30 sec- 4 M</td>
</tr>
<tr>
<td>Alopecia</td>
<td>Less than 3 sites all over the body with partial loss of hair</td>
<td>3-6 sites all over the body with partial loss of hair</td>
<td>More than 6 sites all over the body with partial and/or complete loss of hair</td>
</tr>
<tr>
<td>Scales</td>
<td>Size less than 2 cm in less than 4 sites all over the body</td>
<td>Size 2-4 in 4-8 sites all over the body</td>
<td>Size more than 4 in more than 8 sites all over the body</td>
</tr>
<tr>
<td>Erythema</td>
<td>Less than 2 sites all over the body</td>
<td>2-5 sites all over the body</td>
<td>More than 5 sites all over the body</td>
</tr>
<tr>
<td>Abrasion</td>
<td>Less than 2 sites all over the body</td>
<td>2-4 sites all over the body</td>
<td>More than 4 sites all over the body</td>
</tr>
</tbody>
</table>

Table (3): Efficacy of ivermectin with and without topical treatment against naturally infested buffalo calves by *Psoroptes* spp. mites.

<table>
<thead>
<tr>
<th>Day after treatment</th>
<th>Control</th>
<th>IV only</th>
<th>IV+ Crotamiton</th>
<th>IV+ Permethrin</th>
<th>IV+ Benzyl benzoate</th>
<th>Efficacy</th>
</tr>
</thead>
<tbody>
<tr>
<td>D1</td>
<td>1.85±0.02</td>
<td>1.71±0.03</td>
<td>1.74±0.06</td>
<td>1.65±0.06</td>
<td>1.72±0.03</td>
<td>7.0</td>
</tr>
<tr>
<td>D7</td>
<td>1.84±0.03</td>
<td>1.58±0.05</td>
<td>1.48±0.05</td>
<td>1.45±0.06</td>
<td>1.28±0.06</td>
<td>29.3</td>
</tr>
<tr>
<td>D14</td>
<td>1.85±0.01</td>
<td>1.34±0.07</td>
<td>1.03±0.22</td>
<td>0.86±0.28</td>
<td>0.97±0.2</td>
<td>47.6</td>
</tr>
<tr>
<td>D28</td>
<td>1.91±0.1</td>
<td>0.88±0.18</td>
<td>0.41±0.16</td>
<td>0.18±0.18</td>
<td>0.16±0.16</td>
<td>91.6</td>
</tr>
<tr>
<td>D42</td>
<td>1.88±0.03</td>
<td>0.53±0.24</td>
<td>0.16±0.16</td>
<td>0±0</td>
<td>0±0</td>
<td>100.0</td>
</tr>
<tr>
<td>D56</td>
<td>1.92±0.02</td>
<td>0.45±0.29</td>
<td>0.19±0.10</td>
<td>0±0</td>
<td>0±0</td>
<td>100.0</td>
</tr>
</tbody>
</table>

- **IV** = Ivermectin
- Geometric mean counts (based on transformation to the natural logarithm of [count + 1]).
- Efficacy = 100 × (GM control − GM Treated) GM control)
- Wilks' Lambda test for drug x time interaction, P<0.05

Table (4): Number of treated and untreated animals pre and post administration of drugs against naturally infested buffalo calves by *Psoroptes* spp. mites.

<table>
<thead>
<tr>
<th>Day after treatment</th>
<th>Groups</th>
<th>N</th>
<th>0</th>
<th>7</th>
<th>14</th>
<th>28</th>
<th>42</th>
<th>56</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>Control</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>B</td>
<td>IV only</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>4</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>C</td>
<td>IV+ Crotamiton</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>5</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>D</td>
<td>IV+ Permethrin</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>4</td>
<td>2</td>
<td>5</td>
</tr>
<tr>
<td>E</td>
<td>IV+ Benzyl benzoate</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>5</td>
<td>1</td>
<td>5</td>
<td>6</td>
</tr>
</tbody>
</table>

- **IV** = Ivermectin; **N** = Number of animals in the group; **P** = Number of untreated animal pre administration of drug
- **A** = Number of treated animal after administration of drugs
4. DISCUSSION

Significant effect of age and sex was not examined in this study as the infestation occurs in feedlot males with the same age range. A greater insight into the epidemiology of *Psoroptes* mites infestation in Egyptian animals especially (cattle, buffaloes, sheep and goats) is indeed necessary in order to propose the significant effect of animal species, age, sex on its prevalence. The detected oedema in three of severe infested animal may be attributed to significant decrease in total protein, albumin and globulin that associated with mites infestation (Elkhtam and Mousa, 2016).

The rapid infestation rate in all calves may be attributed to the bad management, poor barn condition, absence of proper and regular antiscabietic application, and the overall non-scientific bases of rearing conditions (Mustafa et al., 2009). Age of the calves (8-14 months ) and season of occurrence may also have influence on infestation (Jabeen et al., 1998). It was supposed that the less developed immune system of the calves may be the cause of the higher ectoparasitic prevalence. The clinical examination revealed that low infestation was detected in head, ear and neck. No lesions were detected in leg or dewlap. The back region (Loin and Chine) was the most affected part as previously showed by Elkhtam and Mousa, (2016). Most animals suffered from moderate infestation according to the selected categories and parameters (Table 2). Poor general performance and weight loss were correlated to the severity of the infection (Munangandu et al., 2010). The rubbing and scratching of the affected areas in an attempt to alleviate the itchiness, results in the shedding of mites in the surroundings for subsequent transmission to other animals. Excessive rubbing and scratching lead to excoriation and haemorrhage on the skin surface.

A rapid effective treatment is strongly requested to obtain a quicker healing of lesions that annoying the animal and limit the infection spreading in the same herd. Ivermectin appeared to be well known drug for the treatment of common parasitic diseases including mange in buffaloes (Crump and Önura, 2011). Efficacy of the drugs was monitored for 56 days post-treatment. Efficacy of ivermectin in the present study was 76.6%. In the previous studies by Gill et al., (1989); Maqbool et al., (1995) and Hayat et al., (1996), the efficacy of
ivermectin against mange in buffaloes was ranged between 90–100% while in Kazmi et al., (2009) the efficacy was 86.66%.

It seems that the ivermectin loss some of it is efficacy with time, which may be due to parasite resistance (Currie et al., 2004). Thus, using of antiscabietic drugs combination can overcome drug resistance or treatment failure (Karthikeyan, 2005). Environmental treatment with butox was used to minimize the re-infestation chance (Kotb and Abdel-Rady, 2011). The permethrin preparation have ability to kill mites and its eggs, with non existence of toxicity potential due to extremely low absorption rate (Pourhasan et al., 2013). However, the combination of permethrin with other acaricides is in need due to potential of resistance development (Oberoi et al., 2007). The 53.5% efficacy of ivermectin plus permethrin at 14 day post application made the permethrin a welcome addition to the available therapies for mange. It has no irritating effect on skin, no objectionable odor, easy to use, cheap in price and unstable (Goldust et al., 2013). The adverse effect including swelling, redness or itching after application that previously attributed to absorption of dead parasite proteins was not detected (Ranjkesh et al., 2013). More study is in need to detect the efficacy of permethrin alone as antiscabietic agent in buffaloes. Combination of ivermectin and permethrin was the most effective treatment of choice for psoroptic mange in this study.

Crotamiton is drug used in lotion form for the treatment of scabies and pruritus (Roth, 1991). Topical ivermectin is more effective than crotamiton 10% cream but regarding side effects, crotamiton was found to be significantly safer than ivermectin (P < 0.05). Ivermectin continued to decrease both the lesions, and the degree of pruritus as compared to crotamiton and this difference was statistically significant (P < 0.05) (Goldust et al., 2014).

Benzyl benzoate has a powerful quick killing effect on mites. It can kill all mites within 15 minutes with the 0.8% dilution, 1h with the 0.4% solution (Kalpaklioglu et al., 1996). In this study no adverse effect was noted for benzyl benzoate however in human it can cause irritations and the post-scabious eczematous reactions were increased after treatment (Haustein and Hlawa,1989). In cattle, a rare case reports of benzyl benzoate irritating effect was detected (Pullin, 1956).

5. CONCLUSION

Mites are the most common cause of infectious skin diseases. The most challenging task is not the diagnosis but the treatment and recovery.

It seems that using multiple drugs with different antiparasitic mechanisms against mites can be effective to treat mange mites. The application of permethrin spray with ivermectin was recommended as a highly effective powerful acaricides in comparison with ivermectin alone or in combination with crotamiton or benzyl benzoate. Finally, animals rearing in poor rural areas need increasing attention from agriculture authority to put the proper deworming and acaricidal application programmes. In that way the owners can be protected from high loss of their reared animal production. More studies of host immune response, the mites biology, mechanisms of drug resistance, vaccine development, safety and efficacy of current and new acaricides are still in need to decrease the gap in our knowledge.

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7. REFERENCES


Munangandu, H.M., Siamudala, V.M., Matandiko, W., Munyeme, M., Chembensof, M., Mwase, E. 2010. Sarcoptes mite epidemiology and treatment in African buffalo (Synceruscaffer) calves captured for translocation from the Kafue game management area to game ranches. BMC Vet. Res. 6: 29-33.


Soulsby EJL. Helminths, arthropodes and protozoa of domesticated animals, 7th ed. USA: Lea &Febiger; 1983.


