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Expedited management of ulcer, colic and diarrhea in 209 horses: An open-labeled observational study of a potency-enhanced sucralfate-like elm phyto-saccharide

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A potency-enhanced polyanionic phyto-saccharide of elm mucilage (PEPPS) was prescribed in 209 horses in an open-labeled field trial. Clients provided informed consent to 32 equine veterinarians to prescribe PEPPS for low grade colic and diarrhea unresponsive to deworming. Most of the horses (n = 175/209) were presumed to have ulcers clinically, while 23/209 were confirmed by gastroscopy to have ulcers and 11/209 horses had colic related to diarrhea. A 4-day/7-dose response rate, determined by veterinarians’ consensus, provided the threshold for a significant clinical outcome, and data was collected through phone interviews over period of 3.5 years. 166/198 horses with ulcer associated colic and 10/11 horses with diarrhea-associated colic responded to PEPPS within 4 days or 7 doses. Using PEPPS alone was associated with positive outcomes in 81% (29/36) of horses. PEPPS added to failed regimens of omeprazole, ranitidine and antacids was associated with positive outcomes in 85% (137/162) of horses. Accelerated healing of ulcers occurred in 8 horses within 14 to 20 days using PEPPS alone and in 15 horses using PEPPS added to failed treatment regimens of omeprazole, ranitidine and antacid. PEPPS appears useful for managing equine ulcer, diarrhea and colic. As an electronegative polyanionic saccharide with substantial muco-adherence, PEPPS was compared to sucralfate. However, a randomized blinded placebo controlled trial is needed to quantify true clinical efficacy.

Key words: Equine, ulcer, diarrhea, colic, elm, sucralfate.

INTRODUCTION

Timely management of disruptive gastrointestinal (GI) symptoms poses a challenge to veterinary physicians (Mair et al., 2002; Tams, 2003). Inappetence, ulcer colic and diarrhea in horses can lead to more severe problems. Therefore, restoration of normal GI function is paramount and management should be expedited. The problem of disruptive equine GI symptoms is fairly significant. Fifty to 92% of the 9.2 million horses in the US (American Horse Council, 2005) have gastric ulcers, depending on their breed and type of containment or athletic activity (Hammond et al., 1996; Mairs et al., 2002; Murray et al., 1996; USDA APHIS, 2001a; Vatistas et al., 1994). In “gastroscopy clinics” conducted over 12 months in 25 states in the US, 60% of 658 horses were found to have ulcerations (Knudsen, 2009). Symptomatic ulcers usually present as low grade colic with food avoidance or inappetence. Inappetence is a principal sign to horse owners of colic and often serves as an early warning for the presence of ulcers. When surveyed, horse owners in Minnesota ranked ulcer colic 5th out of 25 top equine healthcares concerns (Martinson et al., 2006).

Abbreviations: GI, Gastrointestinal; HPS, high potency sucralfate; PEPPS, potency-enhanced polyanionic phyto-saccharide; USDA, United States Department of Agriculture; FDA, food and drug administration.
The USDA National Economic Cost Report on colic (2001b) reporting no geographic variation in the incidence of colic in horses, imply that this sentiment of concern for colic could be generalized to all horse owners. General prevalence positions ulcer colic as a significant disruptive GI symptom in horses and one that require effective management (Hillyer et al., 2001; Kaneene et al., 1997; Tinker et al., 1997; Traub-Dargatz et al., 2001).

Evidence-based treatment of equine diarrhea and nonsurgical colic is limited. However, some published reports (Galvin et al., 2004) suggest that colic can be treated with pain medications such as xylazine (0.5 mg/kg) or butorphanol (0.01 mg/kg). The use of an anti-spasmodic is a reasonable option as well. The initial management of diarrhea entails reducing the mechanical load of the colon chiefly by elimination of large volume fiber sources. Low bulk diet is complimented by the use of psyllium, corn oil and bismuth/salicylate suspension by nasogastric tube which is a current management of equine mucosal erosions, ulcerations and resulting colic and inappetence centers on therapeutic control of acidity (Andrews, 2005; Hammond et al., 1996; Hillyer et al., 2001; Knudson, 2009; Merial, 1999). Acidity is either neutralized by antacids (Andrews, 2005), reduced by histamine-2 blockers (for example, ranitidine, cimetidine, famotidine) (Andrews, 2005) or inhibited by proton pump inhibitors (for example, omeprazole) (Anderson, 2005; Knudson, 2009; Merial, 1999). Neither of these therapeutic agents provides direct relief of colic or direct healing of the mucosa. Instead, colic relief and ulcer healing are secondary consequences of lowering acidity. There are genetically controlled mucosa-specific mechanisms unrelated to acidity that are primarily responsible for direct healing (Tarnawski et al., 1998).

Proof of the presence of such mechanisms is found in FDA trial data on omeprazole (Merial, 1999). There was significant rate of healing observed in horses randomized to sham treatment. While acidity significantly hampered the rate of healing, nevertheless, complete healing occurred in 40% of untreated horses. This of course, was not a "mind over matter" placebo effect, but rather a genetically orchestrated phenomenon that is entirely independent of gastric pH (Jones et al., 1999; Konturek et al., 1991; Pai and Tarnawski, 1998; Skov, 1988; Tarnawski et al., 1998). Sucralfate is a polyanionic saccharide with characteristics similar to sucralfate, being muco-adherent, preferentially engaging mucosa-adherent growth factors and capable of repolarization of voltage-gated nocceptors related to acid pain and nausea. Though PEPPS has been prescribed by veterinarians in the US since 2003 for equine ulcer colic and diarrhea, its use on a large scale has never been formally reported. This report presents observational data in a proof of principle study which may be useful (Dreyer et al., 2010) in prescribing PEPPS in the setting of supervised treatments in an equine practice. Of course, any therapeutic intervention positively associated with the management of inappetence, ulcer colic and diarrhea would be a constructive development.

MATERIALS AND METHODS

Potency enhanced polyanionic phyto-saccharide

Elm mucilage is a polyanionic phytosaccharide (Upton et al., 2011). Unlike sucralfate, PEPPS contains no aluminum or sulfate. It is chiefly a high molecular weight mucilage (> 200,000 Daltons), comprising of galactose-rhamnose disaccharides. Potency-enhanced elm phyto-saccharide is prepared by suspending elm
mucilage in an anion-cation solution similar to that used to formulate high potency sucralfate (HPS) (McCullough, 1995, 2010, 2012). The resultant potency-enhanced phyto-saccharide (PEPPS) is muco-specific and capable of attaining augmented surface concentration of slippery elm. With sucralfate, potency enhancement ranges from 7 to 23 fold 3 h post-administration, having a lower fold increase on normal GI lining and higher fold increase on inflamed or injured mucosa. The exact post-administration surface concentration of PEPPS is unknown. However with PEPPS, the concentration of elm USP administered is less than 2% of slippery elm dose recommended by equine veterinarians (Veterinary Desk Reference, 2011; Ward, 2010). The formulary strength of Elm USP for horses is 1.9%. Administration of PEPPS is in accordance to weight. On average, horses weighing less than 500 lbs received daily doses upwards of 270 mg, those between 500 to 900 lbs received 380 mg and over 900 lbs received 540 mg. PEPPS was administered separate from other medications.

Dosing administration

Participating veterinarians prescribed PEPPS in accordance to weight-dose chart in label instructions. Horses weighing less than 500 lbs (227.3 kg) received 15 ml, between 500 to 900 lbs (227.3 to 409.1 kg) received 20 ml, and over 900 lbs (409.1 kg) received 30 ml. PEPPS was given by drench into cheek pouch twice daily, morning and evening. PEPPS is not absorbed systemically and act as an internal topical coating the GI lining.

Study design and controls

This study was an open labeled non-blinded observational trial. Observational trials are fundamentally distinct from a randomized controlled clinical trial, the least of which there is no control group. Merit is assessed on the basis of outcomes resulting from the introduction of an intervention into a pre-existing clinical setting (Hannan, 2008). Information was collected regarding; the age of horses, the nature of their GI symptoms (the presence of ulcer-suspected colic and diarrhea in horses), and type of treatment regimens prescribed by the practitioner at time of adding PEPPS. The length of illness is not reported. As an observational study, treatment intervention was not randomized. By design, differences in outcomes are observed without regard to similarities or dissimilarities of patient characteristics prior to treatment. In fact, in this type of study, treatment decisions were made by veterinarians prior to use of PEPPS; the selection of PEPPS being made by the veterinarian due to concern that pre-PEPPS treatments were ineffectual.

In this trial the question addressed is not one of the efficacies of PEPPS. Instead, the question addressed is one of the relative merits of PEPPS as a competing treatment or intervention. Outcome of merit is relative to the expectation of the participating veterinarians. As discussed below, a clinical response of 4 days or 7 doses merited note to the veterinarians involved. This study reports the percentage of horses with ulcer related colic and diarrhea related colic who responded to PEPPS while on failing therapies.

Comparative control

As an observational study, there were no control groups (Hannan, 2008). To provide a comparative “control” experience, each veterinarian was asked to reflect on their respective experience and select from a choice of a clinical response times which they would deem to deviate significantly from the expectations of their clinical experience. Most of the small animal veterinarians (80%) felt that a clinical response of 4 days or 7 doses would mark a significant departure from their clinical expectations, and this was based on their experience managing ulcer- and diarrhea associated colic in horses. This consensus of significant departure from expected time of clinical response was used to benchmark the primary outcome and a meaningful response. In essence, expectations of past clinical experience (replete with interventions requiring more time to work) served as a “comparative control” albeit a subjective one.

Consent

All animals were privately owned and owners’ consent was obtained by veterinarians.

Veterinarians participating in the study

Veterinarians placing orders for commercially available PEPPS were recruited to participate in this open-labeled trial. None had professional experience less than 5 years in practice. Veterinarians were familiar with using 2002 original formulation of equine Gastrafa® which contained 10% high potency sucralfate as the active ingredient. Following successful preliminary testing (Translational Medicine Research Center, 2002) high potency sucralfate was replaced in January, 2003 with calcium chelated elm mucilage in the form of PEPPS. Veterinarians were recruited from June, 2003 through December, 2006. All veterinarians prescribing PEPPS were engaged exclusively in primary care of equine animals within valid veterinarian-client-patient relationships. The combined years of practice for equine practitioners who completed the trial were 160. Each was experienced in the standards of care in treating ulcer- and diarrhea associated colic in horses. Out of 46 equine veterinarians, 32 practicing in 20 states of the US completed the study; the remainder lost to follow due to their inability to complete protocol. Veterinarians received no honorarium for their participation.

Sequential participation

Participation in the study was sequential, determined solely by the order of spontaneous requests for product made by veterinarians responding to notification of product’s availability. The veterinarians were self-selected. Information prompting orders pertain to the usefulness of PEPPS in the management of ulcers- and diarrhea associated colic in horses.

Inclusion/exclusion criterion for horses

Included in the observational trial were horses with low grade colic, inappetence or diarrhea for more than 5 days. These horses had failed adequate deworming therapies by owners. Horses were with and without a history of ulcers confirmed by endoscopy. Endoscopy was not required for inclusion. However, in accordance to customary practice of 8 veterinarians, 23 horses underwent endoscopy before and after treatment.

Test population

Horses of all age groups, breeds and ownership purposes were included. The size of the test population was 209 horses within a multi-practice-based study. There were 20 foals under 6 months, 38...
between 6 months to a year, 33 between 1 to 2 years and 118 over age 2. The test population was geographically diverse involving input from 20 out of the top 25 states with highest horse ownership and equine population. As was the custom for 8 of the 32 equine practitioners, 23 horses with ulcer-related colic underwent endoscopy prior to and following treatment with PEPPS.

Conditions managed by equine practitioners

Conditions to be managed in the trial included low grade (non-surgical) colic, ulcer-suspected colic and diarrhea. Low grade colic was heralded by poor appetite or refusal to eat – symptoms that failed to respond to owners’ use of dewormers or adjustments in feed. Ulcer-suspected colic was defined clinically as inappetence (poor feeding/drinking), poor attitude, dull coat and teeth grinding in foals. Diarrhea-associated colic was defined as symptoms identical to ulcer-suspected colic accompanied by loose stools for more than 5 days despite adequate de-wormer therapy by owners. Endoscopy in 23 horses was positive for gastric ulcers. Excluded were horses whose symptoms required surgical intervention.

Conduct of observational field tests

The study was conducted from June, 2003 through December, 2006. Participating veterinarians prescribed PEPPS in accordance to weight-dose chart in label instructions. The dose administration was three times daily for the first day then twice daily. Phone interviews were conducted with veterinarian staff to collect results of adding PEPPS to existing treatment regimens. Results were tabulated as either a positive or negative outcome.

Pre-existing treatment regimens used in study horses

Pre-existing treatment regimens for horses were fairly uniform and centered around four regimens. These were (a) omeprazole alone, (b) omeprazole + antacid, (c) histamine-2 blocker (ranitidine or cimetidine) + antacid or (d) histamine-2 blocker (ranitidine/ cimetidine) + antacid + bismuth suspension for diarrhea-associated colic. The antacid used was aluminum hydroxide/magnesium hydroxide combination. The length of time horses had been on these regimens was not assessed, therefore unknown. When clinical results from pre-existing regimens were deemed suboptimal, participating veterinarians added the polyanionic saccharide, PEPPS.

Horses treated with PEPPS alone

This open-labeled trial was observational with no control groups of untreated horses. However, a small number of horses, 36 of 209 formed a PEPPS only treatment group. Except for a few veterinarians treating the 36 PEPPS only group, previous experience of 80% veterinarians offered no expectation of significant response to a PEPPS intervention within 4 days or 7 doses.

Primary outcome measure in horses

There were two single symptom-related primary outcome measures for this trial. Cessation of ulcer colic and diarrhea were the two single symptom outcomes, respectively. Veterinarian-defined positive responses in horses with ulcer colic and diarrhea would be the restoration normal feed habits and the return of formed stools within 4 days or 7 doses of PEPPS, respectively. This veterinarian-defined response to therapy previously discussed represented a meaningful clinical difference for the management of ulcer colic and diarrhea in horses.

Hypothesis

The hypothesis is that majority of animals with serious and disruptive GI symptoms (of non-surgical etiology) when given PEPPS will experience resolution of symptoms within a timeframe (or dose administration), significant and relevant to the collective historical experience of practicing veterinarians who manage such symptoms routinely. This was a timeframe of 4 days (or in 7 doses).

Analysis

The trial used a per protocol analysis of the data. Both chi-square and two-sample t-test were used to compare percent response to treatment among horses at confidence level of 95 and 99% for confidence intervals.

RESULTS

General

Horses with ulcer-related colic and diarrhea-related colic

All horses eventually responded to PEPPS with variability in the clinical response times extending beyond 4 days. However, Tables 1 and 2 showed that 85% [CI 5.47 (CL 95%)] of 198 horses with ulcer-associated colic and 91% [CI 16.91(CL 95%)] of 11 horses with diarrhea-associated colic responded to PEPPS within 4 days or 7 doses. The percent response using PEPPS alone to manage ulcer-associated colic was 81% [CI 12.9/CL 95%, n = 36]. When PEPPS was added to existing treatment regimens, the percent response was high. Percent response was 83% [CI 10.11/CL 95%] when PEPPS was added to omeprazole (n = 53), 81% (CI 12.64/CL 95%) with PEPPS added to omeprazole + antacids (n = 37), 88% (CI 7.26/CL 95%) with PEPPS added to ranitidine/ cimetidine + antacids (n = 72), and 91% (CI 16.91/CL 95%) with PEPPS added to ranitidine/ cimetidine + antacids + bismuth (n = 11).

PEPPS alone compared to PEPPS augmented treatments

Table 3 shows the results of a two-sample t-test used to compare the percent response using PEPPS alone to the percent response using PEPPS augmented treatments. In each comparison, PEPPS alone versus (PPI + PEPPS), PEPPS alone versus (PPI + AA + PEPPS), PEPPS alone versus (H2B + AA + PEPPS), or PEPPS alone versus (H2B + AA + Bismuth + PEPPS), the t-statistics was not significant at the 0.05 critical alpha levels. There was no statistically significant difference between treatment with PEPPS alone and treatment with PEPPS augmented treatments.
Table 1. Treatment response to PEPPS† stratified by GI symptoms, treatment regimens and age of horse.

<table>
<thead>
<tr>
<th>Treatment regimen</th>
<th>&lt; 6 months (%)</th>
<th>6-12 months (%)</th>
<th>1-2 years (%)</th>
<th>2-8 years (%)</th>
<th>&gt; 8 years (%)</th>
<th>Symptom/disorder treated</th>
<th>Treatment related totals (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>H2B+AA+Bismuth+PEPPS</td>
<td>2/2 (100)</td>
<td>2/2 (100)</td>
<td>2/2 (100)</td>
<td>2/2 (100)</td>
<td>2/3 (67)</td>
<td>Colic-diarrhea</td>
<td>10/11 (91)</td>
</tr>
<tr>
<td>H2B+AA+PEPPS</td>
<td>2/2 (100)</td>
<td>11/13 (85)</td>
<td>9/11 (82)</td>
<td>25/27 (93)</td>
<td>17/19 (89)</td>
<td>Ulcer colic</td>
<td>64/72 (88.9)</td>
</tr>
<tr>
<td>PPI+AA+PEPPS</td>
<td>3/4 (75)</td>
<td>6/7 (86)</td>
<td>6/7 (86)</td>
<td>10/13 (77)</td>
<td>5/6 (83)</td>
<td>Ulcer colic</td>
<td>30/37 (81.0)</td>
</tr>
<tr>
<td>PPI alone+PEPPS</td>
<td>4/4 (100)</td>
<td>10/13 (77)</td>
<td>6/8 (75)</td>
<td>18/21 (86)</td>
<td>6/7 (86)</td>
<td>Ulcer colic</td>
<td>44/53 (83.0)</td>
</tr>
<tr>
<td>PEPPS† alone</td>
<td>7/8 (88)</td>
<td>3/3 (100)</td>
<td>4/5 (80)</td>
<td>10/13 (77)</td>
<td>5/7 (71)</td>
<td>Ulcer colic</td>
<td>29/36 (80.6)</td>
</tr>
<tr>
<td>Age Related Response</td>
<td>18/20 (90)</td>
<td>32/38 (84)</td>
<td>27/33 (82)</td>
<td>68/76 (90)</td>
<td>35/42 (83)</td>
<td>Ulcer colic</td>
<td>180/209 (86)</td>
</tr>
</tbody>
</table>

PEPPS, potency-enhanced polyanionic phyto-saccharide; PPI, proton pump inhibitor (omeprazole); AA, antacids; H2B, histamine-2 blocker (ranitidine or cimetidine); bismuth, bismuth preparations.

treatments. No additional clinical response was obtained over PEPPS alone by the continuance of pre-existing therapies (omeprazole, ranitidine, antacids or bismuth).

**Endoscopy positive ulcers in horses**

Of the 198 horses with ulcer-associated colic, 23 horses were endoscoped by 8 veterinarians prior to and between 14 to 20 days, following PEPPS treatment. Eight of these horses were on PEPPS alone, while the other 15 were on PEPPS augmented therapies. Table 4 shows that while horses in each treatment group had ulcers, all ulcerations were confirmed healed by second endoscopy performed between 14 to 20 days on treatment. With FDA, approved omeprazole require 28 days for a statistically significant healing effect, this observation of PEPPS healing between 14 to 20 days represented accelerated healing.

**DISCUSSION**

**General impression**

Traditionally, the treatment of ulcer colic in horses generally centers on acid reduction (Andrews, 2005; Hammond et al., 1996) for which there are several therapeutic options. Clinical practice to treat equine diarrhea involves the use of psyllium hydrocolloids and bismuth/salicylate combination (Galvin et al., 2004). The empiric use of anti-spasmodics, analgesics and anxiolytics for colic is reasonable as well. However, simultaneous management of ulceration, diarrhea and colic by a single therapeutic agent would be preferred. Potency-enhanced polyanionic phyto-saccharide, PEPPS was prescribed to 209 horses in the private practices of 32 equine practitioners in the US. With the exception of a few cases, the equine practitioners introduced PEPPS into active treatment regimens, deemed to be failing. The study was conducted over a 3.5 year period. The data showed a positive association between the use of PEPPS and the rapid resolution of ulcers, ulcer-associated colic and diarrhea in horse whose symptoms had failed to respond to pre-existing therapies. The hypothesis that the administration of PEPPS is associated with the rapid resolution of ulcer colic, diarrhea and diarrhea associated colic in most horses was supported by the data reported here. In majority of horses on failing therapies, those symptoms resolved within 4 days (or in 7 doses) of receiving PEPPS, a timeframe significantly less than that anticipated from the private practice experiences of veterinarians participating in the trial. Additionally, all horses with ulcerations diagnosed by gastroscopy healed within 14 to 20 days using either PEPPS alone or PEPPS augmented by continuance of existing treatments. In other words, the use of PEPPS in horses with known gastric ulcers was associated with the resolution of ulcer within 14 to 20 days, which is sooner than 28 day therapy required for omeprazole (Merial, 1999) to achieve statistically significant healing effect. A more fair comparison of PEPPS to omeprazole requires evaluation of both under similar clinical restrictions. Be that as it may, 14 to 20 days for ulcer healing is far less than 28 days known to be required for omeprazole.

**Limitations of this observational study**

Observational study of this nature does have distinct limitations (Hannan, 2008). The absence of conventional control groups precludes any statements regarding efficacy. An observational study shows association without commentary to cause. Instead, what is known from this study is
Table 2. Percentage response to treatment in horses with CI (95%).

<table>
<thead>
<tr>
<th>Parameter (Treatment regimen)</th>
<th>Horses with ulcer colic</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Horses with CI (95%)</td>
<td></td>
</tr>
<tr>
<td>PEPPS Alone</td>
<td>81% (n=36)</td>
<td>12.9</td>
</tr>
<tr>
<td>PPI + PEPPS</td>
<td>83% (n=53)</td>
<td>10.11</td>
</tr>
<tr>
<td>PPI + AA + PEPPS</td>
<td>81% (n=37)</td>
<td>12.64</td>
</tr>
<tr>
<td>H2B + AA + PEPPS</td>
<td>88% (n=72)</td>
<td>7.26</td>
</tr>
<tr>
<td>Ulcer colic regimens 2,3,4 collectively</td>
<td>85% (n=162)</td>
<td>5.47</td>
</tr>
</tbody>
</table>

**Horses with diarrhea and colic**

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>H2B + AA + Bismuth + PEPPS</td>
<td>91% (n=11)</td>
</tr>
</tbody>
</table>

PEPPS, potency-enhanced polyanionic phyto-saccharide; PPI, proton pump inhibitor (omeprazole); AA, antacids; H2B, histamine-2 blocker (ranitidine or cimetidine); bismuth, bismuth preparations.

Table 3. T-statistic value comparing PEPPS alone to PEPPS-enhanced treatments in horses.

<table>
<thead>
<tr>
<th>Treatment with PEPPS alone compared to</th>
<th>t-Statistic</th>
<th>Degrees of freedom</th>
<th>2-tailed p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>PPI + PEPPS</td>
<td>0.289</td>
<td>87</td>
<td>0.773</td>
</tr>
<tr>
<td>PPI + AA + PEPPS</td>
<td>0.244</td>
<td>88</td>
<td>0.808</td>
</tr>
<tr>
<td>H2B + AA + PEPPS</td>
<td>1.177</td>
<td>106</td>
<td>0.242</td>
</tr>
<tr>
<td>H2B + AA + Bismuth + PEPPS</td>
<td>0.804</td>
<td>45</td>
<td>0.426</td>
</tr>
</tbody>
</table>

PEPPS, potency-enhanced polyanionic phyto-saccharide; PPI, proton pump inhibitor (omeprazole); AA, antacids; H2B, histamine-2 blocker (ranitidine or cimetidine); bismuth, bismuth preparations.

Table 4. Horses prior to treatment and day 14 to 20.

<table>
<thead>
<tr>
<th>Treatment regimen</th>
<th>Number (%) with gastric ulcers</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Prior to treatment</td>
</tr>
<tr>
<td>PEPPS alone treatment</td>
<td>8/8 (100)</td>
</tr>
<tr>
<td>PEPPS-augmented treatments</td>
<td>15/15 (100)</td>
</tr>
</tbody>
</table>

PEPPS, potency-enhanced polyanionic phyto-saccharide; Treatments augmented with PPI, proton pump inhibitor (omeprazole); AA, antacids; H2B, histamine-2 blocker (ranitidine or cimetidine); bismuth, bismuth preparations.

that a large majority of the horses were observed to improve better than 80% of the study’s equine veterinarians would have thought possible, based on their collective past clinical experience. The historical experience of each veterinarian and their consensus of what constitute a significant deviation from that experience are subjective points. As a result, the data offers little predictive value and is mute as to efficacy. The study design provides an affirmative proof-of-principle supporting a plausible utility of PEPPS in the management of ulcers, diarrhea and colic associated with ulcers and diarrhea.

Another disadvantage of this study is that the manner of recruitment gives rise to bias. Practitioners were self-selected by virtue of responding to advertisements regarding a new gastrointestinal protectant which is resold at profit if the product is prescribed to a patient. Data obtained utilizing this method of recruitment is vulnerable to a self-selection bias that is profit driven. In general, an appropriately randomized, placebo-controlled blinded investigation would best quantify the efficacy of PEPPS and thereby provide a better basis on which to predict the benefit of PEPPS in managing dysfunctional GI symptoms.

**Strengths of this observational study**

Despite limitations on study design, an observational study such as this can provide “real world” setting information useful in evaluating the comparative effectiveness of PEPPS (Dreyer et al., 2010). From this trial, there appear to be positive benefits associated with the use of PEPPS in managing ulcers, diarrhea and colic in horses due to ulcers and diarrhea.
Firstly, the geographic diversity of state-licensed veterinarian was a significant factor. Eighty percent of the equine regions in the US are represented in this study. The data reflected a national experience involving equine practitioners licensed in 20 of the top 25 states, with the highest horse population and ownership. The high percent response associated with PEPPS was uniform. This implies that positive association of PEPPS with symptom resolution was universal with no significant variability due to difference in region or physician practice.

Secondly, the association of a positive outcome with PEPPS demonstrated no deference to the age of the horse. All ages of horses responded similarly. The horse’s age did not alter response to PEPPS. Clinical response was high across all age groups involved, with foals, yearlings and older horses responding equally well to PEPPS.

Thirdly, though horses could have improved in the same timeframe on their existing treatments, it was not likely. The continuation of pre-existing ulcer treatments, to which PEPPS was added, had no added advantage of either treatment over each other. Positive outcomes for ulcer colic in horses on PEPPS alone (81%, 29/36) was indistinguishable from the outcomes in horses treated with omeprazole, antacid and PEPPS. In this study, there did not appear to be any advantage over using PEPPS alone, as 30/37 (81%) horses had virtually the same outcome using PEPPS alone versus PEPPS with the other treatments. Prior to use of PEPPS, the other treatment regimens were failing. The time horses were on the failing treatments was not assessed, however given that the study was conducted within a practice setting, it is assumed initial treatments fail a reasonable test of the horses. Additionally, the extended study period of 3.5 years averaged in the effects on positive outcomes, if any, attributable to seasonality.

Use of PEPPS associated with accelerated healing of ulcers without acid inhibition

Complete healing occurred within 14 to 20 days using either PEPPS alone (n = 8) or PEPPS in addition of clinically ineffective treatments (n = 15) in 23 horses with endoscopic evidence of gastric ulcers. This is 8 to 14 days sooner than the 28 days required for omeprazole to achieve statistically significant healing (Merial, 1999). Healing independent of acid treatment was reported in the Merial study (1999), wherein nearly 40% of horses on placebo either improved or were completely healed, giving support to the notion of an acid-independent ulcer-deterrent system within the mucosa. While 15 of 23 horses with ulcers were on acid reduction therapies with PEPPS, 8 of 23 were on PEPPS alone healed sooner that would otherwise be expected. Acid-mitigating therapies have no known direct interaction with mucosal mechanisms responsible for healing ulcers, but mucoadherent PEPPS does. Accelerated healing in 14 to 20 days was a positive outcome associated with the use of PEPPS alone in horses with ulcers.

Plausible mechanism of action for PEPPS

Slippery elm mucilage is a polyanionic phytosaccharide similar to sucralfate, in that it is a mult-saccharide chain that contains branched substituents with a high electro-negative density. In slippery elm, electro-negative galacturonic acid corresponds to the electro-negative sulfates of sucrose dissacharide of sucralfate. When the elm polysaccharide is placed in a solution of cations and multi-dentate chelators, it is assumed that, similar to sucralfate, the mucosal protective characteristics reportedly existing with PEPPS are indifferent to gastric acidity (Steiner et al., 1982), and its strength of binding, unaffected by pH (Danesh et al., 1988).
Additionally, by virtue of its electro-density, PEPPS binds enteric growth factors as does sucralfate (Jones et al., 2004; Konturek et al., 1991, 1995) and thereby facilitates immediate mucosal regeneration (Tarnawski et al., 1986). Like sucralfate, it is supposed that the electronegative density of PEPPS enables it to modulate voltage-gated nociceptors of the enteric mucosa (Slomiany et al., 1992), thereby repolarizing neural fibers of the vagus nerve reversing colic and pain sensation from acid (Holtzer, 2001, 2004), nausea and colic (Beyak and Grundy, 2005). Just as high doses of sucralfate (25 to 100 mg/kg/dose) by some mechanism stop diarrhea in small animals in 2 to 14 doses (Steiner, 1990), so does PEPPS in the same manner relieve diarrhea in the horse. While the exact mechanism of action of PEPPS, and for that matter, of sucralfate, is not entirely clear, these comments serve as a start for discussion and future analysis. In all, the mechanism of action utilized by PEPPS allows the management of symptoms and signs via direct engagement of enteric elements responsible for mucosal integrity and normative function of the GI tract.

Conclusion

The majority of horses treated with PEPPS for colic due to ulcer and diarrhea were observed to have shortened clinical course to an extent not anticipated by experienced equine practitioners. This data was generated by veterinarians practicing in 20 out 25 most populous equine owning states. While all equine patients eventually responded to PEPPS, a majority of horses responded within 4 days or 7 doses, returning to baseline feeding habits, demeanor and bowel habits. Additionally, whenever PEPPS was used, gastrocape-positive ulcerations healed within 14 to 20 days. This was true in eight horses that healed using PEPPS alone without acid mitigating therapies. The data from this study supports the notion that PEPPS may be useful in the management of equine ulcer, colic and diarrhea. Yet blinded, randomized placebo-controlled trials are needed to assess the true efficacy of potency-enhanced phyto-saccharides.

Acknowledgement

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References


Prevalence of bovine coccidia in Kombolcha district of South Wollo, Ethiopia

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A cross sectional study was conducted from November 2011 up to April 2012 in Kombolcha town to determine the prevalence of coccidia infection in calves. Fecal samples were collected from a total of 288 calves with the age of 1 month to 1 year old which were included in the study purposively. After collection, the samples were transported to the laboratory and examined for the presence of Eimeria oocyst by flotation techniques. For positive sample, a solution of 2.5% potassium dichromate (K₂Cr₂O₇) was added to the feces containing the oocyst for sporulation and identification of the species. Out of 288 calves, 92 (31.9%) were found to be positive for Eimeria species. There was statistically significant difference (P< 0.05) in the prevalence of coccidia infection to different age of animals or fecal consistency. However, the difference was not statistically significant (P>0.05) between coccidia infection and sex, breed, body condition, address or management system. Five species of Eimeria were identified in the study and the most prevalent species were Eimeria bovis (42.3%), Eimeria zuernii (28.3%) and Eimeria auburnensis (13.0%). The other species were E. ellipsoidalis (8.7%), E. alabamensis (4.3%) and unidentified oocyst (3.3%). In conclusion, the present finding has demonstrated that bovine coccidia are one of the important pathogens in calves in the study area. Further epidemiological investigations are required to determine the Eimeria species composition and different agro ecological risk factor on the occurrence of the disease.

Key words: Calves, coccidiosis, Eimeria, Kombolcha, prevalence, risk factors.

INTRODUCTION

Coccidiosis is a parasitic disease of the intestinal tract caused by microscopic organisms called coccidia and is one of the most common and important disease of cattle worldwide. Bovine coccidiosis has been observed in almost all areas where cattle are raised and is usually the most common and important in calves younger than 1 year. All calves managed under conventional systems are exposed to coccidia and become infected early in life. Many studies indicated that under natural conditions, mixed species infections are much more common than mono species infection (Ernst et al., 1987). Coccidiosis in cattle is particularly a problem of confined animals kept under intensive husbandry practices. The disease is more common in housed animals than in those on pastures. In associations with other enteropathogens, coccidia has been indicated as an important cause of diarrhea in calves (Radostits et al., 1994).Coccidiosis spreads from one animal to another by contact with infected feces and is one of the most alarming problems for calf rearing industry. The most common clinical manifestations include in appetite, weakness, and loss of weight, diarrhea, depression and anemia (Soulsby, 1982). The development of clinical coccidiosis in cattle mainly depends on factors like species of Eimeria, age of infected animal, number of oocysts ingested, presence of concurrent infections and type of production system and management practices (Daugschies and Najdrowsk,
More than 13 species of *Eimeria* and one species of *Isospora* have been described to infect cattle and causes of coccidiosis. *Eimeria bovis* and *Eimeria zuernii* are considered the most pathogenic species as they are usually associated with clinical coccidiosis under field conditions. Thus, determination of prevalence, species composition, associated risk factors and animal management and husbandry practice is very useful in designing efficient control strategies (Ernst et al., 1984). *E. zuernii*, *E. bovis* and *Eimeria auburnensis* are the species most often associated with clinical disease in cattle, and other species have been shown to be mildly or moderately pathogenic. Coccidiosis is commonly a disease of young cattle (1 to 2 month to 1 year) and usually is sporadic during the wet seasons of the year (Fraser, 2006).

The prevalence, species composition, and importance of bovine coccidiosis have been documented in various countries of the world. Ernst et al. (1987) reported 82.28% infection rate in the coastal plain area of Georgia (USA); Rodriguez-Vivas et al. (1996) reported 87.8% infection rate in a sub humid tropical climate; Pandit (2009) reported 73.2% infection rate in Kashmir valley. In Ethiopia, Abebe et al. (2008) reported an overall prevalence of 68.1% in cattle in Addis Ababa and Debre Zeit area. However, there is lack of information on the occurrence and losses associated with bovine coccidiosis and very little attention has been given to the role of coccidiosis as the cause of disease and production losses in cattle in Ethiopia, especially in Kombolcha district of South Wollo. Therefore, taking into account the significance of the parasite as one of the most important causes of economic losses and the scarcity of information in the country, the present study was designed to determine the prevalence, species composition, and associated risk factors with *Eimeria* infections of calves in Kombolcha district.

**MATERIALS AND METHODS**

**Study area**

The study was conducted in Kombolcha town, which is found to the North East of Ethiopia in Amhara regional state from November, 2011 to April, 2012. The town is located in a range of altitudes between 1,500 and 1,840 m above sea level with average rainfall of 750 to 900 ml during the study period. Its annual temperature ranges from 25 to 30°C and the relative humidity of the region varies from 23.9 to 79% (NMSA, 2010).

**Study animals**

The study animals were calves within the age of 1 month to 1 year old. A total of 288 fecal samples were collected and examined for coccidia species from different dairy farms and small holders found in Kombolcha. Examined animals were categorized into two age groups as group I = 1 to 6 months age and group II = >6 to 12 months age which was determined by asking the owner of the animal orally (Abebe et al., 2008). Examined animals were also categorized into three according to their body condition: good, medium and poor. This is based on different body visible bone structure and fat deposit (Nicolson and Butterworth, 1986).

**Sampling techniques and sample size determination**

Purposive sampling technique was used to select the study animals from the target population. The sample size required for the study was determined according to Thrushfield (2005) as follows. By taking a 95% confidence interval, 20% expected prevalence whenever there is no information on the prevalence of the disease in the area and 5% desired absolute precision, the sample size was calculated as follows:

\[
\text{n} = \frac{1.96^2 \times P_{\text{exp}} (1 - P_{\text{exp}})}{d^2}
\]

where

- \( n \) = required sample size; \( P_{\text{exp}} \) = expected prevalence, \( d \) = desired absolute precision.

Therefore, 245 calves within the age of 1 month to 1 year old were required from target population in the study area. But, the sample size was maximized to 288 calves.

**Experimental**

**Fecal sample collection**

About 30 g fresh fecal sample was collected from rectum from each calf using sterile disposable plastic gloves. The samples were placed in a labeled clean plastic container (universal bottle) and were transported to the parasitological laboratory of Kombolcha Animal Health and Diagnostic Center on the same day of collection and were preserved at refrigerator until processing within 48 h of arrival. At the time of sampling, the name of the farm (owner), date of sampling, consistency of the feces (soft, watery or normal) and the age, sex, breed, address and management system were recorded for each calf on a data recording format.

**Parasitological investigation**

A 5 g portion of each of the 288 fecal samples collected from the total of 30 g was weighed out using a balance and put in a 50 ml beaker. 42 ml of water was added, mixed thoroughly and poured into a 100-ml glass beaker through a strainer. The 50-ml glass beaker was rinsed with 8 ml of water and the total fluid was poured into four 15-ml conical tip centrifuge tubes. After centrifugation at 1,500 rpm for 5 min, the supernatant was decanted and a sugar solution (specific gravity 1.25) was added to the sediment, until the tube was about half full. The content of each test tube was thoroughly mixed with a wooden applicator stick. With the aid of a conical flux, more sugar solution was added until a convex meniscus was formed on top of the tube. A glass cover slip was placed on top of each tube and was left for 30 min. Then, each glass cover slip was briskly lifted up and placed on a clean glass slide, not allowing formation of air bubbles. The entire area under each cover slip was examined under a binocular microscope at 400× magnification (Hendrix, 1998).

**Sporulation of Eimeria oocysts**

A solution of 2.5% potassium dichromate was added to each fecal sample, which contained most of the *Eimeria* oocyst in a beaker, mixed thoroughly with a wooden applicator and poured into a Petri dish. Each Petri dish was left on the bench in the laboratory to allow
Thereafter, every 24 h, the culture of oocysts was mixed thoroughly and with the aid of medicine dropper, a drop of the culture was placed on a glass slide, covered with a glass cover slip and examined under the microscope to determine when sporulation occurred. When sporulation of oocysts was completed after 14 days, the Petri dish containing oocysts was covered up and stored in a refrigerator at 5°C until needed. Identification of coccidian species will be based on the morphological features of the sporulated oocysts (size, shape, color, and texture of oocyst wall, presence or absence of microple, polar cap) and time of sporulation with the aid of taxonomic keys (Soulsby, 1982; Kennedy and Kralkara, 1987; Sommer, 1998).

**Data management and analysis**

Data collected from study sites were entered and stored in a Microsoft excel spread sheet program and coded for analysis. Statistical analysis was done on Statistical Package for Social sciences (SPSS) 17.0 statistical software. The prevalence was calculated for all data as the number of infected individuals divided by the number of sampled individual and multiplied by 100. Categorical data were analyzed first with the chi square ($\chi^2$) test for independence as a screening process. A P-value < 0.05 was considered as statistically significant.

**RESULTS**

**Prevalence and analysis of potential risk factors for the occurrence of bovine coccidia**

Out of all 288 samples, a total of 92 samples (31.9%) tested positive for *Eimeria* species oocysts as shown in Table 1.

Analysis of the potential risk factor for the occurrence of coccidia has revealed that there were a significant association (P<0.05) with age and fecal consistency of the calves. However, there is no significant association (P>0.05) with breed, sex, body condition, address management system of the calves and coccidia (Table 1).

**Percentage and analysis of *Eimeria* spp. with the suspected risk factors**

Out of 288 calves whose fecal samples were examined, 92 (31.9%) were found to be positive for 5 species of *Eimeria*. The most prevalent species were *E. bovis*, which occurred in 39 (42.3%), *E. zuernii* in 26 (28.3%), *E. auburnensis* in 12 (13.0%), *Eimeria ellipsoidalis* in 8 (8.7%), *Eimeria alabamensis* in 4 (4.3%) and unidentified oocyst were observed in 3 (3.3%) (Table 2).

Analysis of the potential risk factors and species of *Eimeria* has revealed that there is a significant association (P<0.05) between the occurrence of *E. bovis* with body condition and fecal consistency. In addition, fecal consistency has a significant association (P<0.05) for the occurrence of all *Eimeria* spp. However, there is no significant difference (P>0.05) between sex, breed, age, and address and management system for the occurrence of *Eimeria* spp.

**DISCUSSION**

The present study has revealed that the presence of bovine coccidia species parasitizing the gastro intestinal tract of calves under the age of one years in Kombolcha district of South Wollo. The overall prevalence of *Eimeria* spp. is 31.9%, which is lower than previous findings reported in Addis Ababa and Debre Zeit by Abebe et al. (2008) (68.1%), in the coastal plain area of Georgia (USA) by Ernst et al. (1987) (82.28%) and in sub-humid tropical climate by Rodriguez-Vivas et al. (1996) (87.8%). This variation is most likely attributed to the differences in agro-ecology, and husbandry practices of the study animals in different countries (Radostits et al., 2006).

Analysis of risk factor in the association of disease occurrence has revealed that there was no statistically significant association (P>0.05) between breed and coccidia infection. These indicate that body condition does not have influence on the occurrence of coccidia infection. This is due to either equal chance of accessing the oocysts or no difference on protective immunity for the disease. This finding agrees with the report of Abebe et al. (2008). There was no statistically significant association (P<0.05) between sex and coccidia infection. The prevalence in female calves was similar to that of males in this study. This finding agrees with the report of Abebe et al. (2008). There was no statistical significant association (P>0.05) between the address of the animals and coccidian infection. However, the present study disagrees with previous studies indicating that there was a statistical significant association between geographic zone and the occurrence of coccidian infection (Abebe et al., 2008).

There was a strongly significant association (P<0.05) between the age of the calves with the risk of infection in which the prevalence of coccidia appeared to follow an age pattern. Higher infection rate was observed in calves >6 to 12 months of age than calves of 1 to 6 months of age due to the fact that there was good nursing of the colostrum feeding for younger calves. During investigation, almost all the calves older than 6 months were housed in overcrowded condition, less care were given and have easy contact with adult animals. This has given more chance for the animals to lick each other and ingest large number of oocysts, which is in agreement with previous reports (Kennedy, 2001; Abebe et al., 2008; Rodriguez-Vivas et al., 1996; Radostits et al., 2006). Coccidiosis occurs most commonly in young animals with a seasonal incidence when young calves are brought together for weaning or moved into feedlots or fed in small areas for the winter months. The prevalence of infection and the incidence of clinical disease are also age related (Radostits et al., 2006).

There was no a statistical significant association between body condition of the animals and coccidian...
Table 1. Prevalence and analysis of potential risk factors for the occurrence of bovine coccidian.

<table>
<thead>
<tr>
<th>Risk factor</th>
<th>No. of calf examined</th>
<th>No. of positive</th>
<th>Prevalence (%)</th>
<th>$\chi^2$</th>
<th>P- value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breed</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Local</td>
<td>162</td>
<td>53</td>
<td>32.7</td>
<td>0.105</td>
<td>0.750</td>
</tr>
<tr>
<td>Cross</td>
<td>126</td>
<td>39</td>
<td>31.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>103</td>
<td>31</td>
<td>30.1</td>
<td>0.252</td>
<td>0.616</td>
</tr>
<tr>
<td>Female</td>
<td>185</td>
<td>61</td>
<td>33.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Address</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Urban</td>
<td>137</td>
<td>44</td>
<td>32.1</td>
<td>0.004</td>
<td>0.952</td>
</tr>
<tr>
<td>Rural</td>
<td>151</td>
<td>48</td>
<td>31.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1-6 month</td>
<td>112</td>
<td>27</td>
<td>24</td>
<td>5.178</td>
<td>0.023</td>
</tr>
<tr>
<td>&gt;6-12month</td>
<td>176</td>
<td>65</td>
<td>36.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Body condition</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Good</td>
<td>114</td>
<td>34</td>
<td>29.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Medium</td>
<td>106</td>
<td>37</td>
<td>34.9</td>
<td>0.698</td>
<td>0.705</td>
</tr>
<tr>
<td>Poor</td>
<td>68</td>
<td>21</td>
<td>30.9</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Faecal consistency</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normal</td>
<td>176</td>
<td>18</td>
<td>10.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Soft</td>
<td>62</td>
<td>36</td>
<td>58.8</td>
<td>102.278</td>
<td>0.00</td>
</tr>
<tr>
<td>Diarrheic</td>
<td>50</td>
<td>38</td>
<td>76.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Management</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Intensive</td>
<td>109</td>
<td>38</td>
<td>34.9</td>
<td>0.687</td>
<td>0.407</td>
</tr>
<tr>
<td>Extensive</td>
<td>179</td>
<td>54</td>
<td>30.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>288</td>
<td>92</td>
<td>31.9%</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 2. Percentage of *Eimeria* spp. distribution in Kombolcha district.

<table>
<thead>
<tr>
<th>Species of <em>Eimeria</em></th>
<th>Number of positive sample</th>
<th>Prevalence (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>E. bovis</em></td>
<td>39</td>
<td>42.4</td>
</tr>
<tr>
<td><em>E. zuernii</em></td>
<td>26</td>
<td>28.3</td>
</tr>
<tr>
<td><em>E. auburnensis</em></td>
<td>12</td>
<td>13.0</td>
</tr>
<tr>
<td><em>E. ellipsoidalis</em></td>
<td>8</td>
<td>8.7</td>
</tr>
<tr>
<td><em>E. alabamensis</em></td>
<td>4</td>
<td>4.3</td>
</tr>
<tr>
<td>Unidentified oocyst</td>
<td>3</td>
<td>3.3</td>
</tr>
<tr>
<td>Total</td>
<td>92</td>
<td>100</td>
</tr>
</tbody>
</table>

infection (P>0.05). These indicate that body condition does not have influence on the occurrence of coccidia infection. This is due to either the level of infection, sampled size or most of the affected animals harbor the disease without showing clinical signs (Fraser, 2006). However, this finding disagrees with the report of Abebe et al. (2008). In the present study, 76.0% (38/50) of diarrheic calves (blood stained, watery and fetid diarrhea) were found to be positive for *Eimeria*. However, there
were no apparent clinical signs in most of the animals sampled for the study. The influence of management system on prevalence of coccidia has revealed that there was no statistically significant association between them (P>0.05). This finding disagrees with the previous reports by Abisola (2004) and Kennedy and Kralka (1987). This might be attributed to the fact that hygienic system of the barn, nutritional status, contamination of the feed or overcrowding of the animal was similar in both management systems.

The overall prevalence of *Eimeria* spp. in this study was 31.9% (Table 2) and the most prevalent among the 5 species encountered were *E. bovis* (42.4%), *E. zuernii* (28.3%) and *E. auburnensis* (13.0%). These species are the most frequently reported *Eimeria* spp. in outbreaks of coccidiosis throughout the world (Andrews et al., 2004; Abebe et al., 2008; Ernst et al., 1987; Kasim and Al-Shawa, 1985; Oda and Nishida, 1990). This high prevalence of pathogenic species (*E. bovis* and *E. zuernii*) in infected calves and the greater proportions of subclinical infections could negatively influence animals’ productivity and cause economic losses from poor feed efficiency, slow weight gain, weight loss, failure to grow to their full potential, and increased susceptibility to other diseases (Fraser, 2006). Moreover, continuous oocysts shed from subclinical infected calves contaminate the environment or the hair coats and cause severe coccidiosis in highly susceptible new calves that are kept in these areas (Abebe et al., 2008; Radostits et al., 2006).

**Conclusion**

This study has revealed that the prevalence of calves *Eimeria* infection in Kombolcha district was 31.9% and five *Eimeria* spp. namely *E. bovis*, *E. zuernii*, *E. auburnensis*, *E. ellipsoidalis* and *E. alabamensis* were identified in all *Eimeria* positive fecal samples. The high prevalence of *Eimeria* spp. was considered as one of the important infection in cattle farms in the study area. The prevalence of coccidiosis has no significant association with address, sex, breed, body condition, management system of animals examined during the study period. However, the disease has a significant association (P<0.05) with age and faecal consistency of the calf. Results from this study indicate the *Eimeria* infection has a great significance for the livestock producer and need a serious control and preventive issue. Therefore, further epidemiological investigation on coccidia species should be needed in the study area.

**ACKNOWLEDGEMENT**

We would like to forward our appreciation for staff members of Kombolcha Regional Veterinary Laboratory for their cooperation to accomplish this work.

**REFERENCES**


Full Length Research Paper

Gross pathological changes in the reproductive tracts of cows slaughtered at two abattoirs in Southern Ethiopia

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Abattoir based study was conducted to assess the type and prevalence of reproductive abnormalities and pregnancy status of cows slaughtered at Hawassa municipality abattoir and Tula slaughter house. Out of the 345 genital tracts examined, one or two gross abnormalities with different degrees of severity were observed in 124 (35.9%) of genital tract. The most common abnormalities encountered were ovariobursal adhesions (6.38%, n=22), endometritis (4.93%, n=17), and follicular cysts (4.35%, n=15). On anatomical basis, ovarian abnormalities (14.78%, n=51) were more frequent followed by uterine (10.43%, n=36), cervico-vaginal (7.25%, n=25) and oviductal (4.93%, n=17) abnormalities. Both breed and study area showed no statistically significant effect in the prevalence of reproductive abnormalities. Pregnancy was recorded in 26.67% (n=92) of the slaughtered cows, of which 45.65% (n=42) were in the first trimester, while 30.43% (n=28) and 23.91% (n=22) in the second and third trimesters, respectively. The current study revealed that reproductive tract abnormalities are important diseases in the study areas with considerable impact on the reproductive performance of cows. Moreover, the large number of cyclical (36.52%) and pregnant cows (26.67%) slaughtered without any gross abnormalities indicates the absence of proper gynecological examination prior to slaughtering.

Key words: Reproductive abnormalities, genital tract, slaughter, cows, Hawassa, Tula.

INTRODUCTION

The traditional milk production system dominated by indigenous breeds of low genetic potential for milk production accounts for 97 to 98% of the total annual milk production in Ethiopia (Felleke, 2003). This system is constantly challenged by shortage of feed, lack of capital with dairy producers, unimproved animal husbandry system; inefficient and inadequate milk processing skill and prevailing diseases including uterine diseases and anestrus.

Diseases and abnormalities of the female genital tract are believed to be the major reason for economic loss associated with infertility, culling and slaughtering of cows (Singleton and Dobson, 1995; Ashenafi, 2004; Abalti et al., 2006). Since most reproductive tract problems lack additional outward manifestation, hence, examination of gross and microscopic lesions of genital tract play a central role in the identification of these problems. Most of these abnormalities can only be diagnosed when the animal is subjected to postmortem examination (Buregelt, 1997). Though, in different regions of Ethiopia, studies have been conducted on reproductive abnormalities of cows based on abattoir material; in Addis Ababa (Gebrekidan et al., 2009), in Sululta (Simenew et al., 2011), in Tigray (Zerihun, 2001), in Jimma (Amare, 2002), in Nekemte (Samuel, 2002) in Asela (Endalew, 2001), in Bahir Dar (Abalti et al., 2006) and in Awassa (Ashenafi, 2004). However, more systematic work has to be done to assess the problem in depth. Hence, the present study aimed to identify the possible causes of

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slaughtering of female cattle with due emphasis to reproductive organs and to establish the reproductive status (ovarian cyclicity) of slaughtered mature female cattle.

MATERIALS AND METHODS

Study area

The study was conducted at Hawassa Municipal abattoir and Tula slaughter house. Hawassa is the capital city of Southern Nations, Nationalities and Peoples Regional State (SNNPRS). The town is located at 275 km South of Addis Ababa with area coverage of 182,804 hectares. The total human population of the town is about 180,500 (CSA, 2004). The abattoir and slaughter house are located in Tula sub city at 5 and 18 km away from the town, respectively. Active and routine meat inspection is carried out in the abattoir to provide a wholesome meat to the residents of the city. Animals for slaughter at the abattoir are brought mainly from Hawassa, Shashamane, Arsi-Negelle, Dilla and Wolyta Soddo towns. However, cattle slaughtered at Tula slaughter house are not thoroughly inspected and destined for local consumption.

Study animals and design

The study animals included 345 cows (252 local, 58 Holstein Friesian and 35 cross) purchased from the aforementioned markets and brought to the abattoir for slaughtering purpose. All cows slaughtered at the municipality abattoir (n=95) and Tula slaughter house (n=250) during the six month study period were included in the study.

Experimental

Any relevant data about the productivity history like parity, reason for culling, breed, origin, and age of the slaughtered cows were collected from the owner to correlate with pathological finding of the reproductive organs. Age of the slaughtered cows was estimated according to the description given by De-Lahunta and Habel (1986). The genital tracts of all slaughtered cows were collected from both sources, placed in separate plastic bags and transported to Veterinary Parasitology and Pathology Laboratory, School of Veterinary Medicine, Hawassa University, for routine post-mortem examinations. The entire reproductive tract were visually examined and thoroughly palpated. Each reproductive tract was opened along the longitudinal axis starting from the vagina down to the horns and was observed for any abnormalities in color, odor, consistency and morphology (Feyissa and Bekana, 2000). Similarly, both ovaries were incised and inspected for any gross lesions. Moreover, the number of corpora albicantia and status of the corpus luteum (CL) were recorded. In pregnant tracts, the stage of pregnancy was determined based on crown rump length according to Evans and Sack (1973) and Elsayed et al. (1978) and was classified as early (<3 months), mid (3 to 6 months) and late (>6 months) using the formula for estimation of developmental stages (days) \( X = 2.5(y+1); \) where \( y \) is the crown-rump length in cm and \( X \) is developmental stages in days. Methylene blue dye was infused in to the oviduct using disposable syringe through infundibulum to check for non-visible in patent state of oviduct as described by Herenda (1987) and Assey et al. (1998).

Data management and analysis

The results obtained from gross examination of the reproductive tracts of slaughtered cows were recorded on spreadsheet of Microsoft Excel and analyzed using STATA 9 statistical software. Simple descriptive statistics was used for the analysis of the data obtained. Categorical data were analyzed with the Chi-square \( (\chi^2) \) test for independence. In all the analysis P value < 0.05 was considered for significance.

RESULTS

Abnormalities of the reproductive tracts

The overall abattoir prevalence of one or more reproductive abnormalities in cows of the two study sites was 35.9% (124/345). Out of these, eight cows were having two problems at a time. The prevalence of the problem by study sites and breed is summarized in Table 1. During the study period, the most frequently observed abnormalities or disease of the reproductive tract were ovariobursal adhesion with an occurrence rate of 6.38% (n=22) followed by an ovarian cyst of 5.22% (n=18), endometritis of 4.93% (n=17), mucormeta of 3.77% (n=13), vaginitis of 3.19% (n=11) and other less frequently observed abnormalities. Moreover, ovariobursal adhesion, endometritis and follicular cyst were relatively more common in improved dairy cows than the local zebu (Table 2). Based on anatomical classification, abnormalities of the ovary (n=51, 14.78%) were found with highest occurrence rate followed by uterine (n=36, 10.43%), cervico-vaginal (n=25, 7.25%) and oviduct abnormalities 4.93% (n=345). Moreover, one case of macerated fetus was also observed (Table 3).

Pregnancy statuses

The total number of pregnant cows slaughtered during the study period in both study sites was 92. Of the total pregnancies, 45.65% (n=42) were found with early, 30.43% (n=28) were mid and 23.91% (n=22) were late stages. It was also found that the right ovaries had the highest physiological activities than the left ovaries and their percentage were 65.45 and 34.55% on the right and left, respectively.

Ovarian cyclicity

A total of 126 cows slaughtered during the study period were found cyclic. Based on the location, cyclicity was found to be 49.21, 45.24 and 5.56% on the right, left and both ovaries, respectively (Table 4).

DISCUSSION

The prevalence of reproductive tract abnormalities of cows recorded in the present study (35.94%) was in line with previous reports of Teklu (1999) (37.0%, Addis Ababa),
Table 1. Abattoir prevalence of reproductive tract abnormalities in cows (n=345).

<table>
<thead>
<tr>
<th>Risk factor</th>
<th>Observation</th>
<th>Abnormality number</th>
<th>Prevalence (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Study area</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hawassa</td>
<td>95</td>
<td>30</td>
<td>31.58</td>
</tr>
<tr>
<td>Tula</td>
<td>250</td>
<td>94</td>
<td>37.6</td>
</tr>
<tr>
<td>Breed</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Local</td>
<td>252</td>
<td>94</td>
<td>37.30</td>
</tr>
<tr>
<td>Holstein</td>
<td>58</td>
<td>19</td>
<td>32.76</td>
</tr>
<tr>
<td>Cross</td>
<td>35</td>
<td>11</td>
<td>31.45</td>
</tr>
</tbody>
</table>

Table 2. Prevalence of different reproductive abnormalities in different breeds.

<table>
<thead>
<tr>
<th>Abnormality</th>
<th>Average age</th>
<th>Overall prevalence</th>
<th>Breed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ovariobursal adhesion</td>
<td>7.2</td>
<td>6.38</td>
<td>Local 8.62, Holstein 8.30, Cross 5.57</td>
</tr>
<tr>
<td>Endometritis</td>
<td>6.7</td>
<td>4.93</td>
<td>Local 3.97, Holstein 5.17, Cross 4.85</td>
</tr>
<tr>
<td>Follicular cyst</td>
<td>6.8</td>
<td>4.35</td>
<td>Local 3.97, Holstein 5.17, Cross 4.85</td>
</tr>
<tr>
<td>Mucometra</td>
<td>7.8</td>
<td>3.77</td>
<td>Local 4.76, Holstein 1.72, Cross -</td>
</tr>
<tr>
<td>Vaginitis</td>
<td>6.8</td>
<td>3.19</td>
<td>Local 2.38, Holstein 3.45, Cross 2.86</td>
</tr>
<tr>
<td>Hydrosalpinx</td>
<td>8.1</td>
<td>2.32</td>
<td>Local 2.78, Holstein - , Cross -</td>
</tr>
<tr>
<td>Cervicitis</td>
<td>7.2</td>
<td>2.32</td>
<td>Local 1.98, Holstein 1.72, Cross -</td>
</tr>
<tr>
<td>Occluded oviduct</td>
<td>4.8</td>
<td>1.74</td>
<td>Local 1.98, Holstein 1.72, Cross -</td>
</tr>
<tr>
<td>Parovarian cyst</td>
<td>7.1</td>
<td>1.74</td>
<td>Local 1.98, Holstein 1.72, Cross -</td>
</tr>
<tr>
<td>Hydrometra</td>
<td>8.2</td>
<td>1.45</td>
<td>Local 1.59, Holstein - , Cross 2.86</td>
</tr>
<tr>
<td>Ovarian hypoplasia/atrophy</td>
<td>6.5</td>
<td>1.16</td>
<td>Local 1.59, Holstein - , Cross -</td>
</tr>
<tr>
<td>Luteal cyst</td>
<td>6.3</td>
<td>0.87</td>
<td>Local 0.79, Holstein - , Cross 2.86</td>
</tr>
<tr>
<td>Pyosalpinx</td>
<td>8.3</td>
<td>0.87</td>
<td>Local 1.19, Holstein - , Cross -</td>
</tr>
<tr>
<td>Tortuous cervical canal</td>
<td>9.5</td>
<td>0.58</td>
<td>Local 0.79, Holstein - , Cross -</td>
</tr>
<tr>
<td>Cervical ring hypoplasia</td>
<td>5</td>
<td>0.58</td>
<td>Local 0.4, Holstein - , Cross 2.86</td>
</tr>
<tr>
<td>Vaginitis and cervicitis</td>
<td>5</td>
<td>0.58</td>
<td>Local 0.79, Holstein - , Cross -</td>
</tr>
<tr>
<td>Hydropsis/atrophy of uterus</td>
<td>3</td>
<td>0.29</td>
<td>Local 0.4, Holstein - , Cross -</td>
</tr>
<tr>
<td>Macerated fetus</td>
<td>8</td>
<td>0.29</td>
<td>Local 0.4, Holstein - , Cross -</td>
</tr>
</tbody>
</table>


Ovariobursal adhesion (6.38%, n=22) was the most common reproductive abnormality encountered during the study followed by ovarian cyst (5.22%, n=18), endometritis (4.93%, n=17), mucometra (3.77%, n=13), vaginitis (3.19%, n=11). In line with this finding, Abalti et al. (2006) encountered ovariobursal adhesion, endometritis and cystic ovaries as the most common abnormalities with respective prevalence of 5.5, 3.9 and 3.5%. Although, the exact mechanism by which adhesions develop is unclear (Roberts, 1986a, b), extreme adhesions have probably resulted from pregnancy complications that include retained fetal membranes and endometritis (Lewis, 1997). Mild adhesions could result from non-infectious conditions such as physical trauma as a result of rough manipulation of the genital tract during pregnancy diagnosis (Bondurant, 1999). Uterine infections were common findings in cows that had given birth with possible complications especially dystocia and retained fetal membrane.

Examination of ovarian cyclicity revealed 36.52% cyclic ovaries, which was lower than the report made by
Table 3. Prevalence of anatomical reproductive tract abnormalities of cows (n=345).

<table>
<thead>
<tr>
<th>Abnormality</th>
<th>Occurrence rate (%)</th>
<th>Proportional frequency (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ovarian abnormalities</td>
<td>14.78 (n=51)</td>
<td></td>
</tr>
<tr>
<td>Ovariobursal adhesion</td>
<td>6.38</td>
<td></td>
</tr>
<tr>
<td>Ovarian cyst</td>
<td>5.22</td>
<td>38.66</td>
</tr>
<tr>
<td>Parovarian cyst</td>
<td>1.74</td>
<td></td>
</tr>
<tr>
<td>Ovarian hypoplasia/atrophy</td>
<td>11.45</td>
<td></td>
</tr>
<tr>
<td>Uterine abnormalities</td>
<td>10.43 (n=36)</td>
<td></td>
</tr>
<tr>
<td>Endometritis</td>
<td>4.93</td>
<td></td>
</tr>
<tr>
<td>Mucometra</td>
<td>3.77</td>
<td>27.27</td>
</tr>
<tr>
<td>Hydrometra</td>
<td>1.45</td>
<td></td>
</tr>
<tr>
<td>Hypoplasia/atrophy of uterus</td>
<td>0.29</td>
<td></td>
</tr>
<tr>
<td>Cervico- vaginal abnormalities</td>
<td>7.25 (n=25)</td>
<td></td>
</tr>
<tr>
<td>Vaginitis</td>
<td>3.19</td>
<td></td>
</tr>
<tr>
<td>Cervicitis</td>
<td>2.32</td>
<td>18.94</td>
</tr>
<tr>
<td>Cervicitis and vaginitis</td>
<td>0.58</td>
<td></td>
</tr>
<tr>
<td>Cervical ring hypoplasia</td>
<td>0.58</td>
<td></td>
</tr>
<tr>
<td>Tortuous cervical canal</td>
<td>0.58</td>
<td></td>
</tr>
<tr>
<td>Oviduct abnormalities</td>
<td>4.93 (n=17)</td>
<td></td>
</tr>
<tr>
<td>Hydrosalpinx</td>
<td>2.32</td>
<td>12.88</td>
</tr>
<tr>
<td>Occluded oviduct</td>
<td>1.74</td>
<td></td>
</tr>
<tr>
<td>Pyosalpinx</td>
<td>0.87</td>
<td></td>
</tr>
<tr>
<td>Miscellaneous</td>
<td>0.87 (n=3)</td>
<td></td>
</tr>
<tr>
<td>Ovariobursal adhesion and hydrosalpinx</td>
<td>0.29</td>
<td></td>
</tr>
<tr>
<td>Ovariobursal adhesion and ovarian hypoplasia</td>
<td>0.29</td>
<td>2.27</td>
</tr>
<tr>
<td>Macerated fetus</td>
<td>0.29</td>
<td></td>
</tr>
</tbody>
</table>

Table 4. Status of ovarian activities and pregnancy.

<table>
<thead>
<tr>
<th>Status</th>
<th>Right (%)</th>
<th>Left (%)</th>
<th>Bilateral (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cyclic (n=126)</td>
<td>49.21 (n=62)</td>
<td>45.24 (n=57)</td>
<td>5.5 (n=7)</td>
</tr>
<tr>
<td>Pregnancy (n=92)</td>
<td>65.45</td>
<td>34.55</td>
<td>-</td>
</tr>
</tbody>
</table>

Chaudhari and Paul-Bokko (2000) in Nigeria (55.49%), Samuel (2002) in Nekemte (65.55%), Amare (2002) in Jimma (62.2%) and Ashenafi (2004) in Hawassa (66.7%). This variation could be attributed to difference in breed, sample size and management. The presence of active corpus luteum in the right (49.21%), left (45.24%) and both ovaries (5.56%) was in agreement with the occurrence rate reported by Teklu (1999), who reported 55, 43, and 2% for the right, left and both ovaries, respectively. The result indicates that the right ovary is more physiologically active than the left ovary.

Pregnancy wastage (26.67%) noted in the present study was comparable with previous reports made by Amare (2002) in Jimma (24.5%), Samuel (2002) in Nekemte (31.4%) and Ashenafi (2004) in Hawassa (29.91%). However, higher value was reported in East Africa Zebu by Assey et al. (1998) in Tanzania (54%), Abdissa (2000) in Bahirdar (37.8%) and Zeriun (2001) in Raya Valley (36.84%). Such high pregnancy wastage in slaughtered cows could be attributed to inadequate ante-mortem inspection of animals particularly on pregnancy diagnosis. Moreover, cattle owners in the area usually insist on sale of their pregnant cows, because of anticipated pregnancy failure or because of a comparably improved body condition during pregnancy. Though, it is difficult to estimate the total impact of slaughter of pregnant cows;
Chaudhari and Paul-Bokko (2000) concluded that pregnancy wastage accounts for 20 to 25% fall in livestock production particularly in the developing countries, especially sub-Saharan Africa.

Conclusions

Based on the findings of this study and personal communication with cattle owner, low milk yield, infertility and economic conditions were the major factors for slaughter of cows. More than 60% of cows were slaughtered while they were potentially fertile. Slaughtering of potentially fertile cows, that is, pregnant and cyclic cows, indicates the absence of proper antemortem pregnancy diagnosis and lack of record keeping about the different events of reproductive cycle. Coupled with the prevailing reproductive abnormalities, slaughtering of fertile cows cause huge fetal wastage, reduced replacement stock, pose selection pressure and other detrimental effects on the dairy industry. To combat this problem, complete antemortem examination, awareness creation to the owners about the causes and effects of the problem, enforcing laws guiding slaughter of cows are safely recommended.

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Full Length Research Paper

Occurrences and financial significance of bovine cystic echinococcosis in Southern Wollo, Northeastern Ethiopia

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The study was carried out in Kombolcha ELFORA Industrial Abattoir to assess the current status and economic importance of bovine hydatidosis. Hydatid cyst count, characterization and economic loss assessment were conducted out of a total of 535 cattle slaughtered in Kombolcha ELFORA Industrial Abattoir. 93 (17.4%) animals were found harboring hydatid cysts. Thorough meat inspection in the abattoir revealed that 101 visceral organs were found harboring one or more hydatid cyst. The infection of the lung, liver, spleen, heart and kidney were found to be 50.5, 40.6, 1.98, 4.95 and 1.98%, respectively. From the total of 276 hydatid cysts counted, 135 (49.6%), 47 (17%), 20 (7.24%) and 72 (26%) were found to be small, medium, large and calcified cysts, respectively and 178 (67.9%), 12 (4.3%) and 72 (26%) were sterile, fertile and calcified, respectively. The rate of cyst calcification was higher in the liver than in the lung while fertility rate was higher among the cysts of the lung. The annual financial losses from organ condemnation and carcass weight loss due to bovine hydatidosis at Kombolcha ELFORA Industrial Abattoir were estimated to be 288,473.60 Ethiopian Birr (ETB) ($1 = 17.00 ETB). Thus, echinococcosis/hydatidosis is considerably a prevalent disease in cattle, with serious public health concern reflections and causes substantial visible and invisible losses in cattle in the study area.

Key words: Bovine hydatidosis, economic loss, Kombolcha ELFORA, meat inspection, prevalence.

INTRODUCTION

Among the many prevalent livestock diseases, parasitosis represent a major drawback to livestock development in the tropics in general and hydatidosis is among the major parasite diseases contributing to low productivity of meat production due to carcass or organ condemnation, in particular (Lemm et al., 1985). Infection, with the metacestode hydatid cyst of Echinococcus granulosus, stage of parasite tape worms is recognized as one of the world’s major zoonosis affecting both humans and domestic animals (Cringoli et al., 2007).

The definitive host of the parasite, E. granulosus, is dogs which harbor adult tape worms and excrete the parasite eggs along with their feces, while livestock and human are the main intermediate hosts (Oku et al., 2004) for whom the outcome of infection is the development of hydatid cysts in lung, liver or other organs (Muller, 2001; Budke et al., 2006). The incidence of human hydatidosis and the prevalence of the hydatidosis in domestic animals are the highest in countries where there is a large dog population and high sheep production (Gracey, 1986). The absence of proper meat inspection procedures and the presence of large stray dog population are thought to contribute significantly to the prevalence of the disease in Ethiopia (Kebede et al., 2009a). In Ethiopia, a number of researchers reported high prevalence of hydatidosis in different parts of the country. Fuller and Fuller (1981) documented a hyperendemic focus of hydatid disease in South-western Ethiopia in Dassanetch and Nyangatom people. In abattoirs of various locations, researchers indicated that hydatidosis is widespread in

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Ethiopia with great economic and public health significance (Jobre et al., 1996; Sissay et al., 2008; Kebede et al., 2009a, b, c, d, Kebede, 2010). However, there is no current information regarding the prevalence and economic significance of hydatidosis in livestock in South Wollo zone, northeastern part of the country. Hence, it would be essential to have information on the status of hydatidosis with regard to its magnitude of occurrence and economic significance of this disease in the region. Therefore, the present study was aimed at (1) determining the magnitude of the hydatidosis in cattle; (2) studying the localization and fertility/sterility rates of hydatid cysts; and (3) estimating the annual economic loss attributed to the condemned carcasses and organs and duet to weight loss in Kombolcha ELFORA Industrial Abattoir, Northeastern Ethiopia.

MATERIALS AND METHODS

Study area

This study was conducted at Kombolcha ELFORA Industries Abattoir. The abattoir is privately owned which supplies lean meat to Kombolcha Meat Factory for canning. The abattoir is located at Kombolcha, South Wollo, at an altitude of 1500 to 1840 m above sea level and about 375 km away from Addis Ababa in the northeast direction (DoARD, 2006).

Study animals and sampling

A total of 535 cattle presented for slaughter at Kombolcha ELFORA Industrial Abattoir were examined for hydatid cyst in the period between August 2010 and May 2011. The study was an active abattoir survey which included cattle brought from different livestock markets to Kombolcha ELFORA Industrial Abattoir. The sample size was determined by 95% confidence interval at a desired accuracy level of 5% (Thrusfield, 1995) and with expected prevalence of 28.4% (Asrat, 1996). Using purposive sampling method, the samples were selected from cattle registered for slaughtering following the antemortem inspection.

\[ n = \frac{1.96^2 \cdot p_{\text{exp}} \cdot (1-p_{\text{exp}})}{d^2} \]

Where \( n \) = required sample size; \( p_{\text{exp}} \) = expected prevalence; \( d \) = desired absolute precision.

Study design

Postmortem examination

A total of 535 cattle presented for slaughter at Kombolcha ELFORA Industrial Abattoir were examined for the presence of hydatid cyst following the routine meat inspection procedures. The inspection procedure used during the postmortem examination consisted of two steps, namely primary and secondary examination. Primary examination involved usual inspection and palpation of organs and viscera followed by a secondary examination if evidence of metacestode was found. The secondary examination involved further incision into each organ if single or more hydatid cyst(s) were found. The liver, lungs, heart, spleen, mesentery and omentum of each animal were examined grossly. Each organ was also incised once or twice with knife. Whenever the cysts were present they were removed, placed in polyethylene bags separately, labeled and taken to the laboratory for further examination.

Identification of cysts was done in the parasitology laboratory of Kombolcha Regional Veterinary Laboratory based on the criteria described by Soulsby (1982). During the study, detailed records of the species, age of the animals, number, size, location and viability of the cyst(s) were made. All animals slaughtered were local zebu breed of cattle at the age of 4 years and above.

Hydatid cyst characterization

The infected organs from each positive animal were collected and recorded. The total number of hydatid cysts was counted and recorded for each infected organ. The size of the diameter of collected hydatid cysts was measured and classified as small (diameter less than 5 cm), medium (diameter between 5 and 10 cm) and large (diameter greater than 10 cm) (Kebede et al., 2009a, c; Oostburg et al., 2000).

Individual hydatid cysts were carefully incised and examined for protoscolices, which resembled white dots on the germinal epithelium; such cysts were characterized as fertile cysts. Fertile cysts were subjected to viability test. A drop of the sediment containing the protoscolices were placed on the microscope glass slide and covered with cover slip and observed for amoeboid like peristaltic movements with 40x objective. For clear vision, a drop of 0.1% aqueous eosin solution was added to equal volume of protoscolices in hydatid fluid on microscope slide with the principle that viable protoscolices should completely or partially exude the dye while the dead ones absorb it (Macpherson et al., 1985). Furthermore, infertile cysts were further classified as sterile or calcified. Sterile hydatid cysts were characterized by their smooth inner lining usually with slightly turbid fluid in their content. Typical calcified cysts produce a gritty-sound heard at incision (Soulsby, 1982).

Financial loss estimation

Direct and indirect losses were the basis for the estimation of the annual economic losses. Direct losses were calculated on the basis of condemned organs, whereas the indirect losses were estimated on the basis of live weight loss caused by hydatidosis (Polyodorou, 1981; Torgerson and Dowling, 2001).

The parameters considered for the estimation of financial loss were five percent estimated carcass weight loss due to hydatidosis (Polyodorou, 1981), slaughter rates of animals at Kombolcha ELFORA Industrial Abattoir, average carcass weight (dressing percentage) of Ethiopian Zebu cattle breed (126 kg) and the mean retail market price of condemned organs due to hydatidosis such as lung, liver, kidney, spleen. The total financial loss due to hydatidosis was the sum of direct and indirect losses.

Direct and indirect financial losses were calculated on a yearly basis. Average market price of lung, liver, spleen, kidney, heart and a kilogram of beef was found to be 30, 45, 15, 10, 15 and 80 Ethiopian Birr (ETB), respectively. The mean annual numbers of cattle slaughtered during the last one year were 5000. Average number of cattle positive for hydatidosis as it was extrapolated from prevalence findings on ELFORA abattoir were 17.4%.

Direct losses were calculated as follows:

\[ DL = (AS \times CLu \times PLu) + (AS \times CLI \times PLi) + (AS \times CSp \times PSp) + (AS \times CKid \times PKid) + (AS \times CHr \times PHr) \]

Where \( DL \) = direct losses associated with hydatidosis, \( AS = \)

...
estimated mean annual slaughter; PLu = percent involvement of the lung; CLu = local retail price of a lung; PLi = present involvement of the liver; CLI = local retail price of a liver; PSp = present involvement of the spleen; CSp = local retail price of a spleen; PKid = percent involvement of the kidney; CKid = local retail price of a kidney; PHr = percent involvement of the heart; CHr = local retail price of a heart.

Indirect losses (IL) = 5% NAS × PH × CPB × 126 kg

Where 5% = A reduction of 5% in meat production due to hydatidosis established by Polydorou (1981); NAS = average number of cattle slaughtered annually; PH = prevalence rate of hydatidosis; CPB = current average price of 1 kg of beef at Kombolcha; 126 kg is the dressed average carcass weight of adult Zebu cattle (ILCA, 1979).

Total economic loss (TL)

The total economic loss can be evaluated by considering both DL and IL as follows:

\[ TL = DL + IL \]

Data analysis

Data collected from antemortem, postmortem and laboratory finding were entered in to MS Excel and statistical packages (SPSS Version 18) were employed to analyze the results.

RESULTS

This study demonstrated that at Kombolcha ELFORA Industrial Abattoir, 93 (17.4%) were found harboring one or more hydrated cyst. The result obtained from postmortem examination indicated that different organs were affected with hydatid cyst.

Single and multiple infection of organs were recorded out of the total 93 cattle harboring hydatid cysts: 87 (93.5%) were found involving only a single organ and the remaining 6 (6.5%) had a multiple organ involvement. Among the different organs affected, lung and liver constituted the highest infection rate (Table 1).

Cyst size

Exceptionally two large cysts were found in the lung and spleen of cattle measuring 18 and 17 cm in diameter and containing about 1.5 and 1.3 L of fluid, respectively. The total cyst count with respect to size in each infected organ for cattle was described in Table 2.

Cyst fertility, viability and sterility

Fertility and sterility of hydatid cyst was described. The viability percentage of protoscolices was higher in the lung (5.9%) than in the other organs while the percentage of calcified cysts was 36.8%, the highest in the liver (Table 3).

Direct financial loss

Due to cattle hydatidosis, 51 lungs, 41 livers, 5 hearts, 2 kidneys and 2 spleens were condemned during the study period with an economic loss of 1530, 1845, 125, 20 and 30 ETB, respectively (Table 4). This was assessed from the mean retail market price of each organs and the total number of organs condemned during the study period. Annual economic loss on the other hand was estimated considering annual slaughter rate of cattle and prevalence of hydatidosis per organ and was calculated to be 24,423.6 ETB per annum.

Indirect loss

The estimated economic loss from carcass weight loss due to hydatidosis was estimated to be 274,050 ETB. Therefore, the total estimated annual economic loss in cattle at Kombolcha ELFORA Industrial Abattoir due to hydatidosis was 302,023.6ETB ($1 = 17.00 ETB).

DISCUSSION

The occurrence of hydatidosis in cattle was found to be 17.4% during the study period in Kombolcha ELFORA Industrial Abattoir. The current finding is almost similar to that reported as 16.85% (Jemere and Butako, 2011) in Wolaita Sodo, 15.4% (Regassa et al., 2009) in Hawassa and 16% (Kebede et al., 2009b) in Wolaita Sodo. In general terms, throughout the world, there had been different magnitude records of hydatidosis in cattle with low medium and high rates of occurrences. High prevalence rates were registered in other areas of the country such as 61% in Assela (Koskei, 1998), 52.69% in Hawassa (Regassa et al., 2010), 34.05% in Bahir Dar (Kebede et al., 2009a), 46.5% in Debere Zeit (Jobre et al., 1996), 48.9% in Debre Markos (Kebede et al., 2009c), 32.1% in Mekelle (Berhe, 2009), and 22% in Tigray (Kebede et al., 2009d).

Factors such as difference in culture, social activity, animal husbandry systems, lack of proper removal of infectious carcass, and attitude to dogs in different regions might have contributed to the variation in prevalence in different areas of a country (Arbabi and Hooshyr, 2006) and strain differences of E. granulosus that exists in different geographical location (McManus, 2006). In the current study, hydatid cysts were found predominantly in lung and liver representing 50.5 and 40.6 %, respectively. Literature reveals that hydatid cysts are most commonly found in lungs and liver of ungulates (Hubbert et al., 1975) and it is in agreement with the
Table 1. The total number, relative prevalence and number of cysts harbored in affected organ.

<table>
<thead>
<tr>
<th>Organ</th>
<th>Infected number of organ</th>
<th>Relative prevalence</th>
<th>Total number of cyst</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lung</td>
<td>51</td>
<td>50.5</td>
<td>135</td>
</tr>
<tr>
<td>Liver</td>
<td>41</td>
<td>40.6</td>
<td>125</td>
</tr>
<tr>
<td>Heart</td>
<td>5</td>
<td>5</td>
<td>10</td>
</tr>
<tr>
<td>Kidney</td>
<td>2</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>Spleen</td>
<td>2</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>Total</td>
<td>101</td>
<td>-</td>
<td>276</td>
</tr>
</tbody>
</table>

Table 2. Cyst size and counts in relation with organ involvements in infected cattle slaughtered in Kombolcha ELFORA.

<table>
<thead>
<tr>
<th>Organ</th>
<th>Small</th>
<th>Percentage</th>
<th>Medium</th>
<th>Percentage</th>
<th>Large</th>
<th>Percentage</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lung</td>
<td>70</td>
<td>51.9</td>
<td>30</td>
<td>22.2</td>
<td>15</td>
<td>11</td>
<td>115</td>
</tr>
<tr>
<td>Liver</td>
<td>58</td>
<td>46.4</td>
<td>17</td>
<td>13.6</td>
<td>4</td>
<td>3.2</td>
<td>79</td>
</tr>
<tr>
<td>Heart</td>
<td>4</td>
<td>40</td>
<td>-</td>
<td>-</td>
<td>1</td>
<td>10</td>
<td>5</td>
</tr>
<tr>
<td>Kidney</td>
<td>3</td>
<td>75</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>3</td>
</tr>
<tr>
<td>Spleen</td>
<td>2</td>
<td>100</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>2</td>
</tr>
<tr>
<td>Total</td>
<td>137</td>
<td>49.6</td>
<td>47</td>
<td>17</td>
<td>20</td>
<td>7.24</td>
<td>204</td>
</tr>
</tbody>
</table>

Table 3. Type of hydatid cyst sterile, fertile and calcified indifferent organs of infected cattle.

<table>
<thead>
<tr>
<th>Organ</th>
<th>Sterile</th>
<th>Percentage</th>
<th>Fertile</th>
<th>Percentage</th>
<th>Calcified</th>
<th>Percentage</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lung</td>
<td>107</td>
<td>79</td>
<td>8</td>
<td>5.9</td>
<td>20</td>
<td>14.8</td>
<td>135</td>
</tr>
<tr>
<td>Liver</td>
<td>61</td>
<td>48.8</td>
<td>4</td>
<td>3.2</td>
<td>46</td>
<td>36.8</td>
<td>125</td>
</tr>
<tr>
<td>Heart</td>
<td>5</td>
<td>50</td>
<td>-</td>
<td>-</td>
<td>5</td>
<td>50</td>
<td>10</td>
</tr>
<tr>
<td>Kidney</td>
<td>3</td>
<td>75</td>
<td>-</td>
<td>-</td>
<td>1</td>
<td>25</td>
<td>4</td>
</tr>
<tr>
<td>Spleen</td>
<td>2</td>
<td>100</td>
<td>-</td>
<td>-</td>
<td>0</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Total</td>
<td>178</td>
<td>67.9</td>
<td>12</td>
<td>4.3</td>
<td>72</td>
<td>26</td>
<td>276</td>
</tr>
</tbody>
</table>

Table 4. Direct economic losses associated with CE in infected cattle in Kombolcha ELFORA industrial abattoir, Northeast Ethiopia.

<table>
<thead>
<tr>
<th>Organ</th>
<th>No. of organs condemned</th>
<th>Price per organ</th>
<th>Total price ETB</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lung</td>
<td>51</td>
<td>20</td>
<td>1530</td>
</tr>
<tr>
<td>Liver</td>
<td>41</td>
<td>15</td>
<td>1845</td>
</tr>
<tr>
<td>Heart</td>
<td>5</td>
<td>25</td>
<td>125</td>
</tr>
<tr>
<td>Kidney</td>
<td>2</td>
<td>10</td>
<td>20</td>
</tr>
<tr>
<td>Spleen</td>
<td>2</td>
<td>15</td>
<td>30</td>
</tr>
<tr>
<td>Total</td>
<td>101</td>
<td>61</td>
<td>3,550</td>
</tr>
</tbody>
</table>

findings of Bekele and Butako (2011), Njoroge et al. (2002) and Eckert and Deplazes (2004), which show that the lung and liver are the most common sites of hydatid cyst in domestic animals. This could be justified by the fact that lungs and liver possess greater capillary fields, which allow these organs to efficiently filter the ingested oncospheres from the blood liver and lungs undergo sequential filtration of blood, liver undergoes primary filtration of blood from portal veins which is followed by pulmonary filtering actions before other organs are
invaded (Eckert and Deplazes, 2004). Only those oncospheres which transfer the blood will reach the systemic circulation and other tissues (Eckert and Deplazes, 2004; Matosain, 1977).

High numbers of small, medium and large size cysts were found in lungs than in the liver, while the liver harbored higher number of calcified cysts. The reason for higher percentage of small, medium and large cysts in the lungs is due to soft structure of the lung, while the higher yield of calcified cysts in liver could be attributed to relatively higher reticuloendothelial cells and abundant connective tissue reaction of the organ. The higher proportion of a small cysts may be due to immunological response of the host which might preclude expansion of cysts life (Torgerson et al., 1998; Lahmar et al., 1999; Larrieu et al., 2001; Torgerson, 2002). In examining the condition of cyst fertility and viability, the findings of 67.9% sterile, 4.3% fertile and 26% calcified were examined. It may be concluded that most of the cysts in cattle were infertile. The variation infertility rate among different species and in different geographical zone could be due to the differences in the strain of E. granulosus (McManus, 2006). Most of the hydatid cysts from cattle are considered to be sterile (Thompson et al., 1984).

In comparison of the fertility rate among the organs, it was higher in lungs than in liver. It has been stated that the relatively softer consistency of the lung tissue allows easier development of the cysts and the fertility rate of hydatid cysts may show a tendency to increase with advancing the age of the hosts (Himons et al., 1987). This may be attributed to reduced immunological compatibility of animals at their older age of infection. The variation between tissue resistances of the infected organs may also influence the fertility rate of hydatid cysts. The fertility rates observed in this study are law; however, could serve as potential source to infection and per-petuate the cycle of hydatidosis when infected animals are slaughtered and raw offal fed to dogs and also leftovers during backyard slaughter are eaten by wild carnivores. It was observed that majority of the households had livestock, including cattle, sheep, goat, and donkeys, which are the intermediate host of the parasite. Similarly, many households had dogs and cats, which were not dewormed regularly and were managed under free-range system. In this study, hydatidosis was found to incur financial loss that is estimated to be 202,023.60 ETB to the cattle industry per year. Previous studies have also estimated the annual financial losses associated with bovine hydatidosis from other parts of the country. For instance, Bekele and Butamo (2011) reported 410,755.90 ETB (30,202.64 US$), Kebede et al. (2009b) reported 25,608 ETB, Regassa et al. (2010) reported 1,791,625.89 ETB. The difference in the calculated economic loss in the various abattoirs is either due to the variation in the number of slaughtered animals or variation in the prevalence rate of hydatidosis or due to variation in the retail market price of organs. Based on our results, we suggest that bovine hydatidosis is among many of the livestock diseases prevailing in the country incurring both direct and indirect losses to the cattle industry due to condemnation of organs and reduced live weight gain of infected cattle.

In Kombolcha ELFOR Industrial Abattoir, condemned organs and carcasses are buried and some of them are further processed for animal feed. This may reduce the contamination and infection in dogs and other carnivores. Therefore, the decreased prevalence of hydatidosis in this study as compared to the previous studies conducted in different areas of the country could be due to proper disposal of the condemned organ where dogs have no access to infected organs. However, there are backyard slaughter practices during local festivities, the tradition of offering uncooked infected offals to pet animals around homestead, poor public awareness about the diseases, and the habit of disposing dead wild or domestic animals unburied and left open for scavenging carnivores creates favorable condition for maintaining the life cycle of E. granulosus in stray dogs and wild carnivores in the area.

Conclusion

This study highlights the need for thorough investigation to obtain appropriate and more accurate information on the incidence of hydatidosis/echinococcosis in humans, dogs, both in domestic and wild animals to determine the scope and type of relevant control options. In spite of the low magnitude of infection detected, there seems to be an existing socioeconomic condition favorable for hydatidosis, and hence, it remains one of the most important diseases warranting serious attention for prevention and control actions.

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REFERENCES


Full Length Research Paper

Molecular detection of canine parvovirus in Jos, Nigeria


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Canine parvovirus (CPV) remains the most significant viral cause of enteritis in puppies over the age of two months. This study was meant to detect the virus by targeting the VP2 gene, a 583 bp gene (nucleotide 4003 to 4585) of the capsid protein. The detection of the virus was carried out by conventional polymerase chain reaction (PCR) on one hundred and nine samples. Seventy five of these were rectal swabs while thirty four were necropsy tissues, all from dogs presenting with symptoms suggestive of parvovirus enteritis from veterinary clinics in Jos metropolis. Results revealed that 47.70% of the samples were found to be positive for the virus. The rate of detection was more in necropsy tissues (64.71%) relative to rectal swabs (40.00%). A breakdown of the incidence of the infection across breeds of dogs showed that the Rottweilers had 60.00%, Doberman pinchers had 54.54%, Pitbulls had 53.33% while Tan coloured and local breeds of dogs had 50.00 and 21.42%, respectively. With a prevalence rate of 47.70%, the findings have confirmed that the virus is circulating in Jos, Nigeria. As such, stake holders must quickly intervene to arrest the situation given the high economic losses associated with the disease.

Key words: Canine parvovirus, polymerase chain reaction (PCR), Jos.

INTRODUCTION

Canine parvovirus (CPV) is a highly contagious virus mainly affecting dogs. The disease, commonly called parvovirus enteritis is highly infectious and is transmitted from dog to dog by direct or indirect contact with contaminated feces (Appel et al., 1979). Parvovirus enteritis is a relatively new disease that appeared in the late 1970s. It is highly contagious and often fatal in both domestic and wild dog populations with high morbidity (100%) and frequent mortality of up to 10% (Appel, 1978). It is characterized by vomiting, bloody diarrhea, myocarditis and leucopenia (Streck et al., 2009). CPV was first recognized in 1978 and spread worldwide in one to two years (Carmichael, 2005). The virus is a small, non enveloped single stranded DNA virus belonging to the Parvoviridae family and parvovirus genus under the Parvovirinae subfamily (Hong et al., 2007). The virus, which first appeared between 1977 to 1978, probably arose from a very closely related virus in cats, feline panleukopaenia virus (FPLV) through a small number of mutations in the single capsid protein; a species jump which may have involved intermediate passage in other carnivores such as mink or raccoons (Truyen et al., 1996). As early as 1979 the first
variants of CPV2 appeared, termed CPV2a, and they were quickly followed by the appearance of CPV2b in 1984 (Parrish et al., 1985, 1991). The original type 2 virus has now disappeared from the field having been replaced by the 2a and 2b variants; although the relative proportions of these two types varies from country to country (Truyen et al., 1996; Chinchkar et al., 2006; Pereira et al., 2007). However, in Africa, including Nigeria, no reports as regarding the circulation of the virus in the area is available (Kapil et al., 2007). This study is aimed at detecting the virus using polymerase chain reaction (PCR) in Jos, Nigeria.

MATERIALS AND METHODS

A total of one hundred and nine samples were collected. Seventy five of which were rectal swabs while thirty four were necropsy tissues. The rectal swabs were collected from dogs presenting with bloody diarrhea and vomiting signs while necropsy tissues of target organs (intestine, heart, lymph nodes, spleen and liver) were harvested from dogs that died from the disease.

Sample collection

Rectal swabs were collected by the insertion of the cotton bud of a swab stick (Steriline®) through the anal region of the animal into the rectum after the dog has been restrained. The swab stick was carefully turned clockwise and gently withdrawn. A scissors was used to cut off the swab bud into 2 ml cryovial containing virus transport medium (VTM) and transported to the laboratory in a cold box. Necropsy tissues of target organs were collected from dead dogs. They were aseptically delivered into a universal sample bottle (Steriline®) and transported to the laboratory in a cold box.

Table 1. Percentage prevalence according to sample type.

<table>
<thead>
<tr>
<th>Sample type</th>
<th>PCR positive (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Swabs (n = 75)</td>
<td>30 (40.00)</td>
</tr>
<tr>
<td>Tissues (n = 34)</td>
<td>22 (64.71)</td>
</tr>
<tr>
<td>Total (T = 109)</td>
<td>52 (47.70)</td>
</tr>
</tbody>
</table>

Table 2. Percentage positivity of samples based on breed of dog.

<table>
<thead>
<tr>
<th>Breed</th>
<th>PCR positive (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rottweilers (n = 25)</td>
<td>15 (60.00)</td>
</tr>
<tr>
<td>Doberman Pinchers (n = 33)</td>
<td>18 (54.54)</td>
</tr>
<tr>
<td>Pit Bulls (n = 15)</td>
<td>08 (53.33)</td>
</tr>
<tr>
<td>Tan Coloured (n = 04)</td>
<td>02 (50.00)</td>
</tr>
<tr>
<td>Local (n = 28)</td>
<td>06 (21.42)</td>
</tr>
<tr>
<td>Unidentified (n = 04)</td>
<td>03 (75.00)</td>
</tr>
<tr>
<td>Total (T = 109)</td>
<td>52 (47.70)</td>
</tr>
</tbody>
</table>

Polymerase chain reaction (PCR)

Total DNA was extracted using the QIAamp® DNA Mini Kit according to manufacturer’s specifications. Different procedures were adopted for DNA extraction from swabs and DNA extraction from tissues.

CPV-2 specific master mix

The master mix contained 13.88 µl of nuclease free water (Promega®), 2.5 µl of 10× PCR reaction buffer, 0.62 µl of dNTP mixture (10 mM) (Fermentas®), 1.5 µl of magnesium chloride (25 mM), 0.5 µl of forward and 0.5 reverse primers (555 forward and 555 reverse, respectively) for CPV2 (Inqaba Biotech SA®) at 20 pmol/µl and 0.5 units of Taq DNA polymerase (Qiagen®). This gave a total volume of 20 µl master mix. 5 µl of the CPV-2 master mix (above) was added to 5 µl extracted DNA in a new tube for amplification.

Amplification of DNA extracts

The mixture was submitted to a thermal cycling profile of initial denaturation at 94°C for 5 min. This was followed by 30 cycles of denaturation at 94°C for 30 s, annealing at 55°C for 2 min, extension at 72°C for 2 min and 72°C for 10 min in an Applied Biosystem® 9700 PCR machine. The primer sequences used were as follows: 555 forward, 5′-CAGGAAAGATATCCAGAAGGA-3′ and 555 reverse, 5′-GGTGCTAGTTGATATGTAATAAACA-3′. The master mix composition and PCR program was as described by Streck et al. (2009) with minor modifications.

Electrophoresis and analysis

A 10 µl of PCR amplicons was electrophoresed in a 1.5% agarose gel stained with ethidium bromide in the presence of 2 µl of gel loading buffer (Fermentas®). Positive and negative controls were included. The electrophoresis was carried out at 80 volts for 50 min.

RESULTS

Amplicon band sizes of about 583 bp were considered positive for the virus. Results revealed that 52 (47.70%) of the one hundred and nine samples were positive for the virus.

From the seventy five rectal swabs examined, 30 (40.00%) of them were positive for CPV just as 22 (64.71%) out of the thirty four tissue samples examined and were positive for the virus by conventional PCR (Table 1). Statistically, there was no significant difference (P > 0.05) in the number of positive samples from the sample types surveyed (Table 1). Although the sample size was small for breed comparison, Rottweillers had 60.00% incidence, Doberman pinchers 54.54%, Pitbulls 53.33%, Tan coloured 50.00% while local breeds of dogs had only 21.40% (Table 2).

DISCUSSION

Canine parvovirus continues to be an important pathogen...
of dogs and it is responsible for serious occurrences of morbidity and mortality despite the availability of safe and effective vaccines (Decaro et al., 2006a,b). This study revealed that 52 out of the 109 samples examined had the virus. This study therefore revealed a prevalence rate of 47.70% of the disease from this part of the world. The figure is less compared with seven positive samples of CPV 2c strain obtained from nine samples (77.78%) assayed in Brazil (Streck et al., 2009). Also, it is lower than the findings in Uruguay where 24 out of 25 faecal samples (96.00%) were found to be positive for one strain of the virus (Perez et al., 2007). The high percentage of positive tissues against rectal swabs is not unconnected with the fact that the selected tissues had earlier been established as the predilection sites of the agent as such, it is only natural that the virus concentration in such sites be higher (Lobetti, 2003).

Local breeds of dogs were the least susceptible to the infection as against their foreign counterparts. Authors had previously suggested that the former have a greater degree of resistance against the virus than the foreign breeds. They have also been fingered as healthy carriers of the virus (Nelson and Couto, 1998). This is of great epidemiological relevance as their role in the distribution of the virus is once again re-echoed. Since most of the local breeds are free rangers, it makes it possible for them to distribute the virus indiscriminately. This factor poses a great danger to the foreign breed of dogs which are more susceptible to the agent as their local counterparts (Nelson and Couto, 1998).

Although canine parvovirus was implicated in 52 (47.70%) of the samples as causative agent responsible for enteritis in these dogs, the cause of enteritis in 57 (52.29%) PCR-negative samples may be due to other pathogens such as Leptospira or Eimeria species. In general, these findings will no doubt interest both the local and international communities as they have waited all this while for a report on the agent from this part of the world (Kapil et al., 2007).

REFERENCES


UPCOMING CONFERENCES

11th International Congress on the Biology of Fish, Edinburgh, Scotland, 3 Aug 2014

International Conference on Coelenterate Biology, Eilat, Israel, 1 Dec 2013
Conferences and Advert

**October 2013**
11th World Conference on Animal Production, Beijing, China, 15 Oct 2013

**September 2013**
International Conference on Optimizing Productivity of Ruminants, Poultry, Rabbits and Fishes, Marsa Alam, Egypt, 2 Sep 2013