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Use of Mexican Propolis for the Topical Treatment of Dermatomycosis in Horses

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Abstract

Five horses with lesions compatible with dermatomycosis were selected and skin scrapings were taken from all affected areas. Samples were then treated with 10% KOH and observed by direct microscopic examination; fungal cultures using Mycobiotic Agar and Borelli Agar were also preformed; *Trichophyton mentagrophytes* was identified in two cases, whereas *Candida albicans* was identified in the other three. Skin lesions in cases 1 and 2 were treated topically with ketoconazol ointment, while in the remaining cases a weekly bath with propolis-based shampoo and application of propolis-based ointment on the lesions 2 to 3 times a week were implemented. Treatment in all cases lasted 4 weeks. Pictures were taken before and after treatment and, making use of the Image Pro-Express® software, therapy efficacy was determined by measurement of lesion area diameter and a significant reduction in lesion size was observed by the end of the 4 weeks of treatment with ketoconazole and propolis-based products. Thus proving the effectiveness of such propolis-based products, making them a natural therapeutic alternative for the treatment of cutaneous mycoses in horses, and carrying none of the toxic side effects of conventional pharmaceuticals.

Keywords

Mexican Propolis, Horses, Dermatomycosis, Treatment

1. Introduction

Superficial and cutaneous mycoses are fungal infections that involve the superficial layers of the skin, hair, and claws/hoofs. The microorganisms involved are often keratinophilic dermatophytes, such as those belonging to

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the genus *Microsporum*, *Trichophyton* and *Epidermophyton*; however, other fungi like the yeasts *Candida*, *Malassezia* and *Trichosporon* may also produce superficial mycoses [1]. Cutaneous mycoses in horses are of great importance in veterinary medicine due to the zoonotic potential of most of the causative agents, the possibility of transmission between horses and to other species, the discomfort experienced by the infected animals, the aesthetic appearance of the skin, as well as the elevated costs of the pharmacological treatment [2], which tends to extend for long periods of time and can lead to possible side effects, not to mention the contraindications and resistance problems encountered with such treatments. For this reasons, in the last few years, there has been a growing interest in developing therapeutic compounds based on natural antimicrobial products that are easily applied and cheaper. Propolis is one of these products, a resin collected by bees and to which the presence of several compounds in its composition confers it with antibacterial, antiviral, fungicidal, anti-inflammatory, and immunomodulatory activities, all of which have been exhaustively investigated for years [3] [4]. Previous works have already demonstrated the benefits of the use of propolis for the treatment of cutaneous burn injuries in horses [5]. The purpose of this work is to provide a therapeutic alternative for the treatment of superficial mycoses which is practical and efficient, so that any patient can achieve full recovery with the use of a practical, economical and effective topical treatment that poses no risk to both, the patient and humans.

2. Materials and Methods

2.1. Cases

A total of 5 horses of both genders (Thoroughbred and American Quarter) were selected from different stables, ages ranging from 2 to 7 years, and weights between 430 and 540 kg, each of them presented clinical signs compatible with cutaneous mycosis (annular areas of peripherally expanding alopecia, with or without erythematous borders, and fragile hair) but without any other apparent health alteration that could interfere with the results. The number and size of the lesions in each horse, as well as their distribution, was recorded in a work sheet that depicted the anatomical regions of the horse; such evaluation was made before and after treatment. Case 1: American Quarter horse, 5 years old, sorrel. Showed a large alopecic region that covered the caudal aspect of the head, from the ear base to the bottom of the jaw, and another small alopecic lesion on the temple, both on the right side on the face (Figure 1(a)). Each horse was privately owned and thus welfare concerns were directly supervised by staff members of a given stable, which was different for each horse.

Case 2: American Quarter mare, 7 years of age, light sorrel. Showed multiple annular alopecic lesions on the lacrimal, zygomatic, and maxillary bone regions, on the right and left side of the face (Figure 2(a) and Figure 2(c)).



Figure 1. Case 1: Horse, American Quarter. (a) Lesion caused by *Trichophyton mentagrophytes* on the caudal aspect of the head, from the ear base to the bottom of the jaw, and another small alopecic lesion on the temple, both on the right side; (b) Same areas after 4 weeks of ketoconazole ointment treatment.

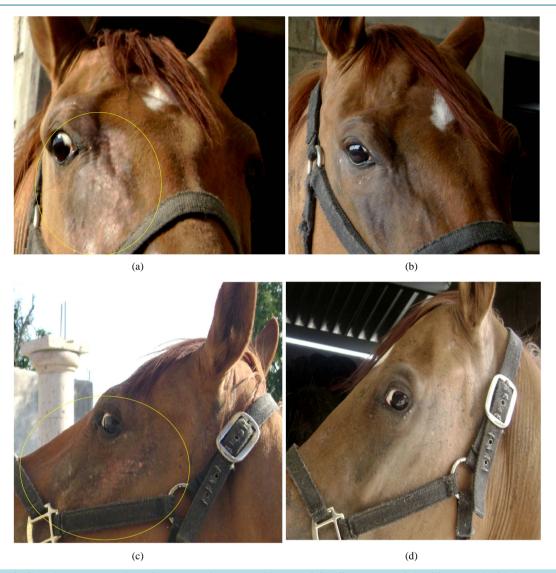


Figure 2. Case 2: Mare, American Quarter. Lesions caused by *Candida albicans*. (a) Right side, treated with ketoconazole ointment; (b) Same area after 4 weeks of treatment; (c) Left side, treated with propolis-based shampoo and ointment; (d) Same area after 4 weeks of treatment.

Case 3: Thoroughbred filly, 2 years of age, dark bay. Showed multiple annular alopecic lesions on the craneoventral part of the barrel (Figure 3(a)).

Case 4: Thoroughbred colt, 2 years of age, brown. Showed multiple annular alopecic on the breast (Figure 4(a)).

Case 5: Thoroughbred filly, 2 years of age, brown. Presented several annular alopecic lesionson the girth, of approximately 2 cm in diameter each (Figure 5(a)).

2.2. Laboratory Procedures

For each mycological examination, hair samples were taken and scales were collected from the edges of thelesions for direct microscopic examination with 10% KOH. These samples were then used to inoculate plates containing Mycobiotic Agar (Bioxon®) and Borelli Agar [6]. After that, Petri dishes were incubated at 28°C for one week, by the end of which we were able to observe the formation of growing colonies. Using Riddel's microculture technique, identification was performed based on the morphological characteristics, using the monographs of Larone [7]. In order to observe yeast-like structures, Gram stains were performed to those colonies

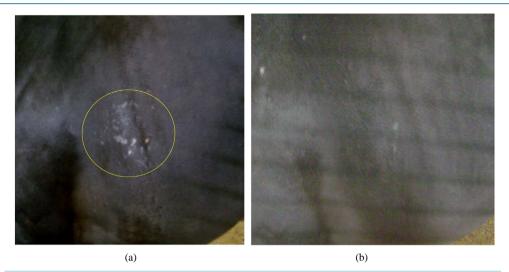


Figure 3. Case 3: Filly, Thoroughbred. Skin infection caused by *Candida albicans* on the craneoventral part of the barrel. (a) Annular alopecic lesions; (b) Same area after 4 weeks of propolis-based shampoo and ointment.



Figure 4. Case 4: Colt, Thoroughbred. (a) Skin lesions caused by *Candida albicans* on the left side of the breast; (b) Same area after 4 weeks of propolis-based treatment.

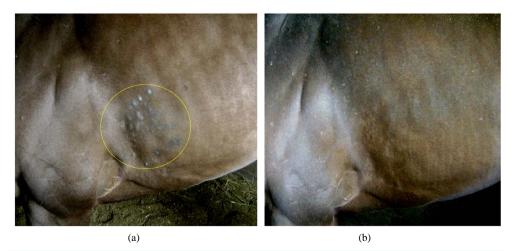


Figure 5. Case 5: Filly, Thoroughbred. (a) Annular alopecic lesions on the girth caused by *Trichophyton mentagrophytes*; (b) Same area after 4 weeks of propolis-based treatment.

whose whitish, creamy appearance corresponded to that observed in *Candida* spp. cultures. A germ tube test, also known asmycelial growth test, was then performed in order to identify the species [8].

2.3. Treatment

In case 1, ketoconazole ointment (Nizoral, Janssen-Cilag[®]) was applied on the affected areas; In case 2, ketoconazole ointment was also applied, but only on the lesions on the right side of the face, whereas propolis-based products were used to topically treat the lesions on the left side. The three remaining cases were only treated with propolis-based compounds, a weekly bath was given with propolis-based shampoo (consisting of both, oily- and soapy-phases, excipients and 3% propolis) and 15% propolis ointment (the composition of which was taken from a previous publication by Cruz [9]) was applied on the lesions 2 to 3 time a week. Treatment in all cases lasted 4 weeks. Propolis' active substances were determined by HPLC analisis, the most relevant being flavones, cinnamic acid and caffeic acid [10]. Both propolis-based products were supplied by the Multidisciplinary Research Unit of the FES-Cuautitlán, UNAM.

2.4. Evaluation and Statistic Analysis

Photographs of the lesions were taken before and after treatment; the pictures were then analyzed with the Image-Pro Express 4.5 software and data corresponding to lesion size changes and distribution were recorded. After that, an ANOVA test (GraphPad Prism software) of the data corresponding to the size of the lesions before and after treatment was carried out and a p value < 0.005 was obtained, so that a statistically significant reduction in lesion size was confirmed.

3. Results

Samples were treated with 10% KOH and a direct microscopic examination was performed. In cases 1 and 5, ectothrix parasitism of hair shafts was observed and, in both cases, the dermatophyte *Trichophyton mentagrophytes* was isolated, whereas in the remaining cases (2, 3, and 4) the yeast *Candida albicans* was identified.

By the end of the 4 weeks of treatment it was evident that the skin lesions of the horses treated with propolisbased shampoo and ointment showed a reduction in the size of the area involved, thus resulting in lesion size reduction of statistical significance. No recidivism of the lesions was observed in any case (Table 1; Figure 1(b), Figure 2(b), Figure 2(d), Figure 3(b), Figure 4(b) and Figure 5(b)).

4. Discussion

Previous works have already shown the antifungal activity of propolis against dermatophytesin dogs, cows and rabbits [4] [11]; also, its *in vitro* activity against *Trichophyton mentagrophytes* has been noted in several studies [12]. Furthermore, the present work not only succeed in demonstrating the effectiveness of propolis for the treatment of dermatophytosis in horses caused by *Trichophyton*, but also proved its viability as an effective treatment for skin infections caused by *Candida albicans*.

Table 1. Summary of results obtained in horses topically treated with ketoconazole and propolis*.

Case	Breed	Age (years)	Gender	Isolated microorganism	Treatment	Area before (cm)	Area after (cm)	Lesion reduction (%)
1	American Quarter	5	Male	Trichophyton mentagrophytes	Ketoconazole	55.27	2.44	95.58
2	American Quarter	7	Female	Candida albicans	Ketoconazole(rs) Propolis(ls)	86.15 31.9	0.24 0.16	99.72 99.50
3	Thoroughbred	2	Female	Candida albicans	Propolis	55.61	0.58	98.95
4	Thoroughbred	2	Male	Candida albicans	Propolis	20.01	0.60	97.05
5	Thoroughbred	2	Female	Trichophyton mentagrophytes	Propolis	20.17	0.30	98.51

p = 0.005; rs = right side; ls = left side.

The effectiveness of propolis for the treatment of *C. albicans* in horses is probably due to its ability to inhibit yeast growth, blastoconidia formation and germ tube formation (an important virulence factor of *C. albicans*), as well as its damaging effect on cell membrane and wall. Such damages have also been reported by [13], who demonstrated that, *in vitro*, a propolis concentration of 0.3 mg/ml is capable of inhibiting yeast growth and blastoconidia formation (yielding the microorganism incapable of reproducing), and creates morphological alterations by means of cellular membrane and wall damage, and even nuclear membrane damage.

A synergic effect with the combined use of propolis-based shampoo and oitment was also observed. Tensoactive activity is provided by several components of the shampoo (sodium lauryl sulfate, amines, sugar alcohols, cocamidopropyl betaine), which eliminate sweat, ceruminous secretions, dead cells, and dirt form the environment, as mentioned by [14]. Once these substances have been removed from the skin's surface, a better residual activity from the propolis contained in the ointment is achieved. A similar study on dermatophytosis in cows was conducted by [11], wherein he mentions that better results are obtained when the application of propolis-based ointment is combined with the application of another oitment that contains benzoic and salicylic acids, instead of using one product alone.

In the present study, topical treatment protocols were implemented for a period of 4 weeks, which has relevance when dealing with extensive skin lesions and correlates with a study in dogs by [9], were patient treatment consisted of a daily wash of the lesions with any commercially available propolis-based soap, followed by the application of a propolis-based ointment; horses were checked every 7 days and complete remission of the lesions and hair growth in affected areas were achieved by the third week of treatment, as well as an overall reduction of pruritus caused by secondary bacterial infection, and increased patient acceptance of treatment application.

The regenerative and wound healing capacities of propolis on skin lesions in horses were also observed in this study, as was proven by size reduction of annular alopecic areas, which was also demonstrated by Howaida *et al.* [5], who evaluated the efficacy of propolis to enhance skin regeneration of old and recent lesions. On the other hand, the work here presented also succeed in demonstrating resolution of skin lesions in horses caused by both, dermatophytes and yeast, after the topical application of propolis-based products, which in turn is associated with the antimicrobial properties of the resin, the presence of free radicals and metabolism stimulation, which speeds up tissue regeneration [15]-[17].

The presence of substances such as caffeic acid (present in our propolis) and kaempferol could also play an important role in tissue regeneration. These substances induce the anagen phase of the hair cycle, as demonstrated by Miyata *et al.* [18], who conducted an investigation in which dilutedethanol-extracted propolis was topically applied to mice that had been previously shaved or wax depilated. Their results showed that the subjects that had received topical treatment with propolis recovered their hair coat faster than those who had not received such treatment, due to the fact that anagen phase induction occurred without any detectable anomalies in hair follicle conformation when this treatment was implemented. The same authors also observed that, after the topical application of the product, the number of cells associated with hair growth increased and keratinocyte proliferation was stimulated.

Regarding case number 2, were the lesions on the right side of the face were treated with ketoconazole ointment, the presence of a mild hyperpigmentation of the area was observed after the resolution of the lesions, which did not happen on the left side, where instead propolis-based products were used, nor did it happen in case 1, where ketoconazole ointment alone was used. Such secondary effect of ketoconazole has not been documented, which lead the authors to consider it an isolated reaction.

The results here presented are non-conclusive, given the small amount of treated animals; however they do provide some guidance for further research on the subject. An important aspect worth of mentioning is that relating to treatment cost per animal, which was of approximately \$100 (USD) for the whole treatment with keto-conazole ointment, and of approximately \$30 (USD) when propolis-based products were used, hence, a significant reduction in treatment cost can be appreciated without compromising the outcome in lesion size reduction, which is similar with both treatments

These research paper adds up to the ever growing scientific literature that present the use of propolis as an attractive alternative therapeutic agent against fungal skin infections that can be safely employed in pediatric, geriatric and pregnant patients, and even in those with renal and/ or hepatic failure, which constitutes a significant advantage over treatments with conventional pharmaceuticals, that while is true are effective, their use is limited in the treatment of such patients, due to their hepatotoxic, embryotoxic and teratogenic effects, as well as many drug interactions [19] and [20].

5. Conclusion

After a thorough review of the currentscientific literature available on the subject, not a single paper about the use of propolis for the treatment of dermatomycosis in horses was found, so it is possible that this work constitutes the first written scientific report of the application of propolis for the treatment of dermatomycosis in this species.

Acknowledgements

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A Survey for Contagious Caprine Pleuropneumonia in Agago and Otuke Districts in Northern Uganda

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Abstract

Background: Contagious Caprine Pleuropneumonia (CCPP) is a devastating disease of goats caused by Mycoplasma capricolum subsp. Capripneumoniae (Mccp). The disease was first confirmed in Uganda in 1995 in Karamoja region. Contagious Caprine Pleuropneumonia negatively impacts on goats' productivity but its extent and magnitude among the local communities in Uganda remain unknown. A cross sectional study was conducted in the districts of Agago and Otuke neighboring Karamoja in Northern Uganda during the months of July and August 2011 to explore for the status of the disease. Methods: Five hundred and four serum samples from goats were obtained from randomly selected unvaccinated herds and 100 goats from vaccinated herds. Serum samples were examined for antibodies against Mycoplasma capricolum subsp. Capripneumoniae (Mccp) by ELISA method. A total of 162 semi-structured questionnaires were administered to selected farmers to obtain information on their understanding of the disease and the risk factors they associated with CCPP. Eight focus group discussions were also conducted with selected farmer groups to obtain detailed qualitative information on CCPP. Results: Among the unvaccinated goats, seroprevalence of CCPP was 32 (17.7%) and 52 (23.3%) for Agago and Otuke districts respectively. Levels of antibodies against Mccp were higher among vaccinated goats than unvaccinated ones (mean optical densities (ODs) of 0.905 and 0.776, p = 0.08). Majority of the farmers 121 (74.7%) had knowledge on CCPP and recognized that CCPP was among the major challenges to goat production in Uganda. Conclusions: This study demonstrated that CCPP was prevalent in Agago and Otuke districts, which are outside but close to Karamoja region where the disease was previously confirmed. There is a need for wider and detailed studies to investigate further CCPP in other districts of Uganda for effective preventive and control of CCPP in Uganda and the neighboring countries.

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Keywords

Contagious Caprine Pleuropneumonia, CCPP Seroprevalence, Uganda

1. Introduction

Contagious caprine pleuropneumonia (CCPP) is a highly infectious and devastating respiratory disease of goats caused by *Mycoplasma capricolum* subspecies capripneumoniae (Mccp). Clinically the diseased is characterized by coughing, respiratory distress and very high morbidity and mortality rates [1]. The disease is included in the list of notifiable diseases of World Organization for Animal Health as it threatens a significant number of goat populations throughout the world causing significant socioeconomic impact mainly in Africa and Asia.

Infection of susceptible goats is mainly through inhalation of contaminated aerosols from infected goats. Due to the high sensitivity of Mycoplasma to the external environment, close contact between infected and susceptible animals is essential for effective transmission of Mccp to take place [2], and overcrowding and confinement have been known to favors close contact and circulation of the Mycoplasma. Stress factors such as malnutrition and movement over long distances have been documented to enhance spread and morbidity of the disease [3] [4]. In Africa where extensive and traditional husbandry is practiced, pathogens have been reported to spread when animals meet at watering points and communal grazing areas.

In Africa where extensive and traditional husbandry is practiced, pathogens have been reported to spread when animals meet at watering points and grazing areas. Latent carriers such as goats or sheep that recovered from the infection without becoming bacteriological sterile were reported responsible for the perpetuation of the disease in herds [1] [5].

Clinically, the disease is characterized by high fever, respiratory distress and high mortality between 60% and 100% in absence of antibiotic intervention [1] [6] [7]. The pathological lesions are characteristic presenting interstitial fribinous pleuropneumonia, interlobular oedema and hepatization of the lung. This involvement is often unilateral, and the affected lungs are known to be port-wine coloured with possible total hepatization [1] [2]. There is involvement of the pleura with the pleural cavity containing straw-coloured exudate with fibrin flocculations [6] [8]. Contagious Caprine Pleuropneumonia (CCPP) is a disease of major economic importance in Asia and Africa, causing major constraint to goat production because of high mortalities [9] but the extent of the disease in Uganda is not well documented. In Uganda CCPP was previously confirmed in the Karamoja region [10], in the North-East of Uganda inhabited by nomadic pastoralists.

Here, we present the findings of a study carried out in two districts Agago and Otuke in Northern Uganda about CCPP and its predisposing factors in goat populations as well as the farmer's attitude about the disease. These districts are outside but close to Karamoja sub region where CCPP had been previously confirmed [10] and yet no documentation of CCPP status in the surrounding areas has been done. This preliminary finding would inform the current situation and provide information about the appropriate preventive and control measures to be instituted against CCPP in the country.

2. Materials and Methods

A cross sectional study was conducted in Agago and Otuke districts adjacent to Karamoja region in Northern Uganda to determine farmers' perception of CCPP and to determine the sero-prevalence of CCPP in these areas. To obtain information on CCPP and other goat diseases in the areas, a structured questionnaire was administered to randomly selected goat farmers and focused group discussions conducted with eight farmers groups using participatory epidemiological methods.

Indirect ELISA for CCPP

To determine the sero-prevalence of CCPP, antibodies against CCPP were measured for serum samples from 504 goats using an in-house indirect ELISA. The ELISA for detection of antibodies against Mccp was developed, optimized and performed in the Central Diagnostic Laboratory at the College of Veterinary Medicine, Animal resources and Biosecurity, Makerere University Kampala, Uganda.

Briefly: ELISA plate, (NUNC Immunoplate 439,454, Thermo Fisher Scientific, Roskilde, Denmark) were coated with 100 μ L per well of inactivated Mccp antigen (Caprivax, Kenya Veterinary Vaccines Production Institute, Nairobi, Kenya) at 1:100 dilution and incubated overnight in at 40 c. The plates were washed, 150 μ L per well of blocking buffer (2% bovine sarum albumin (BSA) in Phosphate buffer saline (PBS)) added and incubated at room temperature for one hour. Test serum samples were diluted at 1:100 in assay diluent (1% BSA + 0.1% Tween 20 in PBS) and added 100 μ L per well. Known positive sera (strong and weak positive) and negative sera were added on each plate. The plates were incubated at room temperature for one hour. Conjugate, protein G HRPO at 1:5000 dilutions was added 100 μ L per well and incubated at room temperature for one hour. ABTS substrate was added 100 μ L per well and incubated at room temperature for 10 minutes. The reaction was stopped after 30 minutes incubation by addition of 1% Sodium dodecylsulphate (VWR BDH) and the optical densities (ODs) read at 450 nm.

An arbitrary cutoff based on the negative control included on each plate was set at 3 standard deviations above the mean OD value of the negative control. The mean OD for the negative sample was 0.1133 and SD = 0.036; hence the cutoff was 0.241. An arbitrary categorization of the results was made as negative (OD < 0.25), trace (OD 0.25 < 0.5), weak positive (OD 0.5 < 1) or strong positive OD > 1). To compute the sero-prevalence, weak and strong positives were considered as positive while those with trace were considered with the negative.

The questionnaire data was entered using EpiDatasoft ware and later imported into stata (Version 9.1) for analysis. The ELISA reading data was automatically recorded into Microsoft excels 2007 and later imported into stata for analysis. The qualitative data from the questionnaire and focus group discussions were analysed thematically and a code sheet to show sequence in responses were developed accordingly. Descriptive statistics including percentages, chi-square test *p*-values are presented.

3. Results

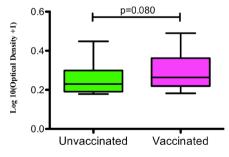
3.1. Sero-Prevalence of CCPP in the Districts of Agago and Otuke

Serum samples were obtained from 404 goats from herds with no history of CCPP vaccination and 100 goats vaccinated for CCPP by MAAIF in the last four months before sample collection. Of the unvaccinated goats sampled, 181 were from Agago district and 223 from Otuke district.

Antibody titers (in terms of ODs) were compared between vaccinated and unvaccinated goats. Of note was that although the antibodies among the vaccinated goats generally tended to be higher than unvaccinated goats this difference was not statistically significant, p = 0.08 (Figure 1). The sero-prevalence of CCPP was 20.8% and was not significantly different between Agago (17.7%) and Otuke (23.3%) districts (Chi square p = 0.165).

3.2. Farmers' Perceptions on Diseases of Goats and Knowledge of CCPP in Agago and Otuke

Questionnaires were administered to 162 individual goat farmers and focused group discussions were conducted with eight farmers' groups. Common goat diseases in the study area in order of importance as listed by the



Vaccination Status of the animals

Figure 1. Levels of antibodies to Mccp antigen compared between the unvaccinated and vaccinated cattle. Shown on the y-axis are the log 10 transformed optical densities. Shown is the Wilcoxon rank-sum (Mann-Whitney) test *p* value.

farmers included endoparasites (39.7%), CCPP (17.9%), mange (16.2%), orf (13.5%), tick infestations (12.7%) for Agago district. Similarly in Otuke districts endoparasites (34.2%), CCPP (25.5%), tick infestations (20.5%), mange (13.7) and orf (6.2%) were highlighted as the major health challenges in goat rearing. Similar patterns were equally obtained in the proportionate ranking of common goat diseases in the FGDs with the farmers' groups.

Of the 162 farmers who responded to the questionnaire, 121 (74.7%) were aware of CCPP and gave the local luo language name of CCPP as "two-oboodyel" or "loukoidyel" (Agago) or "two-iwukudyel" (Otuke). Of the 121 farmers who were aware of CCPP, 86 (82.6%) reported to have had the disease in their herds. They emphasized that the disease was recognized from clinical signs, which included cough/difficult breathing, fever, loss of appetite, nasal discharge, difficult movement and loss of body condition that often culminated into death of the animal. Further, the farmer groups were also able to point out seasonal pattern of CCPP during the participatory focused group discussions. It was apparent that CCPP is understood to be more prevalent in the months of July to August of the annual calendar.

Of the respondent farmers, 86 (82.6%) who reported to have encountered CCPP in their herds, 49 (57%) sought for professional veterinary care for the affected goats (**Table 1**). Surprisingly, 25 (29.1%) of the respondents reported that they never took any particular action in the care of their sick goats while 12 (14.0%) reported to have only applied local herbs as a form of treatment.

It was also noted during the discussion that farmers pointed out herd sizes, farming practices and source of breeding stock as risk factors for occurrence of CCPP. Of the 86 farmers who reported to have encountered CCPP, 74 (86.1%) attributed the disease to rearing small herd sizes managed by communal grazing. Most of the farmers (65 (75.6%)) also pointed out that practicing communal grazing with seldom tethering is likely to increase CCPP infections and spread When farmers were asked on the risk of CCPP infections from breeding stocks, 55 (64%) of the farmers were aware of the risk of introducing the infection and preferred to acquire breeding stocks from far off districts they considered free of the disease.

4. Discussions

The main aim of this survey was to explore the occurrence of CCPP beyond the boarders of Karamoja region where the disease had been previously confirmed [10]. This study confirmed that indeed CCPP negatively impact on goat health and production in Agago and Otuke but its extent and magnitude among local communities remains to be fully elucidated. The ranking of CCPP by farmers in Agago and Otuke as second among the health challenges of their goats corroborates with Southern Ethiopia participatory investigation of CCPP where farmers ranked the disease major constraint in goat production [11]. The clinical signs and postmortem lesions identified by farmers in Agago and Otuke were closely related to earlier documented clinical signs of the disease [2] [12].

The seasonal pattern of CCPP identified by farmers during onset of rains agreed with previous reports from Southern Ethiopia [11]. This also affirmed previous reports that seasonal calendars were important tool in designing disease control programs [13] [14] and this could be equally targeted as best time for control of CCPP in Agago and Otuke districts.

The farmers' attitude towards treatment of CCPP in Agago and Otuke was largely related to use of modern veterinary drugs than use of local herbs even though a few used both. This observation is of interest and contrary to previous reports that purely pastoral communities depend highly on local materials mainly plants to manage livestock health problems as first line of treatment [15] [16]. However, of note was that up to 29.1% of the farmers did not take any action against CCPP infections in their herds and this could probably signify the low value some farmers still attached to small ruminant.

Table 1. Farmers' approach to management of cases of Contageous Caprine Pleuropneumonia.

District	Sought veterinary service and applied veterinary drugs	Use of local herbs	Sought veterinary help and also applied local herbs	No action taken
Agago $(n = 45)$	19 (42.2%)	8 (17.8%)	2 (4.4%)	16 (35.6%)
Otuke $(n = 41)$	22 (53.7%)	4 (9.8%)	6 (14.6%)	9 (22.0%)
Overall	41 (47.7%)	12 (13.9%)	8 (9.3%)	25 (29.1%)

All the goat husbandry practices in Agago and Otuke were still traditional irrespective of herd sizes reared by farmers, source of breeding stock and farming practices. Such traditional husbandry practices were previously reported to favor spread of CCPP when animals meet at watering points and grazing areas because of increased contact rates between infected goats and naïve ones essential for effective transmission of Mccp [2]-[4].

The present study estimated the overall sero-prevelance of CCPP at 20.8%. This is close to what has been reported (15% - 18%) in southern Ethiopia [4] [11]. Our finding was at variance with reports of studies done in Ethiopia [17] [18] and in Beetal goats in Pakistan Hussain, 2012 #51 as well as in East Turkey [19] who documented higher sero-prevalence between 31% and 38%. The sero-prevalence of the disease was not significantly different between the two districts, Agago (17.7%) and Otuke (23.3%). This is not surprising since Agago and Otuke lie within the same agro ecological zone, a similar explanation earlier reported in Ethiopia [18].

Important to note was that comparison of antibody levels (ODs) among the vaccinated goats were slightly higher than unvaccinated but the difference still insignificant (p = 0.08). We expected a significant difference in the antibody levels between two. This may suggest that vaccination may not be provoking sufficient sero-conversion thus putting the efficacy of this vaccine into question. Never the less, this findings were in disagreement with high efficacy reported with inactivated Mycoplasma strain F38 saponin vaccine in natural infection with CCPP [20]-[22] and experimentally lyophilized killed F38 vaccine that conferred 100% protection against mortality and 95% clinical disease of Mycoplasma species strains F38 [23]. Seroprevalence done in Tanzania [24] and in Ethiopia [4] [18] have also shown no sex difference in CCPP epidemiology.

5. Conclusions

The preliminary questionnaire survey, focus group discussions and the serological findings strongly demonstrated that CCPP is prevalent in Agago and Otukedistricts, which are outside but close to Karamoja region where the disease was previously confirmed. This study also showed most farmers to be aware of CCPP and the use of participatory disease investigations approaches which could be of relevancy in designing and timing CCPP control programs in these districts.

The study covered only Agago and Otuke; therefore, more detailed and bigger studies should be undertaken to investigate further CCPP in rest of the districts in Uganda to pave way for effective preventive and control measures against CCPP in the country. Secondly, there is a need to develop a diagnostic test which will be easy to use and readily available in Uganda.

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Factors Affecting Technology Adoption in Small Community Farmers in Relation to Reproductive Events in Tropical Cattle Raised under Dual Purpose Systems

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Abstract

The impact of technology adoption among farmers in developing countries has been an issue difficult to measure and interpret. Hence, research to improve production has been centered in technology that possible is not adequate for this region. The present review aims to utilize subjects such as Genetics, Reproduction, Nutrition, Management and Animal Health as examples of research endeavors illustrating the scarcely bond existing between research and adoption of technology. A final section is an attempt to illustrate the possible reasons why the transfer of technology is failing to be of any consequence at least under the conditions of developing countries.

Keywords

Tropics, Artificial Insemination, Embryo Transfer, Nutritional Supplementation

1. Introduction

The production of beef and milk in the majority of countries in the tropics has been transformed through time into more intensive systems with a high concentration of animals in smaller areas. However, in spite of this tendency, the traditional systems for dual purpose continue to be the principal subsistence for numerous families

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even though they lack infrastructure and sound production. Nonetheless, these systems yield about 41% of the total milk and 50% of beef for the area [1].

There are numerous and diverse systems of production in the tropics but a common bond in all of them is the purchasing power and economic ability of the producer to invest in the enterprise. Based on these premises, researchers have arbitrarily divided farmers in subsistence, medium income and those with sound budgets. The shortcomings and virtues of these divisions will be discussed. Adoption of technology has to be closely connected to the economic stability of the farmer. Success in the enterprises with limited budgets was related in the past to government or international initiatives based on subsidies, when this support ensued, farmers were left with scarce possibilities to implement the recommended strategies.

The present review aims to highlight some productive events in the farm as examples of past and current technology and the pitfalls to absorb and implement these techniques, and moreover, to illustrate the little impact of experimental research in tropical animal production.

2. Genetic Programs

Producers have based their efforts on an indiscriminative crossing of diverse breeds, ending up with animals characterized without a defined phenotype or production performance. These cattle are hardy, resistant to the environment but with a limited ability to produce either beef or milk [2]. Other researchers, particularly in Latin America have attempted to reincorporate the native breeds brought by the Spaniards before the XIX century, to improve production to the already existing systems. There are quite a number of papers concerning the rescue of native breeds (either *Bos taurus* or *Bos indicus*). However, generally based on the numbers of animals of any particular breed, it seems to be that this technology has not been properly adopted in the tropics.

Innumerable efforts have been vested to import specialized European breeds such as the Holstein Friesian, Jersey or Brown Swiss to the area, more failures than success stories have been accounted for in the literature, probably due to the low adaptation of these animals to the harsh environmental conditions in the tropics [3].

Finally, in the 90's considerable interest was given to a more selective program based on the crossbreeding of European breeds with *Bos indicus* types, in effect, assays comparing purebred animals with F1 or other levels of crossing have concluded that the F1 animal is by far the best choice for meat or milk production [4]. Thus, the hybrid vigor product of crossing two purebreds proved to be an incentive for farmers [5]. Even more, researchers experimented alternative crossing of two breeds to ended up in the production of synthetic breeds (the majority 5/8 of one bred and 3/8 of the other) such as the Cuban Syboney, Australian Milking Zebu or Jamaican Hope to cite a few. It is beyond the scope of this review to describe the advantages and disadvantages of the different combinations experimented in the 90's. However, it gives the impression to be an agreement that these systems are not applicable in small community enterprises as the volume of animals is limited; hence it is not feasible to implement an alternative crossbreeding program. To conclude, evidence points out that if the animal has more than 75% of European genes, the adaptation to the local environment is hazardous; on the other hand if the animal possesses 75% of zebu genes, then expression of production indicators are very limited.

Recently [6], evaluated the feasibility of using embryo transfer in F1 crossbred animals (Holstein × Indobrasil) placing an F1 embryo in the undefined animal hence avoiding long periods of crossbreeding and favoring the hybrid vigor enhancing the production of either beef or milk. The economic calculations of these authors for the moment make this approach not a valuable strategy in small community farmers [7] [8]. This is probably why most of the reviews on embryo transfer are of experiences obtained in the use of the technique in *Bos taurus* animals [9]. Nonetheless, recent evidence from Brazil has shown promising results in *Bos indicus* cattle using embryo transfer in large herds [10].

3. Reproduction

Traditionally, farmers have used natural mating (NM) as the most advantageous procedure to reproduce their animals. Unfortunately, the genetic progress encountered through this system is minimal and for this reason, government policies have been geared towards using artificial insemination (AI) as a vehicle to introduce new and improved breeds in the tropics [6]. The adoption of this technology is somewhat dissimilar. Progressive farmers have found that the system can be economically sound if investment in training of personnel directly involved in the implementation of the technique is applied. This investment is geared towards solving problems

such as inefficient estrus detection and poor fertility following AI. With the advent of pharmacological procedures to manipulate the estrous cycle [11] it is possible to a certain degree to eliminate the problem of estrus detection [12]. The economic feasibility of these procedures remain to be tested as the physiological state of the animals (anestric, with a calf suckling or poor nutrition) affects the capacity of producers to implement AI [13]. In effect Alonso *et al.*, [14] found in an experiment where cows were AI by appointment but observed continuously for 100 h following synchronization that 85% of the animals displaying estrus ovulated, contrasting with only 63% that did not. Fertility in the whole herd was 25%; however, 86% of these pregnancies were detected in animals that displayed estrus.

In essence, it is relatively easier to have the bull running with the cows at all times [15] hence reducing the human error consequence of poor estrous detection efficacy and precision, or mistakes in the technique itself (poor implementation of thawing procedures, inaccurate placing of the semen in the body of the uterus and inefficient storage in the liquid nitrogen container to mentioned just a few [16]. How much extension services would recommend the use of AI among small producers is a debatable issue that deserves further investigation. For example, there are several procedures to facilitate the milk let down of the cows, starting from the suckling of the calf and a prompt removal after milk flow, or allowing the calf to suckle one teat after milking or giving the feed to the calf from a pool of residual milk [17] [18]. What effect those any of these systems bear on the onset of ovarian activity?? How these methods affect the performance of the calf?? All these interventions deserve further research to recommend one over the other.

4. Feeding

This procedure is possibly the major hurdle affecting production in the tropics. As most systems are based on grazing as the main element for feeding, farmers face two very different scenarios for adopting an adequate procedure. During the dry season, fodder is scarce and of poor quality and in some countries, actually quite dramatic, especially nowadays with the constant climatic changes due to global warming. In contrast, during the rainy season, pasture seems to be available in quantity but pending on the conditions, probably with large concentrations of water. In spite of the fact that these events are well known, surprisingly there are very few sound strategies recommending the use of particular techniques to overcome these difficulties. In general it can be said that farmers should think that there is going to be little rain during the season so they should prevent for the possible draught. Unfortunately this is easier to recommend, not so to implement. Feeding procedures are certainly quite different between farmers with poor as opposed to sound budgets for investment. In general this is reflected in cow performance, particularly milk production. Animals properly fed, usually have productions above 8 lt per day; this is obviously closely related to the ability of the breed to adapt to the climatic conditions in the area and their capacity to dissipate heat [19]. Seasonal production of milk poses a serious problem in the commercialization of the product. At the moment, in the majority of tropical countries there is abundant production of milk in the rainy season (following a peak of calvings in the spring) and very little milk at the end of the rainy season. However, this statement deserves a qualification as there are specialized units in the tropics capable of producing milk the whole year around. Feeding strategies are mostly aimed to utilize local resources, the days of importing feedstuffs from more temperate areas (mostly grains with high energy and protein content) are likely over. A challenge to feed a balanced diet with scarce fodders rich in protein is a quest that specialists should have in their forefront of their research.

For example, Kamanzi, and Mapiye [20] in a study in Rwanda considered that land scarcity was ranked as the most important cause of feed shortage followed by inadequacy of forage planting material and lack of knowledge on forage production and utilization. To ensure sustainable viability of smallholder dairying in densely populated highlands, the following recommendations have been put forward: a) screening and evaluation of high-yielding and easily propagated pastures, b) incorporation of forages into cropping systems, c) value addition of low quality roughages and d) training farmers on forage production and utilization. The amount of milk produced will depend largely on two issues, the percentage of European genes that the animals have and the feeding regime imposed. However, it seems to be evidence in the literature [21] that the quantity so much as the quality of the food, determines milk production. If this statement is correct, would it be possible to implement a diet based on the ability of certain bacteria to synthesized fiber? Some studies [22] suggested that this is possible. The question still remains, are the animals existing today capable of a substantial improvement in milk production if the quality, quantity or both can be manipulated with local products???

5. Management

An interesting study was published [23], in dual crossbred cattle specialized in milk production, who found that if the animal had comfortable accommodation the key for calving was comfort, hence the spread of this event was in the cooler winter months. In contrast, cattle raised under primitive installations and poor management, the calving season was mostly concentrated in the summer when fodder was more abundant. Management plays a very important role in the fate of a production unit in the tropics; generally accommodations are rustic, fairly basic and old. Farmers have to deal with these types of facilities and this is not a small feat. Unfortunately, the investment required for improving their farms is quite considerable and not permissible under the prices currently found in the market for animal products. There is a need for a more serious and long term program from governments in developing countries to overcome this issue. The lack of an effort will translate in more migration from the countryside to the urban areas. The average age of a farmer in certain tropical areas is 55 years [24]. How long these systems will be sustainable if there is lack of incorporation of younger farmers?

For example, Hostiou et al., [25] found that most of the tasks (milking, grazing management) are performed manually. A study, based on the quantification of working time shows that most of the activities are carried out exclusively by family manpower; at times, helped by wage earners, volunteer workers and farmers' mutual aid. The study reveals the high variability of routine work (from 913 to 3955 hours per year for milk production) and seasonal work (from 17 to 328 days per season). Also, Lopez-Pereira [26] stated that the continuing soil erosion and land degradation in these low-input staple crop production hillside farming systems lead to be pessimistic about increasing the agricultural incomes of these farmers. However, this study shows that the appropriate combination of improved technologies and agricultural policy or alternative production diversification strategies, can advance the incomes of small-scale hillside farmers in by over 50%. The technology components considered are stone walls and ditches combined with living tree barriers to prevent erosion of the hillsides, and a package of improved sorghum seed, and modest doses of nitrogenous fertilizer. The study concluded that erosion-control devices and yield-increasing crop varieties and fertilizers are effective technologies for the erosion-prone hillside landholdings found in many areas of the tropics. A similar strategy was undertaken years later [27] observing that by only using trees as shade from the blazing sun would improve the forage capacity of the pasture by preventing early desiccation and dryness of the fodder. Murgueitio et al., [28] proposes that extensive use of the land deeply rooted in the culture and rural economy of the region, requires an urgent transformation if it is to become both more efficient and environmentally friendly. Silvopastoral systems incorporating native trees and shrubs are instrumental for the productive rehabilitation of cattle production and for biodiversity conservation in agricultural landscapes

6. Animal Health

The welfare of the cow and the calf soon after calving is an event where little information has been afforded by researchers in the area. This is probably the consequence of a sequel of normal events during this stage. In effect, diseases such as retained placenta, hypocalcaemia or ketosis are rarely reported and it seems that calf survival is the major issue soon after calving. The majority of cows tend to calve outdoors hence predators are important. Health issues seem to be concentrated in the control of parasites both external and internal which undoubtedly affects animal performance. For example an interesting study was performed by reference [4], where different combinations of crossbred animals ($Bos\ taurus \times Bos\ indicus$) were compared. The best performance and disease resistant was by far the F1 crossbred animal.

7. Adoption of Technology

Tey and Brindal [29], in a review concurred that adoption of technology is driven by factors such as, 1) socio-economic, 2) agro-ecological, 3) institutional, 4) information, 5) farmer perception, 6) behavioral and 7) technological issues. Some of these relatively "fixed" factors are useful for market segmentation and targeting purposes. Other "modifiable" factors can be reshaped through interventions. On the other hand, reference [30] found significant correlation coefficients between and within farm household characteristics and technology adoption. For example, male-headed households adopted significantly a higher number of technologies than female-headed households. Technology adoption rates increased significantly with the stage of education and family size and decreasing distance from market or trade centers. The level of technology adoption by small-

holder farmers is still unsatisfactory and is highly dependent on gender, family size, location and level of education [31]. The authors concluded that the geographical site where the farms were situated, the size of the farm and the level of education of the farmers were very important in the success of an enterprise. However, the frequency of the visits of the farm owners and technicians alike, were also major players to bring stability to the farm.

Bellows *et al.*, [32] reported that the adoption of land and agrochemical-intensive methods by resource-poor farmers cultivating steep lands, resulted in decreased environmental and economic sustainability. Farmers with adequate resources were able to maintain viability by transferring land out of beans and into other commodities, particularly cattle. However, this shift in resource use affected social equity by decreasing farm labor opportunities for smallholders and landless farmers and diminishing land available for tenants. This study indicate that the impact of technology introduction on farming system sustainability can be assessed effectively by conducting integrated socioeconomic and agronomic analyses across farms representing various in land-use practices and intensities.

All these factors reported above are all important in decision making in the farm, but also in the researchers interpretation involving the magnitude of each factor in their recommendations to improve animal production in the tropics. The failure of many international and national programs has been the consequence of their inability to understand the local conditions thus applying a suitable program basically tailor made for the region in question. In fact, researchers in emerging countries should focus their efforts and resources in developing technologies based on the local conditions and needs of people, instead of investing them in studies which application is often expensive and unpractical.

In general there is agreement that there are mainly four different systems from transferring information to the farmer. Probably the most ancient one is related to the data generated in experimental stations or universities and transmitted to farmers in a variety of systems, either by technical bulletins, field days or conferences given by technicians in forums directed to farmers. The most obvious system, extension services to the farmer, is practically nonexistent in developing countries for lack of a budget to support this important department of diffusion of knowledge generated by the researchers in the experimental station. Moreover, the results obtained in the experimental stations are hardly reproduced by the local farmer for many reasons but probably the most important is the difficulty in transferring data from a controlled environment (such as the one in an experimental condition) to the farmers working with different infrastructure, economy and labor. The second in importance and gaining popularity in recent years, is the diffusion of data generated in the farm itself either by experiments carried out in situ, or by retrieving information that the farmer has documented in one way or another. This method has created the need for a more active institutional input to diffuse this knowledge. There are however, successful examples of this method. In Costa Rica a program started about 15 years ago to digitalize their information using software that could be implemented by the majority of farmers hence facilitating the diffusion of productive parameters via the network or having field days among the participating farmers. Similar examples are becoming more frequent in different parts of the developing world. The third system or method of transferring technology has been the support or government or international organizations in defined programs to improve the conditions of farmers' enterprises. Experience has shown that this type of effort is successful as long as the subsidy is present, once the program ended; farmers find it hard to accept that there is a valid need to invest in their farms. This is particularly so in the long term programs where results are not immediate and visible to the farmer.

8. Conclusion

Finally, a relatively untested system is the diffusion of information between farmers themselves, either by informal chatting or visits to other farms to learn something that they could implement in their farms. This system of diffusion of technology should be accurately measured to understand their dimensions.

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