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BULLETIN OF ANIMAL HEALTH AND PRODUCTION IN AFRICA

BULLETIN DE LA SANTE ET DE LA PRODUCTION ANIMALES EN AFRIQUE

OAU/STRC
OUA/CSTR

March 1990
Mars

Vol. XXXVIII No. 1

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BUREAU INTERAFRICAIN DES RESSOURCES ANIMALES

P.O. Box 30786, NAIROBI, KENYA

BULLETIN

March
Mars 1990 VOLUME 38 No. 1

ORGANISATION OF AFRICAN UNITY
ORGANISATION DE L'UNITE AFRICAINE

COMMISSION SCIENTIFIQUE TECHNIQUE ET DE LA RECHERCHE

SCIENTIFIC, TECHNICAL AND RESEARCH COMMISSION

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BULLETIN OF ANIMAL HEALTH AND PRODUCTION IN AFRICA

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I. ORIGINAL ARTICLES

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 1-4

SEASONAL FAECAL EGG COUNT CHANGES IN KENYAN GOATS THROUGH A ONE-YEAR PERIOD

L.W. WAMAE and M.A.K. IHIGA
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CHANGEMENTS SAISONNIERS DU DENOMBREMENT D'OEUFS DANS LES FECES DES CHEVRES KENYANES PENDANT UNE ANNEE

Résumé

La numération d'oeufs dans les fèces de 30 chèvres kenyanes a été relevée pendant une année. Les variations saisonnières du dénombrement d'oeufs sont discutées en rapport avec la pluviosité et la température. Les espèces de ver qui vivent en parasite chez ces chèvres furent déterminées par la méthode de culture fécale et les caractéristiques de l'oeuf. Il s'agit des vers ci-après: *Haemonchus, Oesophagostomum, Trichostrongylus, Cooperia, Nematodirus, Strongyloides* et *Moniezia*. Il a été établi que, pour les strongyles, le dénombrement d'oeufs dans les fèces a atteint le maximum pendant la saison chaude, tandis que pour les *Strongyloides*, on a obtenu le maximum pendant la saison humide et chaude. Il a été donc proposé que le déparasitage soit effectué juste avant l'arrivée des pluies afin de réduire la contamination des pâturages pendant la saison pluvieuse.

Summary

Faecal egg counts of 30 Kenyan goats were recorded over a one-year period. Seasonal fluctuations of the eggs are discussed with reference to rainfall and temperature. The worm species parasitizing these goats were determined by the faecal culture method as well as by egg characteristics as *Haemonchus, Oesophagostomum, Trichostrongylus, Cooperia, Nematodirus, Strongyloides* and *Moniezia*. It was established that faecal egg counts for strongyles peaked during the dry months while those of *Strongyloides* peaked during the wet and warm weather. It is therefore suggested that deworming should be done just before the rains in order to reduce pasture contamination during wet weather.

INTRODUCTION

Helminthiasis in goats is caused by many genera of worms. The infective larvae of these worms are picked up individually from the environment. These worms usually have a free-living (non-parasitic) as well as a parasitic phase. The nematodes which are the subject of this paper usually contaminate the environment when their eggs are passed in faecal matter of infected animals. These eggs develop, under suitable conditions, into L_1 through to L_3 which is infective. These nematodes cause a parasitic gastro-enteritis which results in unthriftiness, various degrees of diarrhoea and death in over-whelming infections.

Seasonal changes in faecal egg counts have been documented before by many workers[1,2,3,4,5]. It has already been established that these changes occur in different animals at the same time of the year[1,2]. So, it is in order to assume that the said changes would also occur in goats although in the tropics the times of the year when these changes occur will be different from those in the temperate areas.

Materials and Methods

Thirty adult goats selected randomly from a group of 100 goats, recently brought to the National Veterinary Research Centre (Muguga) from Machakos (Eastern Kenya) were used in this exercise. The whole group was ini-

tially confined in isolation as a measure of disease control in the station. During this period, one goat died and on autopsy was reported to have died of 'worms' during the month of October, 1986.

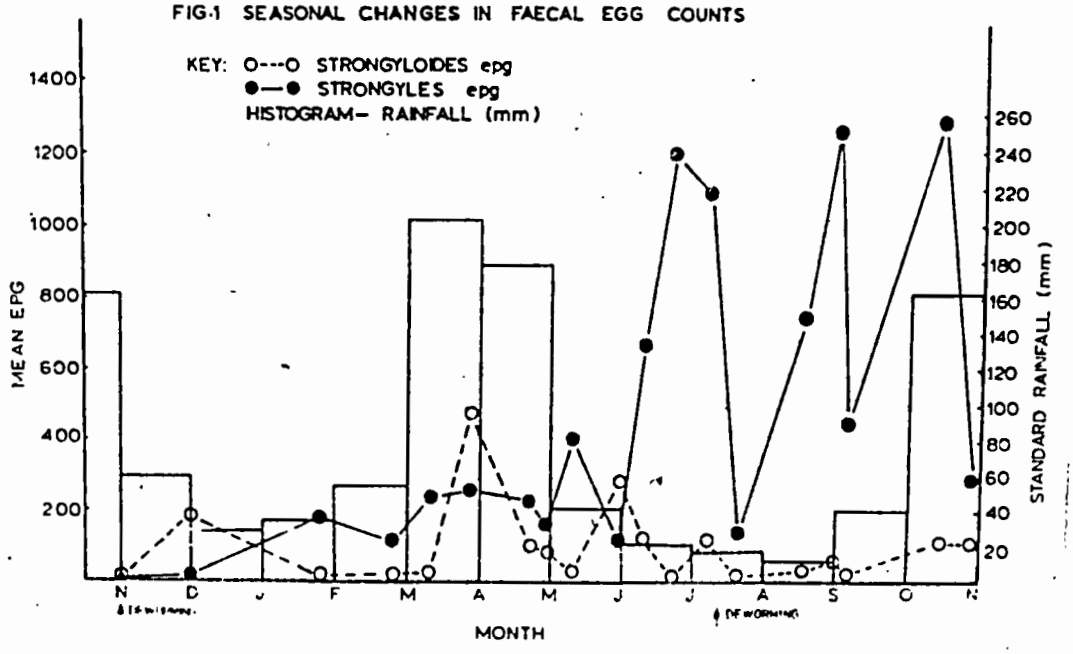
On the 12th of November, 1986 all the goats were dosed with Febantel (Rintal(R) Bayer) at the recommended dose rates. Faecal samples taken 9 days after this were devoid of any eggs. Consequently the goats improved in bodily condition and seemed in much better health. Sampling was then continued henceforth in order to monitor reinfection. Faecal samples were subjected to the direct Macmaster technique for estimation of eggs per gram (e.p.g). The eggs could then be differentiated into those of strongyles, *Strongyloides* and *Moniezia*. Oocysts were also observed. Attempts were also made to identify the worms parasitizing the goats by the faecal culture method[6] as well as by egg characteristics. This exercise was carried out once a month from November to March when sampling every fortnight was then begun in order to closely monitor egg count changes.

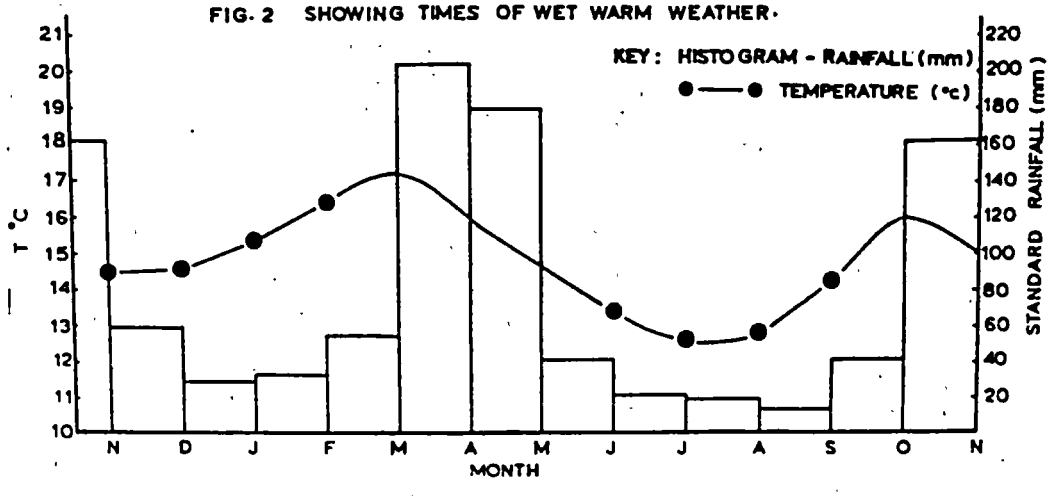
The goats were grazed freely on various paddocks in the Centre and housed for the night in stalls with cement floor overlaid with hay beddings. The goats probably superimposed their pasture contamination on the already existing one.

Results

On commencement of this experiment, the goats had high mean faecal egg counts of strongyles (over 1700 e.p.g.) and *Strongyloides* (over 2900 e.p.g.). During the course of this work, *Nematodirus* eggs were also encountered. As shown in Figure 1, anthelmintic treatment in November (Rintal(R), Bayer) brought egg counts down to nil. However, worm build-up started in the dry month of December. Sampling was not done in January due to unavoidable circumstances. However, by February the rise in egg counts was apparent although it was more modest for *Strongyloides*. During the rainy months of April and May there was only a mild rise in egg counts and *Strongyloides* eggs were more than those of strongyles. However, as the rains receded in the months of June, July and the dry spell eventually got under way, strongyle egg counts shot up while those of *Strongyloides* were low and remained thus throughout the dry spell with a slight rise in November when again the rains

FIG·1 SEASONAL CHANGES IN FAECAL EGG COUNTS





were considerable. With the coming of the rains in November strongyle counts started to drop towards the end of this month. Temperatures were somewhat high during the rains and this presented a wet, warm climate during the rainy months, Figure 2. Coccidia oocysts were also encountered in these goats but no attempt was made to identify or count them.

The worms in these goats were identified either by faecal culture or characteristic eggs, as *Haemonchus Oesophagostomum, Trichostrongylus, Cooperia, Nematodirus, Strongyloides* and *Moniezia.* At no time were *Fasciola* spp. or *Paramphistome* spp. eggs encountered either by the sedimentation method or the Zinc Sulfate flotation method.

Discussion

These results point to the fact that the goats passed large numbers of strongyle eggs during the relatively dry months rather than during wet ones. This fact has been recorded previously in ewes[7,8] and goats[9]. It is during the dry months that larvae per unit area of pasture are concentrated due to scarcity of herbage and therefore a single goat will pick a lot of larvae as it grazes on a bigger surface area. This situation is reflected in the results of this study. However, the situation is reversed during the wet season when a dilution factor is in operation due to the lush growth of herbage. Additionally, rain carries larvae into the ground. It is interesting to note that faecal egg counts of *Strongyloides* were somewhat low when strongyle egg counts were high and did not peak simultaneously. This trend was specially so during the wet, warm months. It has been suggested[10] that self-cure mechanisms may limit egg deposition during wet seasons and if this is the case, it only appears true as far as strongyles are concerned in this study.

Perhaps one of the reasons for this peaking of *Strongyloides* infections is reflected by their very life cycle. It is known that during warm and wet seasons, the free-living adults on the ground multiply much faster and therefore produce large numbers of infective L_3 larvae (heterogonic cycle). These, after infection, then mature in about 9 days and so start laying eggs. This is in contrast to the homogonic cycle which occurs when environmental condition are adverse[11] and a non-parasitic life cycle is then operational.

It has been suggested that increases in egg counts are primarily due to an increase in worm burdens rather than an increase in the egg laying capacity of worms[5]. The role of inhibited larvae has been shown to be of little importance in Haemonchosis in Kenya where only about 13% inhibition was described[10]. However, the role of this phenomenon in

Kenya as regards other worm species encountered in this study is not clear.

This study therefore demonstrates and supports other workers' findings that faecal egg counts are highest during the dry weather rather than the wet weather. It is, therefore, the time when the rate of pasture contamination is also highest and dosing of livestock should be done at this time just before the rains. This would get rid of worm burdens and so reduce amount of contamination to further augment the dilution factor on pastures during rainy seasons and so keep them with a low level of infective larvae.

Acknowledgement

We are thankful to Dr. Litamoi of Bacteriology Division for supplying us with the goats. The authors are also grateful to Mr. John Sangura and the Helminthology Division staff who helped in collection and examination of the samples. We wish to thank Mr. Muga, for providing meteorological data. This paper was published with the permission of the Director, National Veterinary Research Centre, Muguga.

References

1. Zawadosky, M.M. and Zvjaguintzenvisin (1933). *J. Parasitol.*
2. Taylor, E.L. (1935) *J. Parasit.* **21**:175.
3. Truner, H.W. and Moon, J.G. (1928) *J. Amer. Vet. Med. Ass.* **74**:199.
4. Schmid, F. (1933). *Zent fur. Bakt. etc.* **130**:338.
5. Morgan, D.O.; Parnell, I.W. and Rayski, C. (1951). *J. Helm.* **24**: (3) 101.
6. Ministry of Agriculture, Fisheries and Food (1971). Manual of Veterinary Parasitological Laboratory Techniques. Technical Bull. No. 18, London pg. 14.
7. Allonby, E.W. (1981). Cited by Uluund (1984) as personal communication.
8. Ulvund, M.I.; Maine, E.; Marano, J., Bhogal, M.S. (1984) *Kenya Vet.* **8** (1):9.
9. Asanji, M.F. and Williams, M.O. (1987) *Bull. Anim. Hlth. Prod. Afr.* **35**:308.
10. Allonby, E.W. (1974) Studies on Haemonchus contortus infection in Merino sheep, Ph.D. thesis, University of Glasgow, J.S. Heron Ltd.
11. Soulsby, E.J.L. (1968) Helminths, Arthropods and Protozoa of domesticated A animals 6th ed. London, Tindal, Bailliers and Cassell, p.178.

Received for publication on 23rd February 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 5-6

INCIDENCE OF *FASCIOLA GIGANTICA* INTRAMOLLUSCAN STAGES IN *LYMNAEA NATALENSIS*, THE INTERMEDIATE HOST, OVER A ONE-YEAR PERIOD INKENYA

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INTRODUCTION

Liverfluke disease in cattle in Kenya is transmitted by the snail intermediate host *Lymnaea natalensis*. Few records in literature indicate incidence of *F. gigantica* in *Lymnaea natalensis*[1]. It has been documented in Kenya that cattle fascioliasis is limited to defined endemic areas where rainfall is suitable for its propagation,[2], and some parts of the country have much higher incidences than others.

Work done in other parts of the world indicate that incidence of liverflukes in cattle vary from one season to the next[3,4,5].

This work was designed to study monthly incidences of *F. gigantica* intramolluscan stages in *Lymnaea natalensis* from an area in Kenya where bovine fascioliasis had been previously established as occurring in cattle[6].

Lymnaea natalensis snails were collected each month from endemic sites at Ruthagati and Kianjogu dams in Nyeri district and taken to the Veterinary Investigation Laboratory in Karatina. The snails were dissected out to check for any intramolluscan stages which were then differentiated into either liverfluke or non-liverfluke[7]. Additionally the snails were also checked for *Chaetogaster limnei* which live in snail shells as commensals.

A total of 414 snails were collected and dissected out and results are shown in Table 1. Out of these a total of 130 (31.4%) had liverfluke intramolluscan stages (mostly rediae) while 252 (60.9%) harboured *Chaetogaster limnei*. Fork-tailed cercariae were encountered in some snails which were not infected with *Fasciola* developmental stages while, in others single tailed non-encysting cercariae were seen. No snails were found infected with more than one cercarial type during this exercise.

Although highest incidences of *Fasciola* infected snails (over 40%) were encountered during the months of March, May, June, and July infected snails were present most of the year (Table 1).

The observation that infected snails were present most of the year is important epidemiologically in that susceptible hosts remain at risk of being infected throughout most of the year as has been reported before in Kenya[8]. Most cattle sampled from this area at different times of the year were found to be infected with liverfluke. It was also observed that many cattle are brought to this communal area to graze and drink and that cattle populations are higher during the dry season. These factors therefore produce almost perfect conditions for disease transmission. Metacercariae were also observed on vegetation in these areas and so were readily available to the cattle grazing there.

This vicious circle gets worse during the rainy season when snail populations increase rapidly. At this time a lot of fresh water is available and so suitable snail habitats are formed. It appears that with fresh water, snails are stimulated to lay a lot of eggs and also attain short generation intervals.

Findings in this study therefore reflect those of other workers[1] who have found that snails infested with rediae increase with rainfall while those shedding mature cercariae decrease with rainfall.

As mentioned above some fork-tailed cercariae were also seen as well as single-tailed non-encysting cercariae. The single-tailed non-encysting cercariae were identified in the laboratory as belonging to the family *Plagiorchiidae* which are intestinal parasites of vertebrates while the fork-tailed ones are those of the family *Strigeidae* and *Diplostomatidae* which are intestinal parasites of birds and mammals[7].

Table 1: Showing incidences of various parasites in *Lymnaea natalensis*

	Number of Snails dissected	Fasciola rediae/ cercariae	Chaetogaster limnei	Fork-tailed cercariae	Non-encysting single-tailed cercariae
February 1987	117	13	62	7	0
March	59	27	47	5	3
April	18	2	16	1	7
May	54	24	44	11	7
June	56	24	51	0	0
July	62	26	23	0	15
August	–	–	–	–	–
September	34	12	8	0	6
October	6	1	0	0	1
November	4	1	1	0	1
December	3	0	0	0	0
January 1988	0	0	0	0	0
February 1988	1	0	0	0	0
TOTAL	414	130(31.4%)	352 (60.9%)	24 (5.8%)	40 (9.7%)

Also seen were *Chaetogaster limnei* which live in the mantle cavity of the snails. It was noted that rise in number of snails infested with this commensal resulted in a decline in developmental stages of trematodes. This phenomenon has been reported before and attributed to these commensals swallowing miracidia and therefore interfering with their host finding capability[9].

Acknowledgements

We wish to thank Dr. Tangus and staff of VIL Karatina for putting their laboratory facilities at our disposal as well as Mr. John Sangura for technical help.

This paper was published with permission from the Director, National Veterinary Research Centre, Muguga.

References

1. Ogambo-Ongoma, A.H. (1979) 4th *Intl. cong. Parasitol.* Warszawa, Section C.
2. Cheruiyot, H.K. (1983) *Bull. Anim. Hlth. Prod. Afr.* **31 (4)**:364.
3. Ollerenshaw, C.B. (1959) *Vet. Rec.* **71**:957.
4. Ross, J.G. (1967) *Vet. Rec.* **80**:214.
5. Ross, J.G. (1967) *Vet. Rec.* **80**:368.
6. Cheruiyot, H.K. and Wamae, L.W. (1988) *Bull. Anim. Hlth. Prod. Afr.* **36**: (1):55.
7. Frandsen, F. and Christensen, N.O. (1984) *Act. Trop.* **41**:181.
8. Preston, J.M. and Castelino, J.B. (1977) *Br. Vet. J.* **133**:600.
9. Khalil, L.F. (1961) *J. Helminth.* **35**:269.

Received for publication on 14th April 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 7-10

AN ENZYME-LINKED IMMUNOSORBENT ASSAY (ELISA) FOR LIVESTOCK HYDATIDOSIS BASED ON A PARTIALLY PURIFIED THERMO-STABLE ANTIGEN

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TITRAGE AVEC IMMUNOADSORBANT LIE A UNE ENZYME BASE SUR UN ANTIGENE THERMOSTABLE EN PARTIE PURIFIE POUR LA DETECTION DE L'HYDATIDOSE CHEZ LE BETAIL

Résumé

Au total, 227 sérums de bétail ont été examinés. Ces sérums étaient répartis comme suit: 68 sérums de bovins (dont 35 de bovins qui avaient des kystes hydatiques lors de l'examen post-mortem et 33 de bovins n'ayant pas de kystes hydatiques); 80 sérums de moutons (dont 25 de moutons qui avaient des kystes hydatiques et 55 de moutons n'ayant pas de kystes hydatiques); 79 sérums de chèvres (dont 16 de chèvres qui avaient des kystes hydatiques et 63 de chèvres n'ayant pas de kystes hydatiques).

Lorsqu'un titre ELISA de 1:50 et plus a été considéré comme étant positif, une sensibilité de 68% et une spécificité de 54% ont été obtenues avec les sérums ovins. On a pu obtenir une sensibilité de 88% et une spécificité de 40% avec les sérums caprins, tandis que les sérums bovins ont donné une sensibilité de 51% et une spécificité de 70%.

Compte tenu de la faible sensibilité et spécificité obtenues avec la technique ELISA, il a été conclu que le test basé sur l'antigène thermostable de liquide de kyste hydatique (LKH) ne serait pas nécessaire pour faire le diagnostic de l'hydatidose chez le bétail. Il faudrait donc identifier d'autres antigènes *Echinococcus granulosus* dans LKH qui pourrait permettre d'obtenir une meilleure sensibilité et spécificité que l'antigène thermostable.

Summary

A total of 227 livestock sera were examined. These consisted of: 68 sera from cattle, of which 35 were from cattle which had hydatid cysts at post-mortem inspection and 33 from cattle which did not have hydatid cysts; 80 sera from sheep, of which 25 were from sheep with hydatid cysts and 55 from sheep which did not have hydatid cysts; 79 goat sera, of which 16 were from goats with hydatid cysts and 63 from goats without hydatid cysts.

When an ELISA titre of 1:50 and above was considered positive a sensitivity of 68% and a specificity of 54% were obtained with sheep sera. A sensitivity of 88% and a specificity of 40% were obtained with goat sera, while the cattle showed a sensitivity of 51% and a specificity of 70%.

Due to the low sensitivity and specificity achieved with the ELISA method, it was concluded that the test based on the thermo-stable antigen of hydatid cyst fluid may not be useful in diagnosis of hydatidosis in livestock. There is need therefore, to identify other *Echinococcus granulosus* antigens in HCF which may yield better sensitivity and specificity than the thermo-stable antigen.

INTRODUCTION

A variety of serological tests and their attendant problems in sero-diagnosis of hydatid disease in man and livestock have been described[1] The usefulness of enzyme-linked immunosorbent assay in diagnosis of livestock hydatidosis has been doubted by various workers. Light-owlers *et al.*[2] reported that the specificity of ELISA in diagnosis of hydatid disease in sheep was poor, and that there was

overlap of serological reactions between infected and non-infected sheep, thus making individual diagnosis of hydatid infection unreliable. Gathura[3] and Kagiko[4] reported low ELISA specificity and sensitivity in diagnosis of hydatid disease in cattle.

A sensitive and specific serological test for hydatidosis in animals is desirable both for epidemiological studies and possible application in hydatid disease control programmes, by facilitating selective removal of the hydatid disease positive animals.

This study was initiated with a view to investigate the possible application of ELISA based on the hydatid cyst fluid (HCF) thermo-stable antigen in diagnosis of hydatid disease in cattle, sheep and goats.

Materials and Methods

Sera: Sera were collected in universal bottles at the time of bleeding of the animals to be slaughtered. Clots were removed, the sera centrifuged at 2,000 x g and stored at 4°C with 0.1% sodium azide.

Antigen Preparation: The antigen was prepared according to a method described by Njeruh *et al.*[5,6]. The method briefly consisted of the following steps: cattle hydatid fluid was concentrated twenty-fold by ultrafiltration through amicon, PM 30 filter paper, with a cut-off point of 30,000 daltons. The concentrated fluid was heated in a pressure cooker at 1.66 atmospheres and a temperature of 110°C for 10 minutes, then centrifuged at 2,000 x g. The supernate was used as source of antigen for the enzyme-linked immunosorbent assay.

The Test Procedure: The enzyme-linked immunosorbent assay, was carried out according to a method described by Engvall and Perlmann[7] and Van Weemen and Schuurs[8]. An antigen solution containing 175μg/ml protein was diluted five hundred times in a coating buffer. The coating buffer consisted of: 0.01M phosphate buffer, pH 7.2 with 2% polyethylene-glycol (PEG), 0.04M sodium chloride and 0.02% sodium azide. One hundred microlitres of the 1:500 antigen dilution was pipetted into each well of Titertek U-shaped microtitre plates. The plates were incubated for 48 hours in a humid chamber at room temperature. The plates were then washed 5 times with a washing buffer which consisted of: 1,000 mls phosphate buffer saline (PBS), pH 7.2, 0.05% Tween 80 and 9,000 mls of distilled water. The test sera were diluted serially from 1:50 upto 1:6,400. The diluent at pH 7.5 consisted of: 0.05M phosphate buffer, 7.5% potassium chloride, 0.1% EDTA, 0.25% benzoic acid, 0.5% Tween 80 and 5% normal rabbit serum. After addition of 100μl of test sera to each well the plates were incubated for one hour in a humid chamber at 37°C. One hundred microlitres of anti-goat, anti-sheep and anti-cattle IgG-glucose oxidase conjugate was added to each well of separate plates, and the plates incubated at 37° for one hour in a humid chamber. The plates were washed five times in the washing buffer, and a substrate for glucose oxidase added. The substrate consisted of: Beta-D-glucose (17.5 Ω μg/ml), horseradish peroxidase (HRPO) type VI (8.93 μg/ml) and ABTS (0.225 μg/ml) dissolved in 0.05M citrate/ammonium acetate buffer, pH 5.0. Before the substrate was added to the plates, the mixture was decolourized with sodium dithionite until only a hazy green colour was left. After the substrate was added to the plates, the plates were incubated at room temperature for one hour and optical density (OD) readings taken using a Dynatech micro-ELISA reader. An OD reading of 50 and above was considered a positive reaction.

Known positive and negative controls were included in all the test plates. A total of 227 livestock sera which consisted of 68 sera of which 35 originated from cattle with hydatid cysts and 33 from cattle without hydatid cysts; 80 sheep sera of which 25 originated from sheep with hydatid cysts and 55 from sheep without hydatid cysts; 79 sera from goats of which 16 originated from goats with hydatid cysts and 63 from goats without hydatid cysts, were examined by the ELISA technique.

Results

When a titre of 1:50 and above was considered positive, 18 out of 35 sera from cattle with hydatid cysts were identified correctly, thus giving a sensitivity of 51%, while 23 out of 33 sera from cattle without hydatid cysts gave negative results, thus showing a specificity of 70%. The results of examination of cattle sera are shown in Fig. 1.

When a titre of 1:50 and above was considered positive, 17 out of 25 sera from sheep with hydatid cysts were identified correctly giving a sensitivity of 68%. Thirty out of 55 sera from sheep without hydatid cysts gave negative reactions, thus a specificity of 55% was recorded. The results of ELISA on sheep sera are summarized in Fig. 2.

When a titre of 1:50 and above was considered positive, 14 out of 16 sera from goats with hydatid cysts were identified correctly, thus giving a sensitivity of 88%. Twenty five out of 63 sera from goats without hydatid cysts gave negative reactions, thus showing a specificity of 40%. The results of ELISA on goats sera are summarized in Fig. 3.

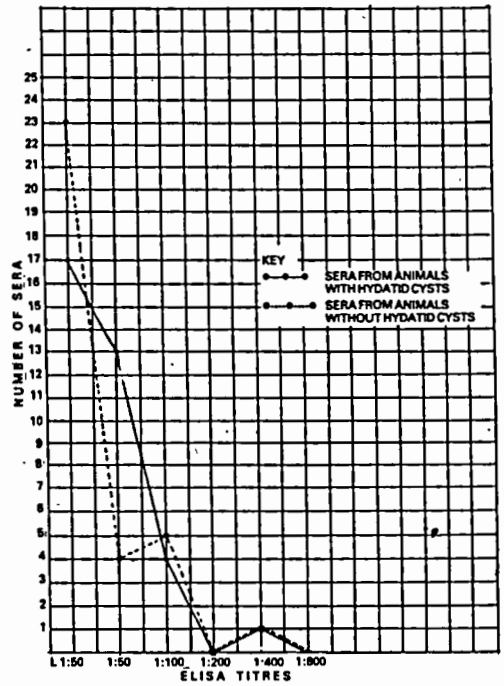

FIG. 1: ELISA TITRES OF SERA FROM CATTLE WITH AND WITHOUT HYDATID CYSTS

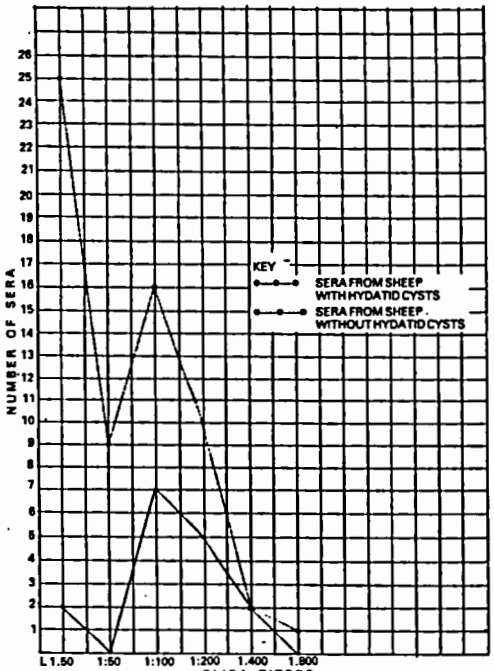

FIG. 2: ELISA TITRES OF SERA FROM SHEEP WITH AND WITHOUT HYDATID CYSTS

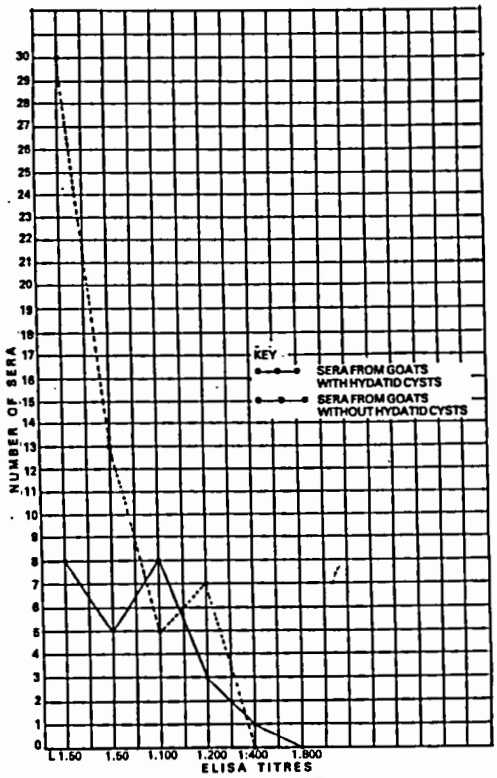

FIG. 3: ELISA TITRES OF SERA FROM GOATS WITH AND WITHOUT HYDATID CYSTS

Discussion

Serological diagnosis of hydatid disease in livestock appears to have attracted little attention from various research workers. However, possible application of sero-diagnosis of hydatid disease in livestock and selective slaughter of sero-positive animals as a tool for control of hydatid disease should be considered. This is particularly important in areas of high hydatid disease endemicity e.g. the Turkana District of Kenya, where standard methods applied in hydatid disease control have proved not particularly useful due to several factors — e.g. lack of slaughter-houses, the nomadic life-style of the Turkana people, low literacy levels of the nomads, and hence inability to appreciate the life-cycle of the *E. granulosus* parasite. It is important therefore to develop a reliable serological test which may be applied to identify serologically positive animals in such endemic areas for selective removal.

The ELISA sensitivity results of 51%, 68%, and 88% recorded with cattle, sheep and goat sera respectively are comparable to the 56% reported by Gathura[3] who used 'Arc 5' in serodiagnosis of hydatidosis in cattle. Kagiko *et al.*[9] used a combination of HCF antigens, and reported an ELISA sensitivity of 98% with cattle sera. The specificities of 69.0%, 54.0% and 54.0% recorded with cattle, sheep and goat sera respectively are comparable to the 59% reported by Gathura[3]. Kagiko *et al.*[9] reported an ELISA specificity of 70% when they examined 180 cattle sera. Lightowlers *et al.*[2] reported that the specificity of ELISA in diagnosis of hydatid disease in sheep was poor. There was overlap of serological reactions between infected and non-infected sheep, thus making individual diagnosis of hydatid infection unreliable.

The low specificity recorded with the ELISA in diagnosis of hydatid disease in livestock may be mainly attributed to presence of cross-reacting antigens between HCF antigens and other parasites which are common in livestock. Kagiko[4] and Njeruh and Lindqvist[10] identified various cross-reactions between HCF antigens of *E. granulosus* origin and other parasites of livestock.

Due to the low sensitivity and/or specificity of ELISA recorded with various species of livestock, it is concluded that this test is not useful in the diagnosis of hydatid disease in livestock.

Acknowledgements

We are grateful to the National Council for Science and Technology and the Leverhulme Trust for the financing of the research project. We appreciate the help from Mrs. Lily W. Mwihuri in typing the manuscript.

Reference

1. Rickard, M.D. (1979). *Austr. Vet. J.* **55**: 99.
2. Lightowlers, M.W., Rickard, M.D., Honey, R.D., Obendorf, D.L. and Mitchell, G.F. (1984). *Austr. Vet. J.* **61**: 101.
3. Gathura, P.B. (1984). MSc. Thesis University of Nairobi.
4. Kagiko, M.M. (1985). PhD Thesis, University of Nairobi.
5. Njeruh, F.M., Gathuma, J.M. and Tumboh-Oeri, A.G. (1986). *East Afr. Med. J.* **63**: 312.
6. Njeruh, F.M., Gathuma, J.M. and Tumboh-Oeri, A.G. (1986). *East Afr. Med. J.* **63**: 318.
7. Engvall, E. and Perlmann, P. (1971). *Immunochem.* **8**: 871.
8. Van Weemen, B.K. and Schuurs, A.H.W.M. (1971). *Febs. Leh.* **15**: 232.
9. Kagiko, M.M., Gathuma, J.M. and Lindqvist, K.J. (1986). *Int. J. Zoon.* **13**: 241.
10. Njeruh, F.M. and Lindqvist, K.J. (1982). Proceedings of the Third Annual Medical Scientific Conference (KEMRI/KETRI). 26.

Received for publication on 30th June 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 11-14

TICK-WILDLIFE ASSOCIATION IN RIVERS STATE, NIGERIA

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L'ASSOCIATION TIQUE/FAUNE SAUVAGE DANS L'ETAT DE RIVERS AU NIGERIA

Résumé

La faune sauvage de la plaine de la forêt pluvieuse, dans l'Etat de Rivers au Nigeria, a été examinée pour la détection des tiques (adultes et nymphes) au cours de la période allant de mai 1985 à avril 1986. Les animaux sauvages suivants ont été détiqués: *Neotragus batesi* (antilope naine de Bate), *Potamochoerus porcus* (sanglier de forêt), *Cephalophus maxwelli* (céphalophe de Maxwell), *Hyaena hyaena* (hyène), *Pathera pardus* (léopard), *Nandina binotata* (marte des palmiers), *Genetta pardina* (genette des forêts), *Atherurus africanus* (porc-épic de buisson), *Thoronomys Swinderlanus* (rat de jonc) et *Cricetomys gamblanus* (gros rat).

On n'a recueilli que deux tiques spp. *(Boophilus decoloratus* et *Hyalomma truncatum).* Un nombre élevé de ces deux espèces nous a été signalé pendant la saison sèche et la saison des pluies. On a aussi rassemblé quelques nymphes des deux espèces de tique (3,17%–4,43%). Les mâles constituaient 61,5 à 64,7% des adultes. L'hyène, le sanglier de forêt et la marte des palmiers étaient les hôtes préférés des tiques. Les raisons qui ont permis d'obtenir les résultats publiés dans cette communication sont exposées.

Summary

Wildlife from the lowland rain forest in Rivers State, Nigeria, were examined for ticks (adults and nymphs) during the period, May 1985 – April, 1986. The following wildlife were de-ticked: *Neotragus batesi* (Bate's dwarf Antelope), *Potamochoerus porcus* (forest hog), *Cephalophus maxwelli* (Maxwell's duiker), *Hyaena hyaena* (Hyaena), *Pathera pardus* (Leopard), *Nandina binotata* (Palm civet), *Genetta pardina* (Forest genet), *Atherurus africanus* (Bush-tailed Porcupine), *Thoronomys swinderianus* (Cane Rat) and *Cricetomys gamblanus* (Giant Rat).

Only two tick spp. *(Boophilus decoloratus* and *Hyalomma truncatum)* were collected. High numbers of both spp. were recorded in the dry and rainy seasons. Few nymps (3.17-4.43%) of both spp. were collected. Males accounted for 61.5-64.7% of adults. Hyaena, Forest Hog and Palm Civet were the most favoured hosts. Probable reasons for the results obtained are presented.

INTRODUCTION

The incessant drought affecting the Northern States in Nigeria has highlighted the need for the utilization of pastures in the Southern States of the country for livestock production. The Southern States include Rivers States. The Rivers State Government recently unveiled plans to develop cattle production in the state. Reports from the State Ministry of Agriculture (Livestock Division) indicate an increasing incidence of various arthropod-borne diseases of livestock: trypanosomiasis, babesiosis and anaplasmosis. The importance of acarines as vectors of major diseases of livestock in Nigeria has been documented[1a,1b,2a]. Our present knowledge on the spp. complex and distribution of Nigerian ticks is largely due to several surveys in the last 3 decades[3,4,5,6,1a,2b,2c,7]. The tick surveys were conducted in the Southwestern and Northern sections of the country and restricted to rangeland and livestock. The area presently designated Rivers State was not covered in any of these surveys. Preliminary observations by Okiwelu et. al[8] on cattle grazing in the rangeland near Port Harcourt (capital of Rivers State), during the dry and rainy seasons of 1981-1982, showed high populations of nymphal and adult ticks. The rainy season populations were significantly higher. The dominant species was *Amblyomma variegatum,* the vector of the rickettsia, *Cowdria ruminatum,* the causative organism of Heartwater in cat-

tle. Mohammed[1b] described this disease as endemic in Nigeria. Other spp. encountered in these preliminary investigations were *Boophilus geigyi* and *Hyalomma rufipes.* Boophilus spp. are vectors of Babesiosis in Nigeria. This disease is also endemic[2a].

Since wildlife are known to habour ticks[9], it is imperative that studies of the ecology of ticks in a habitat, should include ivestigations of those occuring on wildlife. There are few studies on wildlife-tick associations in Africa[10,11,12]. In continuation of studies on tick ecology in Rivers State, investigations were conducted on tick-wildlife associations during the rainy (May-October, 1985) and dry (November 1985-April, 1986) seasons. Its is envisaged that the results obtained will complement those reported from observations on cattle[7]. Survey of ticks on other livestock and rangeland is in progress.

Study Area

The Rivers State occupies an area of 28.00km² and lies between 4° 17N and 5° 29N, 5° 31'E and 7° 36E. Two-thirds of it lies within the Niger Delta Basin. The land surface of the State is dominated by plains of mainly 0-50m, except the north-east tip, where it is 50-100m above sea level. The annual maximum temperature for the capital, Port Harcourt, is 30°C. The hottest months are February, March and April. The coldest month is usually August. The RH is high throughout the year, because it is affected by humid maritime airmass all year round. The lowest mean annual rainfall is 2500mm in the extreme north-east. Along the coast, it is more than 4000mm. The vegetation consists of: lowland rainforest, with Pennisetum-dominated grass species in the North-East, freshwater swamp forest, saltwater swamp-forest in the south (except at the coast, where coastal grass spp. dominate).

Materials and Methods

Studies were undertaken during the period, May 1985 - April 1986. The wildlife examined in this study were collected from the lowland rainforest. Ticks were collected from wildlife shot by local hunters, De-ticking proceeded immediately on arrival at the wildlife meat market at Omagwa. The following wildlife were deticked: *Artiodactyla-Neotragus batesi* (Bate's dwarf antelope), *Potamochoerus porcus* (Forest hog), *Cephalophus maxwelli* (Maxwell's duiker) Carnivora – *Hyaena hayaena,* (Hyaena), *Panthera pardus* (Leopard), *Nandina binotata* (Palm civet), *Genetta Pardina* (Forest Genet) and rodents *Atherurus africanus* (Bush tailed Porcupine), *Thryonomys swinderianus* (Cane Rat) *Cricetomys gambianus* (Giant Rat). All wildlife examined were collected from the lowland rainforest of the State.

Ticks were examined and identified. Nymphs and adults of each tick species on each wildlife sp. were counted. Monthly data were recorded.

Results

Only two tick species *Boophilus decoloratus* and *Hyalomma truncatum* were recorded (Table 1). More *H. truncatum* were collected; it accounted for 65% of all ticks collected. This ratio was consistent both in the rainy and dry seasons. Both species occurred throughout the year. Nymphs accounted for only 3.17% and 4.43% of the *Hyalomma* and *Boophilus* collections respectively and the rest were adults. Males accounted for 61.5% of adult *Boophilus* and 64.73% of adult *Hyalomma.* Larvae were not recorded.

Ticks were found on all wildlife examined, throughout the year (Table 2). High numbers of ticks, in a descending order, were collected from Hyaena, Forest hog, Palm civet and Cane rat, during the rainy season. Few ticks, in a descending order, were collected from Antelope, Genet, Duiker and Giant rat during the rainy season. No leopard was examined in the rainy season.

In the dry season, many ticks were recorded, in a descending order, from Hyaena, Cane rat, Forest hog and Antelope. Few ticks, in a descending order, were collected from Genet, Leopard, Civet, Duiker and Giant rat.

Table 1: Seasonal and monthly numbers of tick spp. collected off wildlife in Rivers State (June, 1985 – May, 1986)

	P		E		R		I		O		D		
Tick spp.	**RAINY**					**DRY**							**Grand Totals**
	May	**June**	**July**	**Aug.**	**Sept.**	**Oct.**	**Nov.**	**Dec.**	**Jan.**	**Feb.**	**Mar.**	**April**	
Boophilus decoloratus	107	205	39	327	157	43	136	112	64	146	119	102	1,557
Hyalomma rufipes	387	142	13	480	286	314	280	128	135	406	170	162	2,903
Season Totals	2,500 (Rainy)					1,960 (Dry)							

Table 2: Numbers of ticks on various wildlife hosts

Season	Host*	*Boophilus decoloratus*		*Hyaloma truncatum*	
		Adults	Nymphs	Adults	Nymphs
Rainy (May – October 1985)	*H. hyaena*	392	14	162	2
	N. batesi	233	0	18	2
	P. porcus	192	0	845	3
	N. binotata	66	14	0	0
	A. africanus	131	0	519	35
	G. pardina	16	0	16	0
	C. maxwelli	7	3	38	2
		827	31	1,598	44
Dry (November 1985 – April, 1986)	*A. africanus*	151	1	410	26
	H. hyaena	340	9	100	4
	G. pardina	22	0	10	0
	C. maxwelli	7	0	50	4
	N. batesi	62	15	55	3
	P. porcus	68	4	568	9
	P. pardus	16	0	12	8
	N. binotata	0	0	2	0
		666	33	1,207	54

*Numbers of hosts examined varied.

Discussion

In Hoogstraal's dendogram of tick phylogeny, *Boophilus* and *Hyalomma* are among the genera of the sub-family Metastriata[9]. It is therefore not surprising that the closely related genera *Boophilus* and *Hyalomma* are closely associated on the same hosts. Hoogstraal stated that *Boophilus decoloratus* has co-evolved with the magnificient variety of artiodactyls that enrich the extensive African savannas[9]. It is obvious that *B. decoloratus* has extended its range, beyond the artiodactyls, since many were collected from carnivores (hyaena, civet) and rodents (cane rat). Hoogstraal also notes that in parts of West Africa, *B. decoloratus* has been replaced by *B. geigyi*[9]. This phenomenon may explain the replacement of *B. decoloratus* by *B. geigyi* on cattle examined at the main abbaittor at Port Harcourt, capital of Rivers State[8].

Ixodids had also been recorded on different groups of large and small mammals by other workers. Hoogstraal stated that ticks were found on medium to small

rodents[11]. Sachs and Sachs collected ixodids from bovids[12]. Yeoman and Walker also recorded the closely related ixodid, Rhipicephalus on carnivores (Mongoose, Genet, Hyrax)[11].

Finally, it should be mentioned that the high numbers of ticks on Hyaena, Red river hog and Cane rat were probably also attributable to the significantly higher frequencies of these mammals among all wildlife examined.

References

1a. Mohammed, A.N. (1974). *The seasonal incidence of ticks on cattle in Northern Nigeria with particular reference to their role in the transmission of bovine piroplasm.* Ph. D. Thesis. Ahmadu Bello University, Zaria, Nigeria.

1b. Mohammed, A.N. (1980). *Occ. Publ. Ent. Soc. Nigeria* **(25)**:53-58.

2a. Dipeolu, O.O. (1976). *Occ. Publ. Ent. Soc. Nig.* **(18)**:24-39.

2b. Dipeolu, O.O. (1975). *Bull. Anim. Hith Prod. Afr.* **(XXIII)**:165-167.

2c. Dipeolu, O.O. (1975). *Trop. Anim. Hith Prod.* **(7)**:35-39.

3. Metta, R.W. (1951). Nigeria, 1949-50, Annual Report Veterinary Department. p. 32.

4. Gambles, R.M. (1957). Nigeria, 1950-1951, Annual Report Veterinary Department, p. 39.

5. Unsworth, K. (1952). *Ann. Trop. Med. Parasitol.* **46**:331-336.

6. Strickland, K.L. (1961). *Studies of the tick of domestic animals of Northern Nigeria. Apr. preliminary to Disease Investigation.* M. Sc. Thesis, Trinity College, Dublin.

7. Iwuala, M.O.E. and I. Okpala (1978). *Bull. Anim. Hith Prod. XXVI* **(4)**:339-350.

8. Okiwelu, S.N., B. Darwang and c. Ndoma (1983). *Bull Anim. Hith. Prod. Afr.* **23** (2):127-135.

9. Hoogstraal, Harry (1986). *Bull. Ent. Soc. Amer.* **32** (1):23-35.

10. Yeoman, G.H. (1966). *Bull. Epiz. Dis.* **14**:113-140.

11. Yeoman, G.H. and J.B. Walker (1967). *The Ixodid Ticks of Tanzania.* Commonwealth Institute of Entomology, London.

12. Sachs, R. and C. Sachs (1968). *Bull. Epiz. Dis.* **16**:455-472.

Received for publication on 10th May 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 15-18

CASEOUS LYMPHADENITIS OF SHEEP AND GOATS IN KENYA

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LYMPHADENITE CASEIFORME DES OVINS ET DES CAPRINS AU KENYA

Résumé

Dans une étude sur la lymphadénite caséiforme chez les ovins et les caprins indigènes au Kenya, 54 des 757 chèvres (7,13%) et 6 des 378 moutons (1,6%) avaient un ou plusieurs abcès. L'abcès était très fréquent dans les ganglions lymphatiques prescapulaires (68,25%) suivis par les ganglions lymphatiques précruraux (14,28%). On a observé six abscès sous-cutanés près des ganglions lymphatiques superficiels qui n'ont subi aucune infection. L'histopathologie des abcès a révélé une inflammation sous forme de granulome. *C. pseudotuberculosis* a été isolé de 79,36% des abcès dans des cultures pures ou mélangées. D'autres micro-organismes furent isolés de 11,1% des abcès, tandis que 9,52% étaient stériles.

Summary

In a survey of caseous lymphadenitis in local sheep and goats in Kenya, 54 of 757 goats (7.13%) and six of 378 sheep (1.6%) were found to have one or more abscesses. Abscessation was commonest in the prescapular (68.25%) followed by the precrural (14.28%) lymphnodes. Six abscesses were located subcutaneously close to uninvolved superficial lymphnodes. Histopathology of the abscesses showed granulomatous inflammation. *C. pseudotuberculosis* was isolated from 79.36% of the abscesses in pure or mixed cultures. From 11.1% of the abscesses other micro-organisms were isolated while 9.52% were found sterile.

INTRODUCTION

Caseous lymphadenitis (CLA) is a chronic insidious disease mainly of sheep and goats which is caused by *Corynebacterium pseudotuberculosis*[1,2,3]. The disease is characterised by caseous lesions in peripheral lymphnodes but can also spread to visceral lymphnodes and organs[1,4,5]. CLA is usually clinically inapparent and is observed during slaughter although the disseminated form causes a chronic debilitating condition[5,6,7].

CLA has a worldwide distribution and causes high economic losses through condemnation of affected carcasses and organs at slaughter,[4,3] and through losses in production and reproduction[5,9,10].

In Kenya, two cases of CLA have been reported in an exotic sheep flock[11]. Infections by *C. pseudotuberculosis* have also been reported in cattle,[12,13], and in rhinoceros[12]. This investigation was aimed at obtaining information on CLA in local sheep and goats in Kenya with respect to the prevalence, the pathological manifestation and the isolation of the aetiological agent.

Materials and Methods

Animals

Carcasses of sheep and goats were examined during post mortem meat inspection in four slaughterhouses in Nairobi. Animals slaughtered in these slaughterhouses come from different parts of the country but mainly from the nomadic North-Eastern, Rift Valley and Eastern provinces. The breeds of goats were mainly the Galla and the East African. Sheep breeds were the Dopper, Black-head Persian, the Red Maasai and cross-breeds.

Post mortem Examination Procedure

The superficial and internal lym-

phnodes of the carcasses were incised and examined for abscesses. Visceral organs were examined by palpation and incisions. Any abscessed lymphnode or lesion was excised, put separately into plastic containers and transported to the laboratory.

Bacteriology

Pus from abscesses was cultured on 5% ox blood agar and incubated at 37°C for a maximum of two days. Bacterial isolates were characterised and identified on the basis of their cultural characteristics, cellular morphology and staining and biochemical reactions[14].

Histopathology

Abscessed lesions and lymphnodes were fixed in 10% formalin, embedded in paraffin wax and sectioned at 5μm thickness (Rotary microtome, Leitz, West Germany). Sections were stained with Haematoxylin and Eosin (H&E).

Results

Abscesses

A total of 757 carcasses of goats were examined. Fifty four carcasses (7.13%) had abscesses in one or two locations. Altogether, 56 abscesses were observed (Table 1). Fifty of them were located exclusively in peripheral lymphnodes (40 prescapular, six precrural, three parotid, and one submaxilar), two involved precrural lymphnodes as well as the adjacent subcutaneous tissue, while four affected only subcutaneous tissue adjacent to peripheral lymphnodes (three in prescapular regions and one in a precrural area).

Three hundred and seventy eight sheep carcasses were examined. Six of them (1.6%) had abscesses in one or two locations. A total of seven abscesses were observed, which were distributed as follows:- three in prescapular lymphnodes; one in a precrural lymphnode; one each in the subcutaneous tissue adjacent to a prescapular and a precrural lymphnode; and one in the lung parenchyma (Table 1).

Lymphnode abscesses affected either the entire node, one pole, or occurred as foci in different areas of the node parenchyma. The pus was either creamy or greenish-yellow in colour, and the consistency either caseous, granular or hard and calcified. Pus in one lymphnode abscess from a sheep was arranged in onion-like concentric circles. Subcutaneous abscesses were all greenish-yellow and caseous and none was calcified.

Table 2: The type and frequency of isolation of microorganisms from caseous sheep and goat abscesses

	Culture type		
Organism	**pure**	**mixed**	**total**
C. pseudotuberculosis	36	14	50
Bacillus spp.	1	8	9
Streptococcus spp.	2	4	6
Staphylococcus spp.	3	1	4
Coliforms	0	4	4
Pseudomonas spp.	0	1	1
None	–	–	6

Table 1: The number and distribution of caseous abscesses in sheep and goats

	Number of abscesses			
Location of abscesses	**Sheep**	**%**	**Goats**	**%**
L.N. prescapular	3	42.85	40	71.42
L.N. precrural	1	14.28	8	14.28
L.N. parotid	0	–	3	5.35
L.N. submaxillar	0	–	1	1.78
Lung	1	14.28	0	–
Subcutaneous	2	28.57	4	7.14
Total	7		56	

L.N.=Lymphnode

Histopathology

Microscopically, the lesions consisted of a necrotic centre surrounded by a zone of neutrophils, giant cells, macrophages, plasma and epithelioid cells. Lesions were encapsulated by a dense layer of fibroblasts.

Bacteriology

Table 2 shows the types and frequency of isolation of micro-organisms from the sheep and goat abscesses. *C. pseudotuberculosis* was isolated from 50 of 63 abscesses (79.36%) either in pure cultures (36 of 50 instances) or together with other organisms (14 cases). Seven abscesses (11.1%) yielded other organisms *(Bacillus sp., Streptococcus sp., Staphylococcus sp.,* Coliforms or *Pseudomonas sp.,)* in pure or mixed cultures, while another six (9.52%) including the pulmonary abscess (from a sheep) were bacteriologically sterile.

Discussion

The present results show that CLA is prevalent in local sheep and goats in Kenya and that *Corynebacterium pseudotuberculosis* is an important causative agent. The organism was isolated from majority of the lesions (79.4%) either in pure or mixed cultures. Other organisms (pyogenic and non-pyogenic) were recovered from 11.1% of the lesions. These other organisms were presumably secondary invaders that had overgrown the primary pathogen in the lesions. Apart from *C. pseudotuberculosis,* other pyogenic organisms such as *Staphylococcus aureas* and *Corybacterium pyogenes* have only experimentally (in sheep) been shown to cause abscesses in superficial lympnodes[15]. These other organisms are also unlikely to cause the typical caseous granulomatous lesions of *C. pseudotuberculosis*[3]. A condition of subcutaneous abscesses located close to lymphnodes in sheep (Morel's disease), caused by a Gram positive *Micrococcus,* has been reported in Kenya[11]. No such organism was however isolated from the subcutaneous abscesses, which were observed in both sheep and goats. Only *C. pseudotuberculosis* was isolated from one of such abscesses.

The prevalence of caseous lesions in goats (7.1%) was similar to those reported in other parts of the world[16,17,18]. The observed prevalence in sheep (1.6%) was much lower than those reported by other workers[2,19]. The commonest route of infection by *C. pseudotuberculosis* in sheep is through superficial wounds such as those inflicted during shearing[20,21]. The local sheep in Kenya are not normally shorn. The unshorn wool possibly in turn protects the sheep against thorns and other sharp objects that may inflict superficial wounds.

The commonest site of abscessation in both sheep and goats was the prescapular lymphnodes. With respect to sheep in the present results, the site was in agreement with other reports[1,21,22]. Lymphnodes of the head region are reportedly the commonest site of infection in goats[10,16,18,19]. This possibly results from infections through traumatised buccal mucosae[16]. The majority of the goats examined in this work came from arid and semi arid areas with vegetation of thorny bushes. Superficial scratches inflicted during browsing would therefore be the most likely route of infection, with subsequent high infection rate of superficial body lymphnodes.

Only in one (out of four) lymphnode abscess from sheep was the pus arranged in onion-like concentric circles although this is considered a classical feature in ovine *C. pseudotuberculosis* lymphadenitis[1,3] and a major distinction between caprine and ovine CLA[19]. No goat abscess had such a pattern of pus arrangement but insipissation and calcification which is considered rare,[18] was frequently observed.

In the present investigation, CLA of local sheep and goats in Kenya under nomadic conditions was found to have a low and a moderate prevalence rate respectively. The pathological manifestation of the disease in both species was mild. None of the affected carcasses had more than two abscesses and abscesses in all carcasses (except one) were superficially located. The disseminated form of the disease was not observed.

Acknowledgement

This work was supported by University of Nairobi Deans' committee grant No. 655-071.

References

1. Marsh, H. (1965). Diseases caused by bacteria in *"Newsoms sheep diseases"* 3rd Ed, Williams and Wilkins, Baltimore, md. pp. 87-90.
2. Williamson, P. and Nairn, M.E. (1980). *Aust. Vet. J.* **58**:496.
3. Jubb, K.V.F. and Kennedy, P.C. (1970). *Pathology of domestic animals,* 2nd Ed. N.Y. Acad. press Inc. vol. 1, pp. 373-375.
4. Addo. P.B. and Eid., F.I.A. (1978). *Bull. Anim. Hlth. prod. Afr.* **25**:37.
5. Renshaw, H.W., Graff, V.P. Gates, N.L. (1979). *Am. J. Vet. Res.* **40**:1110.
6. East, N.E. (1982). *Comp. Contin. Educ. Pract. Vet.* **4**:419.
7. Blood, D.C., Rodostits, O.M., Handerson, J.A. (1983). *Veterinary Medicine,* 6th Ed., Pitman press Ltd. U.K. pp. 517-518.
8. Turner, L.W. (1973). *77th proc. Annual Meeting of U.S. Ani. Hlth Asso. 650.*
9. Mostafa, A.S., Khater, A., Soyour, E.M. and El sawaf, S. A. (1973). *J. Egypt. Vet. Med. Ass* **33**:127.
10. Burrel, D.H. (1981). *Aust. Vet. J.* **57**:105.
11. Shirlaw, J.F. and Ashford, W.A. (1962). *Vet. Res* **74**:1025.
12. Purchase, H.S. (1944). *J. Comp. Path.* **54**:232.
13. Kariuki, D.P. and Poulton, J. (1982). *Trop. Anim Hlth. Prod.* **14**:33.
14. Buchanan, R.R. and Gibbons, N.E. (1974) *Bergeys manual of Determinative Bacteriology* 8th Ed., Williams and Wilkins and Wilkins, Balti more, Md.
15. Richard, Y., Fontaine, P. (1979). *Comp. Immun Microbiol. Infect. Dis.* **2**:125.
16. Ashfaq, M.K. and Campbell, S.G. (1979) *VM/SAC.* **74**:1161.
17. Hein, H.R. and Cargill, C.F. (1981). *Aust. Vet. J* **57**:498.
18. Batey, R.G., Speed, C.M., Kobes, C.J. (1986) *Aust. Vet. J.* **63**:33.
19. Brown, C.C. and Olander, J.J. (1987). *Vet. Bul* **57**:1.
20. Nagy, G. (1976). *J.I. S. Afr. Vet. Med. Ass.* **47** 197.
21. Nairn, M.E. and Robertson, J.P. (1974).*Aust. Ve J.* **50**:537.
22. Ayers, J.L. (1977). *JAVMA.* **171**:1251.

Received for publication on 25th April 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 19-21

PREVALENCE OF BOVINE BRUCELLOSIS IN DAIRY HERD PRE AND POST ISOLATION OF REACTORS

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PREVALENCE DE LA BRUCELLOSE BOVINE CHEZ UN TROUPEAU LAITIER AVANT ET APRES L'ISOLEMENT DES ANIMAUX INFECTES

Résumé

La prévalence de la brucellose bovine a fait l'objet d'études chez un troupeau laitier sous exploitation intensive pendant deux années consécutives. Les taureaux reproducteurs avaient le taux d'incidence le plus élevé (71,4%) suivis par les vaches taries (37,6%). En dépit de l'isolement de tous les animaux infectés, la moyenne globale du taux d'incidence au cours de la deuxième année d'enquête était élevée contrairement aux prévisions (19,9% pour la première année contre 5,9% pour la deuxième année).

Le test de fixation du complément (CFT) s'est avéré le test de diagnostic sérologique le plus fiable que l'on pourrait utiliser pour confirmer les résultats négatifs et incertains obtenus avec le test sur Plaque de Rose Bengal (RBPT) et le test d'agglutination du sérum (SAT).

Summary

Prevalence of bovine brucellosis was studied in a dairy herd under intensive husbandry management for two consecutive years. The serving bulls showed the highest incidence rate (71.4%) followed by dry cows (37.6%). Inspite of isolation of all reactors, the overall average incidence rate during the second year survey was unexpectedly high (19.9% for the first year compared to 5.9% for second year).

The Complement Fixation Test (CFT) was found to be the most reliable serological diagnostic test that could be utilized for confirmation of Rose Bengal Plate Test (RBPT) and Serum Agglutination Test (SAT) negative and doubtful results.

INTRODUCTION

Despite the prevalence of bovine brucellosis among Sudanese cattle with fairly high incidence rate[1,2,3] and despite the nature of the disease as a public health hazzard and it's great economic importance, the disease has been over shadowed till recently by major killing diseases like rinderpest. However, with the developing livestock industry, the economic significance of this disease and it's potential danger to human health cannot be disregarded. Therefore, an efficient control programme is desirable. The efficacy of such programmes necessitates close assessment of the tools used in surveying the disease and their ability to detect latent carriers[4].

In this study the most currently used serological tests for diagnosing brucellosis were compared. The efficacy of reactors isolation policy as a means for controlling bovine brucellosis was observed.

Materials and Methods

The herd used in this study belonged to a well managed dairy farm around Khartoum. The total number of cattle surveyed was 371 for the year 1985 and 270 for year 1986. During these surveys no animal was introduced to the farm. The herd was composed of milking cows, dry cows, heifers, bulls and male calves. The breed types of the herd members were crossbred (local Zebu X European Friesian) and recently (1984) pure European Friesian cows were admitted to the farm.

None of the animals tested were vaccinated against brucellosis.

Blood from the jugular vein was collected into sterile vaccutainers and serum was separated by centrifugation. Rose Bengal plate (RBPT) was assayed

immediately while serum Agglutination Tast (SAT) and Complement Fixation Test (CFT) were performed latter on the stored sera.

All serological tests were carried out according to the standard procedures described by Morgan[5].

The CFT was applied in year 1986 for confirming the sera that were either doubtful to RBPT or negative SAT. The animal was considered positive to SAT whenever it gave a titre of 2/20 or more, this is equivalent to 31 IU/ml. A titre of 1/10 or more is regarded as positive for CFT since none of the animals tested were exposed to vaccination. Reactors found in the year 1985 were isolated from the herd.

Results

The incidence of brucellosis in the herd was 19.9% for the year 1985 compared to 5.9% for the year 1986 Table (1). All the recently introduced Friesian cows were negative for the three serological tests. The serving bulls showed the highest prevalence rate (71.1%) followed by dry cows (37.6%) then heifers (14.2%) and the milking cows being the least (12.5%). The male calves were found negative to all tests. During the year 1986 a marked decrease in the incidence of brucellosis among all members of the herd was recorded after isolation of the reactors tested in year 1985.

During surveillence in 1985 RBPT and

Table 1: Incidence of brucellosis in dairy herd based on three serological tests for two consecutive years

Animal condition	Year of testing	Total Number Tested	RBPT +	RBPT −	RBPT ±	RBPT % Reactors	SAT +	SAT −	SAT % Reactors	CFT +	CFT −	CFT % Reactors
Milking cows	1985	128	6	112	0	12.5	16	112	12.5			
	1986	94	8	86	0	8.5	8	86	8.5	8	86	8.5
Dry cows	1985	85	32	53	0	37.6	32	53	37.6			
	1986	146	5	139	2	3.4	6	146	4.1	7	139	4.8
Bulls	1985	7	5	2	0	71.4	5	2	71.4			
	1986	3	0	3	0	0	0	3	0	0	3	0
Heifers	1985	148	21	127	0	14.2	21	127	14.2	21	127	14.2
	1986	39	2	37	0	5.1	2	37	5.1	2	37	5.1
Male calves	1985	3	0	0	0	0	0	3	0			
	1986	8	0	8	0	0	0	8	0	0	8	0
Total	1985	371	74	297	0	19.9	74	297	19.9			
	1986	290	15	273	2	5.2	16	274	5.5	17	273	5.9

+Positive, −negative, ±doubtful results of tests applied.

Table 2: Distribution of titres for positive results of CFT and SAT

Animal condition	Year of testing	Positive titres for SAT 2	3	2	2	3	2	3	Positive titres for CFT 1	1	1	1	1	1	1
Milking		20	40	80	160	320	640	1280	10	40	40	80	160	320	640
Cows	1985	2	5	6	2	1	0	0							
	1986	1	2	1	1	3	0	0	4	0	2	0	0	2	0
Dry cows	1985	4	3	3	4	5	5	8							
	1986	0	2	1	1	0	2	6	3	2	0	0	1	0	1
Bulls	1985	0	1	1	1	1	0	1							
	1986	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Heifers	1985	0	2	3	4	3	4	5							
	1986	0	0	1	1	0	0	0	0	0	0	1	1	0	0
Male	1985	0	0	0	0	0	0	0							
	1986	0	0	0	0	0	0	0	0	0	0	0	0	0	0
% of each titre	1985	8.1	14.9	17.6	14.9	18.5	12.2	18.9							
	1986	6.3	2.5	18.8	18.8	18.8	12.2	0	14.2	11.8	11.8	5.9	4.8	4.8	5.9

SAT gave doubtfull or negative results; those were confirmed positive by CFT. with incidence rate of 3.4% for RBPT, 4.1% for SAT 4.8% for CFT. It is quite clear that the distribution of titres (Table 2) during the first and second surveillence reflected a marked differences in the infection rate. However, in the second survey most of the reactors were more or less concentrated in the lower level of serological titres.

Discussion

The high prevalence rate of bovine brucellosis during the first year survey at both, herd and group levels, was probably due to the intensive husbandary management adopted where the reactor animals are in permanent contact with healthy ones. The high incidence rate among serving bulls (71.4%) may be a factor in dissiminating the disease. It is worth-mentioning that the cross-bred bulls used for serving the Friesian cows were found to be free of the disease through out the two test-periods.

Despite the dramatic decrease in overall incidence rate for the second year survey following the isolation of all reactors, yet the situation was disappointing. The results reflected either the limited ability of the currently used serological tests for detection of brucella antibodies or the occurrence of latent carriers. It has been reported that a breakdown occurs in heifers which were negative to brucellosis blood tests up to two years after the removal of reactors[4]. Other breakdowns had been recorded in brucella free-zone areas[5]. These observations were in a close agreement with the present findings suggesting the possibility of an in apparent latent infection of long standing nature. Moreover, whatever the number of latent carriers is small, the damage they might cause due to the contagious nature of the disease, can be extensive. Therefore, to rely on test and slaughter policy in conjuction with proper sanitary measures[7] were found unsatisfactory for eradication of brucellosis. In addition to the above mentioned measures, the routine survey of brucella antibodies together with isolation of reactors could hinder the spread of infection and localize it.

It was clear that the reactors in the second year surveillence possessed less degree of infection. This might point towards the inability of the RBPT and SAT to detect the very low level of the circulating brucella antibodies. Hence the cases that were classified as doubtful by RBPT or negative by SAT found positive by CFT. In general this is in agreement with Stemshorn[8]. Though RBPT is essentially employed as screening test in serological diagnosis of bovine brucellosis together with SAT, Whenever, there are conflicting results between RBPT and SAT, application CFT as a confirmatory procedure is necessary.

References

1. Nasri, M.E. (1960) *Veterinary Record* 72 pp 1200.
2. Dafaalla, N. (1962) *Sudan Journal of Veterinary Science and Animal Husbandary*, 3 pp 80.
3. Ibrahim, A.E. & Habiballa, N. (1975). *Tropical Animal Health and Production* 7 pp. 245.
4. Dolan, L.A. (1980) *Veterinary Record* **106** pp. 241.
5. Morgan, W.J.B. Mackinnon, D.J., Gill, K.P.W. Gower. S.G.M. & Norris, P.I.W. (1978). *Brucellosis Diagnosis standard laboratory Techniques.* Second edition Ventrsl Veterinary Laboratory, New Haw, Weybridge, Surrey KT 15 #HB. Ministry of Agriculture Fisheries and Food pp. 126.
6. Lapraik R.D. & Moffat. (1982) *Veterinary Record* **111** pp. 578.
7. Hussein, A.S., Singh. S.S, & Haji (1978) *Bulletin of Animal Health and Production in Africa* **26** pp. 150.
8. Stemshorn, B.W., Forbes, L.B., Eaglesome, M.D., Nielsen, K.H., Robertson, F.J. & Samagh, B.S. (1985). *Canadian Journal of Comparative Medicine* **49** pp. 391.

Received for publication on 27th April 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 23-25

BRUCELLOSIS IN SHEEP AND GOATS IN CENTRAL ETHIOPIA

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LA BRUCELLOSE CHEZ LES OVINS ET LES CAPRINS DANS LE CENTRE DE L'ETHIOPIE

Résumé

Des échantillons de sérum prélevés de 1.507 moutons et 1.197 chèvres à l'abattoir d'Addis-Abéba, à Debre Berhan et à Abomessa ont été examinés pour détecter la brucellose à l'aide du test sur Plaque de Rose Bengal. Les taux globaux de prévalence étaient respectivement de 1,5% et 1,3% chez les moutons et les chèvres. Il est proposé de recourir à la vaccination avec la souche Rev. 1 comme moyens éventuels de lutte contre la brucellose chez les petits ruminants.

Summary

Serum samples collected from 1507 sheep and 1197 goats at Addis Ababa abattoir, Debre Berhan and Abomessa were tested for brucellosis using the Rose Bengal test. The overall prevalence rates were 1.5 and 1.3% in sheep and goats respectively. Vaccination with strain Rev. 1 is suggested as the possible means of control of brucellosis in small ruminants.

INTRODUCTION

Brucellosis is the cause of abortion and infertility in sheep and goats[1]. It is a disease which occurs due to infection with *Brucella melitensis* or *Brucella abortus* and at times with other brucella organisms.

Although there are 23 million sheep and 18 million goats, there are limited reports on brucellosis in small ruminants in Ethiopia. *Brucella abortus* was isolated from cattle and *Brucella melitensis* from goats[2,3]. Ovine and caprine brucellosis have also been reported by the Office International des Epizooties[4].

Brucellosis has been reported in small ruminants in neighbouring countries. Prevalence rates of 1.7% of 2227 sheep and 1.5% of 5674 goats in the Sudan[5], 6.01% of 505 sheep and goats in Kenya[6] and 2.8% of 250 goats, 7.20% of 250 sheep and 5.29% of 340 goats in Somalia[7,8] were reported. Chukwu[9] gave a detailed review on the prevalence of brucellosis in domestic animals in different African countries. The disease is also endemic around the Mediterranean, North Africa and the Middle East[10].

Since the prevalence of brucellosis has not been studied in small ruminants in Ethiopia, this epidemiologic work was conducted from mid 1985 to mid 1987.

Materials and Methods

Serum samples were collected from 1062 sheep and 561 goats slaughtered at the Addis Ababa abattoir and from a field population of 445 sheep and 636 goats. The sera were preserved at -20°C until tested.

The origin of most sheep and goats slaughtered at Addis Ababa abattoir were from the central parts of Ethiopia[11]. The sheep originated mainly from the highlands of Arsi, Welo and Shewa regions while the goats originated from the lowland areas. The two major sources of supply of goats were Ameya and Mecha which are 200 km west of Addis Ababa. The field samples were obtained from Debre Berhan and Abomessa, 130 and 200 km north and east of Addis Ababa respectively (Figure 1).

All sera were tested with the Rose Bengal test (RBT)[1,3,12,13] using the antigen supplied by the Central Veterinary Laboratory, Weybridge, United Kingdom.

Data were analysed using Chi-square method[14].

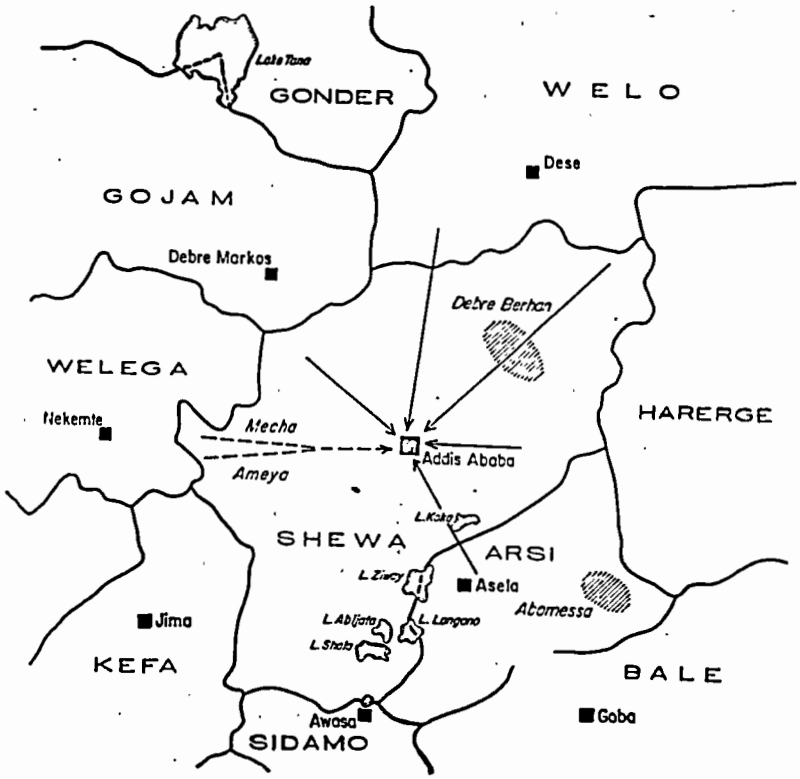

Figure 1. Central Ethiopia

Table 1: The Prevalence of brucellosis using Rose Bengal test

Place of sample collection	Sheep			Goat		
	No. of sample	No. positive	% positive	No. of sample	No. positive	% positive
Addis Ababa abattoir	1062	22	2.1	561	1	0.
Abomessa	–	–	–	636	15	2.
Debre Berhan	445	0	0	–	–	
Total	1507	22	1.5	1197	16	1.

Results and Discussions

The results of the RBT are presented in Table 1. The prevalence of brucellosis was 2.1% of 1062 sheep and 0.2% of 561 goats tested from Addis Ababa abattoir. The field samples gave a value of 0% of 445 sheep for Debre Berhan and 2.4% of 636 goats for Abomessa. The overall prevalence rates were 1.5% of 1507 sheep and 1.3% of 1197 goats.

The values obtained for sheep and goats from Addis Ababa abattoir and field samples differ significantly ($P<0.05$).

The prevalence rates range from 0 to 2.1% for sheep from Debre Berhan and those slaughtered at Addis Ababa abattoir. The variation is 0.2 to 2.4% for goats from Addis Ababa abbattoir and Abomessa. Such differences signify the varation in the prevalence of brucellosis in different localities. It is unlikely that the

discrepancy resulted from the slaughter of culled animals that harbour brucella organisms since it is reversed in sheep and goats.

The overall prevalence of brucellosis is low in Ethiopia. This agrees with findings in neighbouring countries such as the Sudan[5], Kenya[6] and Somalia[7,8]. But since the present serological methods are not reliable indicators of brucella infections in sheep and goats, the interpretation of agglutination tests should be guarded. A positive serological reactions suggest only the presence of the disease in a population[13]

The present serological techniques are not adequate to enable serological screening and elimination of positive reactors as a means of control; but vaccination with Rev. 1 through the conjunctival route may be helpful to control the disease in sheep and goats[3].

Acknowledgement

The authors wish to acknowledge the coordination of blood sample collection from sheep and goats at Debre Berhan and Abomessa by Drs. Amare Tadesse and Yosef Sheramo, the veterinarians of the Ministry of Agriculture.

References

1. Nicoletti, P. (1982). Diagnosis and vaccination for the control of brucellosis in the Near East. FAO. *Anim. Prod. Hlth. Pap.*, No. 38, 42 pp.
2. Booker Agriculture Limited (1981). *Regional Animal Diseases Control Project, Volume III, Annexes-Ethiopia.* Edinburgh.
3. Fensterebank, R. (1986). *Rev. Sci. Tec. Off. Int. Epiz.*, **5**,605.
4. O.I.E. (1983). *Zoosanitary Situation in Member Countries In 1982. Paris, France.*
5. Abdulla, A.E.D. (1966). *Sud. J. Vet. Sci. Anim. Husb.*, **7**,28.
6. Waghela, S. (1976). *Bull. Anim. Hlth. Prod. Afr.*, **1**,53.
7. Falade, S. Hussein A.H. (1979). *Trop. Anim. Hlth. Prod.*, **11**,211.
8. Andreani, E., Prosperi, S., Salim, A.H., and Arush, A.M. (1982). *Rev. Elev. Med. Vet. Pays. Trop.*, **35**,329.
9. Chukwu, G.G. (1985). *Bull. Anim. Hlth. Prod. Afr.*, **33**,193.
10. Alton, G.G. (1973). *Wld. Anim. Rev.*, **5**,16.
11. Getachew, A. (1977). The organization of meat market in Addis Ababa. ILCA, Addis Ababa, 59 pp.
12. Falade, S. and Ezwnwane, E. (1984). *Bull. Ani. Hlth. Prod. Afr.*, **32**,43.
13. Kolar, J. (1984). *Prev. Vet. Med.*, **2**,215.
14. Snedecor, G.W. and Cocharn, W.G. (1967). *Statistical Methods.* Ames, Iowa, USA, The Iowa State University Press.

Received for publication on 15th April 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 27-31

SPIRAMYCIN ADIPIQUE-INDUCED IMMUNOSUPPRESSION IN CHICKENS

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EFFET IMMUNODEPRESSEUR PROVOQUE PAR LA SPIRAMYCINE ADIPIQUE CHEZ LES POULETS

Résumé

L'effet de la spiramycine adipique à une dose de 50mg/kg de poids vif (avant, au cours ou après la vaccination) sur la réaction immunitaire au vaccin contre le virus de la maladie de Newcastle (souche Lasota) a fait l'objet d'études chez 180 poulets Hubbard. Ce médicament a augmenté le nombre de lymphocytes mais a, par contre, réduit celui des éosinophiles et des hétérophiles circulants. La réaction lymphoprolifère à la phytohémagglutinine s'est accrue; en revanche celle de la protéine dépurée a baissé. La migration des macrophages (Indice de migration) n'était pas affectée. Le titre d'anticorps n'a pas connu de changement significatif, tandis que le niveau de globuline gamma du sérum a diminué et le rapport albumine/globuline a augmenté. Le retard d'hypersensibilité a été également réduit et le taux de mortalité dû à l'exposition au virus vélogène virulent a augmenté chez les poulets ayant reçu une piqûre de spiramycine adipique.

Summary

The effect of spiramycin adipique in a dose of 50 mg/kg b.wt. (before, during or after vaccination) on the immune response to Newcastle disease virus vaccine (Lasota strain) was studied in 180 Hubbard chickens. This drug increased the lymphocyte count but decreased that of circulating heterophils and oesinophils. The lymphopreliferative response to phytohemagglutinin was increased while that of purified protein derivative was decreased. The migration of macrophages (Migration Index) was not affected. The antibody titre showed no significant change while the level of serum gamma globulin was decreased and the albumin/globulin ratio was increased. The delayed hypersensitivity was also reduced and the percentage of mortality due to challenge with virulant velogenic virus was increased in spiramycin adipique injected birds.

INTRODUCTION

Several antibiotics have been found to supress the immune response to specific antigens in man and animals[1,2,3,4 and 5].

Spiramycin is commonly used for treatment of mastitis in cattle, enzootic pneumonia in pigs and toxoplasmosis and foot rot in sheep[6,7,8,9 and 10]. It is also used for its potent antibacterial activity against *Mycoplasma gallisepticum* in chickens[11,12 and 13].

The effect of spiramycin on the immune system could not be traced in the available literatures therefore, the present work was designed to investigate the effect of spiramycin adipique which is commonly used in poultry production on the immune response to Newcastle disease virus vaccine in chickens.

Materials and Methods

Spiramycin adipique injectable pure powder was obtained from SPECIA RHONE, PARIS. It was freshly dissolved in normal saline solution to a concentration of 10% just before injection.

Chickens

One hundred and eighty, one day old Hubbard chicks were obtained from the General Poultry Company, Egypt and

*National Organization for Drug Control and Research, Giza, Egypt.

reared under strict hygenic measures till they were 42 days old. Chicks were fed standard ration, watered *ad lib* and received no medication or vaccination except those under investigation. One hundred and sixty birds were vaccinated (intraocularly at the age of 42 days) with Newcastle disease virus vaccine (Lasota strain). The other chickens were left non vaccinated non treated. The vaccinated birds were grouped into 4 equal groups. Chickens of the 1st group were injected with equal volume of sterile saline and kept as vaccinated control. The 2nd, 3rd and 4th groups were injected subcutaneously with spiramycin adipique in a dose of 50 mg/kg b.w.t. twice (48 hrs interval) before, after or before and after vaccination respectively.

Heparinized blood samples (3ml) were taken by the wing vein puncture of 10 chickens from each group before and 1, 2, 3, 4 and 5 weeks after vaccination to separate the mono-nuclear leucocytes[14]. The pripheral blood lymphocytes, heterophils and eosinophils counts were determined as explained by[15]. The macrophage migration inhibition (Migration Index) and the lymphoproliferative response to phytohemagglutinin — p (PHA) and purified protein derivative (PPD) were estimated as described by[16] and[17]. Another blood sample was taken from the wing vein of 5 chickens from each group, left to clot to obtain clear serum. The hemagglutination inhibition antibody titre, albumin, globulin and gamma globulin were determined in serum as described by[18] and[19]. The effect of spiramycin adipique on the delayed hypersensitivity to Newcastle disease virus vaccine was tested on 5 chickens from each group by intradermal injection of 1000 HA units of virulent virus into the wattle[20]. Moreover, the effect of spiramycin adipique on the percentage of protection against 1 locally isolated velogenic viscerotropic Vewcastle disease virus (titre 10/0.1 ml) was studied as explained by[21] on 20 vaccinated chickens from each group and 20 non vaccinated non treated chickens.

The obtained results were statistically analysed using the Student's "t" test.

Results

Administration of spiramycin adipique in chickens after or during vaccination increased the lymphocyte count. Insignificant changes in the lymphocyte count of chickens injected with the drug before vaccination. Heterophils and eosinophils counts were significantly decreased in all spiramycin treated chickens (table 1).

The number of tranformed lymphocytes of spiramycin injected chickens was increased in response to 10 ug/ml of phytohemagglutinin but it was decreased with 10 ug/ml of purified protein derivative. The migration index of macrophages was not changed in all spiramycin treated chickens (table 2).

Subcutaneous injection of spiramycin adipique in a dose of 50 mg/kg b.w.t. caused insignificant changes in the antibody titre in all treated chickens over the experimental period. The gamma globulin was significantly decreased in the *2nd*, *4th* and *5th* weeks after vaccination. The albumin/globulin ratio was increased (table 3).

Delayed hypersensitivity of chickens in response to 1000 hemagglutination units of purified concentrated Newcastle disease virus was decreased in spiramycin injected chickens. Prolonged effect was recorded in chickens receiving the drug before vaccination as compared with vaccinated non-treated control (table 4).

Administration of spiramycin adipique caused 10% mortality in chickens challenged with velogenic visceropic Newcastle disease virus corresponding to 5 and 100% in vaccinated and non-vaccinated non-treated chickens, respectively (table 4).

Discussion

Spiramycin adipique had no effect on the lymphocyte count when given before vaccination but increased their number when given after or during vaccination. However, it decreased the number of immunologically non specific effectors, heterophils and eosinophils in all trated birds. the effect of drugs on the immune response includes redistribution of cir-

Table 1: Effect of subcutaneous injection of spiramycin adipique (50mg/kg b.wt.) on lymphocytes, heterophils and eosinophils counts (%) of vaccinated chickens (n=10).

Administration of spiramycin	Parameters	Prevaccination level	Weeks after vaccination				
			1st	2nd	3rd	4th	5th
	Lymphocytes	48.4±1.12	61.4±0.68	66.4±0.24	69.6±0.51	64.0±0.45	63.8±0.49
	Heterophils	39.20±1.16	27.2±0.37	19.6±0.24	19.2±0.37	25.8±0.49	26.0±0.45
	Eosinophils	1.60±0.20	1.20±0.20	2.60±0.24	2.20±0.20	1.80±0.20	1.60±0.24
	Lymphocytes	48.4±0.81	60.2±0.58	66.6±0.40	70.8±0.37	65.4±0.68	65.4±0.81
before	Heterophils	38.8±0.97	28.2±0.87	17.8±0.66*	18.0±0.32*	23.8±0.37**	23.4±0.24***
vaccination	Eosinophils	1.80±0.20	1.20±0.20	1.40±0.24**	1.40±0.24*	1.60±0.24	1.40±0.24
	Lymphocytes	50.0±1.32	61.4±0.60	70.4±0.24***	72.0±0.63**	68.6±0.24***	68.2±0.50***
after	Heterophils	36.0±1.89	27.6±0.24	16.2±0.37***	15.8±0.66***	19.2±0.37**	19.2±0.37***
vaccination	Eosinophils	2.00±0.45	1.40±0.24	1.40±0.24**	1.80±0.20	2.20±0.20	2.20±0.20
before and	Lymphocytes	50.8±1.36	64.6±0.24**	69.4±0.24***	70.4±0.24	66.4±0.24***	66.4±0.24***
after	Heterophils	36.8±1.02	21.0±0.32***	17.0±0.55*	17.6±0.40**	21.2±0.37***	22.2±0.37***
vaccination	Eosinophils	1.80±0.20	1.20±0.20	2.80±0.37	1.60±0.24	1.60±0.24	1.60±0.24

**Significant at P<0.01 ***Significant at P<0.001

Table 2: Effect of subcutaneous injection of spiramycin adipique (50mg/kg b.wt.) on the proliferative response of lymphocytes in response to 10 ùg/ml of phytohemagglutinin (PHA) and purified protein derivative (PPD) and on the migration index (M.I.) of macrophages of vaccinated chickens in (n=5).

Administration of spiramycin	Parameters	Prevaccination level	Weeks after vaccination				
			1st	2nd	3rd	4th	5th
	PHA	13.50±0.44	21.67±0.48	51.17±0.37	44.00±0.37	37.83±0.21	33.67±0.22
	P.P.D.	11.42±0.15	20.25±0.28	21.42±0.40	15.67±0.38	12.83±0.30	11.67±0.26
	M.I.	33.94±1.48	38.59±3.57	36.26±2.09	31.09±3.80	28.48±3.48	21.97±5.47
	PHA	12.50±0.29	20.50±0.29	55.92±1.73*	50.67±0.33**	44.00±0.49***	42.17±0.24***
before	P.P.D.	10.67±0.56	16.67±0.56***	20.33±0.70	15.50±0.26	13.42±0.23	11.33±0.36
vaccination	M.I.	40.25±5.05	50.36±5.05	38.65±6.21	38.50±5.84	31.23±2.82	23.37±3.12
	PHA	11.75±0.78	19.75±0.88	59.42±0.31***	50.33±0.38***	44.83±0.44***	41.00±0.60***
vaccination	P.P.D.	11.00±0.21	19.17±0.27	21.00±0.39	16.00±0.49	12.83±0.37	12.35±0.22
	M.I.	38.64±4.32	34.93±2.95	35.82±1.47	40.72±5.42	31.14±6.99	21.87±7.04
before	PHA	12.83±0.37	20.50±0.31	58.33±0.38***	50.83±0.41***	45.50±0.23***	40.00±0.48***
and after	P.P.D.	10.50±0.38	18.75±0.39**	20.17±0.46	15.67±0.31	12.58±0.34	12.67±0.62
vaccination	M.I.	41.39±4.97	32.97±2.88	29.73±3.61	26.42±1.32	25.78±3.76	20.33±7.49

*Significant at P<0.05 **Significant at P<0.01 ***Significant at P<0.001

culating leucocytes[22] which may explain the effect of spiramycin on the leucocytes count.

The response of lymphocytes of spiramycin treated chickens to Phytohemagglutinin (PHA) was markedly increased. On the contrary the response of those lymphocytes to purified protein derivative (PPD) was reduced. These observations may indicate that both PHA and PPD stimulate different populations of lymphoid cells. Similar conclusion for the response to PHA and concanavalin-A was previously recorded in mouse thymic cells[23].

Spiramycin adipique markedly decreased the serum gamma globulin and increased the albumin/globulin ratio. This indicates that the drug may interfere the synthesis of immunoglobulin bearing

cells to purified protein derivative. In turkey, antibiotics have been shown to decrease the immunoglobulin bearing cells[24]. In this respect, interferance in protein or immunoglobulin synthesis, lowered phagocytosis and reduced exposure

Table 3: Effect of subcutaneous injection of spiramycin adipique (50mg/kg b.wt.) on hemagglutination inhibition antibody titre (HI), gamma globulin, albumen/globulin (A/g) ratio in serum of vaccinated chickens (n=5).

Administration of spiramycin	Parameters	Prevaccination level	Weeks after vaccination				
			1st	2nd	3rd	4th	5th
	HI titre	0	0	1.51±0.17	1.99±0.24	2.35±0.15	2.23±0.07
	gamma globulin (g)	0.33±0.03	–	0.67±0.05	–	0.47±0.01	0.73±0.01
	A/g ratio	0.89±0.03	–	0.89±0.01	–	1.04±0.004	0.84±0.03
before vaccination	HI titre	0	0	1.81±0.19	2.41±0.14	2.59±0.07	2.35±0.15
	gamma globulin (g)	0.40±0.01	–	0.28±0.05***	–	0.41±0.04	0.35±0.02**
	A/g ratio	0.84±0.01	–	1.10±0.04***	–	1.17±0.03***	1.07±0.05***
after vaccination	HI titre	0	0	1.87±0.15	2.29±0.15	2.47±0.18	2.35±0.11
	gamma globulin (g)	0.33±0.03	–	0.37±0.01***	–	0.4±0.01***	0.34±0.02***
	A/g ratio	0.89±0.03	–	0.91±0.03	–	0.99±0.01**	1.07±0.02***
before and after vaccination	HI titre	0	0	1.93±0.07	2.11±0.19	2.41±0.10	2.17±0.15
	gamma globulin (g)	0.34±0.04	–	0.32±0.02***	–	0.29±0.02***	0.43±0.01***
	A/g ratio	0.91±0.03	–	1.18±0.01***	–	1.25±0.07**	1.08±0.003***

**Significant at P<0.01 ***Significant at P<0.001

Table 4: Effect of subcutaneous injection of spiramycin adipique on the thickness of watties intradermally injected with 1000 HA units of velogenic viscerotropic virus and on the protection percent against virulent velogenic viscerotropic virus in chickens

Groups	Thickness of wattle in mm (n=5)							Protection (n=20)	
	hours after injection							Number	%
	3	6	9	12	15	16	24		
Non vaccinated control	–	–	–	–	–	–	–	0/20	0
Vaccinated control	2.40±0.10	2.70±0.04	2.00±0.10	1.80±0.10	1.70±0.10	1.70±0.10	1.60±0.10	19/20	95
Spiramycin before vaccination	2.30±0.02	2.00±0.10***	1.60±0.10*	1.40±0.10*	2.00±0.10	2.00±0.10	1.90±0.10	18/20	90
Spiramycin after vaccination	2.60±0.02	2.10±0.04***	1.70±0.10	2.00±0.10	1.40±0.20	1.50±0.10	1.60±0.10	18/20	90
Spiramycin before and after vaccination	2.50±0.10	1.90±0.20**	2.10±0.10	1.90±0.20	1.80±0.20	1.80±0.20	1.70±0.10	18/20	90

*Significant at P<0.05
**Significant at P<0.01
***Significant at P<0.001

to antigens have been indicated as possible mechanisms of immunosuppression of antibacterial drugs[2,25,3,26,27 and 28].

Insignificant changes in hemagglutination-inhibition antibody titre was observed in spiramycin-treated chickens when compared to vaccinated control. These findings agree with that obtained after administration of tylosin, gentamycin and chlorotetracycline in turkey[24].

Delayed hypersensitivity to intradermal injection of the virulent virus was reduced in spiramycin treated chickens. Moreover, challenge of vaccinated spiramycin treated birds with virulent viscerotropic Newcastle disease virus caused 10% mortality corresponding to 5% in vaccinated control. The present decrease in the delayed hypersensitivity correlates with the decrease in the number of circulating heterophils and eosinophils which are considered as non specific effectors involved in the delayed hypersensitivity[29 and 22].

It could be concluded that spiramycin adipique suppress the immune response to Newcastle disease virus vaccine in chickens and should be withdrawn during vaccination.

References

1. Stevens, K.M. (1953): *J. Immunol.* **71**:119.
2. Siegal, A.C.; Johnson, E.E. and Stollerman, G.H. (1961): *New England J. Med.*, **265**:266.
3. Weisberger, A.S.; Daniel, T.M. and Hoffman, A. (1964): *J. Expt. Med.*, **120**:183.
4. Panigraphy, B.; Grumbles, L.C.; Miller, D.; Naqi, S.A. and Hall C.F. (1979): *Avian Dis.*, **23 (2)**:401.
5. Tu, T.D. (1980): *Acta Veterinaria*, **28, (3)**:297.
6. Westmarch, E. (1969): *Finsk. Vet. Tidskr.*, **75**, 534.
7. Plonait, H. (1970): *Dt. Tierarztl. Wschr.*, **77**:473.
8. Du reuil, G. (1972): These Ecole National Veterinaire, Lyon. P. 139.
9. Nicolas, J.A. (1975): *Bulletin Mensuel de la Societe Veterinarire pratique de France*, **59 (7)**: 367-373.
10. Harper, T.S.; London, W.T. and Sever, J.L. (1985): *Am. J. Trop. Med. Hyg.*, **34 (1)**:50-57.
11. Ota, S.; Watnabe, S. and Kuniyasu, C. (1970): *Natn. Inst. Anim. Hlth; Qt., Tokyo*, **10**:1-10.
12. Lucas, J.H.S. and Injlis, J.M. (1970): *Caheirs Med. Vet.*, **39**:43-52.
13. Kuniyasu, G.; Yoshida, Y. and Takano, M. (1974):*National Institute of Animal Health Quarterly, Japan*, **14 (2)**:48-53.
14. Burrels, S. and Wells, P.W. (1977): *Res. Vet. Scl.*, **23**:84-86.
15. Hudson, L. and Hay, F.C. (1980): "Practical immunology", 2 *nd* ed., Blachwell Scientific publication, Oxford, London, Edinburgh, Boston, Melbourne.
16. Morita, C. Okada, T.; Izawa, H. and Soekawa, M. (1976): *Avian Dis.*, **20 (2)**:230-235.
17. Barta, O. and Oylkan, P.P. (1981): *Comp. Immun. Microbiol. Infect. Dis.*, **4 (2)**:209-221.
18. Bierer, B.W. (1961): *Am. J. Vet. Res.*, **30**:2237-2240.
19. MaxBrugh, Jr.; Beard, C.W. and Wilkes, W.J. (1977): *Avian Dis.*, **22 (2)**:320.
20. Cheville, N.F. and Beard, O.W. (1972): *Lab. Inves.*; **27**:129.
21. Reda, I.M. and Sheble, A. (1979): Isolation and characterization of local viscerotropic NDV. Personal communication.
22. Stities D.P.; Stobo, J.D.; Fudenberg, A.H. and Wells, J.V. (1982): "Basic and Clinical Immunology" 4th ed. Lauge Medical Publications, Los Altos, California; P. 227-314.
23. Jacobson, H. and Blomgren, H. (1974): *Cell. Immunol.* **11**, 427.
24. Cook, J.; Naqi, S.A.; Sahin, N. and Wagner, G. (1984): *Am. J. Vet. Res.* **45 (10)**:2189-2192.
25. Daniel, T.; Suhrland, L.G. and Weisberger, A.S. (1964); *J. Lab. Clin. Med.*, **64**:850-851.
26. Weisberger, A.S.; Moore, R.D. and Schoenberg, H.D. (1966): *J. Lab. Clin. Med.*, **67**:58-69.
27. Fineh, R. (1980): *J. Hyg. Epidemiol Microbiol Immunal.* **23**:220-225.
28. Hauser, W.E. and Remington, J.S. (1982): *Am. J. Trop. Med.*, **72**, 711-716.
29. Olah, I.; Kittner, Z.; Toro, I. and Glick, B. (1981): *Poult. Scin.* **60 (6)**:1321-1342.

Received for publication on 11th May 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 33-38

A CASE OF BOVINE THROMBOEMBOLIC MENINGOENCEPHALITIS ON TWO FARMS NEAR HARARE

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UN CAS DE MENINGO-ENCEPHALITE THROMBOEMBOLIQUE BOVINE DANS DEUX FERMES PRES DE HARARE

Résumé

Un syndrome qui ressemble du point de vue clinicopathologique à la méningo-encéphalite thromboembolique (TEME) est apparu dans deux fermes laitières près de Harare (Zimbabwe) et a fait l'objet de description. Il a été observé chez les veaux âgés de 3 à 6 mois, et les signes cliniques comprenaient la pyrexie, l'affaissement, la suffocation, l'ankylose, les troubles du système nerveux central, le fléchissement des boulets et la position couchée. La necropsie de quatre veaux a révélé la présence de la polysérite, la polyarthrite fibrineuse et la méningo-encéphalite avec la thrombose dans le cerveau. Il n'a pas été possible de cultiver d'organisme avec les prélèvements, probablement à cause d'un traitement à la pénicilline administrée auparavant, mais il semblerait que le syndrome soit dû à *Haemophilus somnus*.

Summary

A syndrome which clinicopathologically resembles thromboembolic meningoencephalitis (TEME) occurring in two dairy farms near Harare, Zimbabwe, is described. It was seen in calves between 3 and 6 months of age and signs included pyrexia, depression, respiratory embarassment, stiffness and central nervous system (CNS) disturbances, knuckling at the fetlocks and recumbency. Necropsy on 4 calves showed the presence of polyserositis, fibrinosus polyarthritis and meningoencephalitis with thrombosis in the brain. No significant organism was cultured from the samples presumably because of earlier penicillin treatment, but the syndrome is thought to have been caused by *Haemophilus somnus*.

INTRODUCTION

Thromboembolic meningoencephalitis (TEME) is a disease syndrome caused by *Haemophilus somnus* in cattle and is characterised by fever, septicaemia, respiratory distress, locomotor disturbances with the organism finally localizing in the central nervous system[1,2,3,4]. The disease has also been associated with weak calf syndrome (WCS)[5,6,7] and sleeper syndrome[8]. Other clinical signs described for the disease include reproductive disease including abortion and endometritis[9,10,11,12,13]. In male ruminants the organism has been associated with ovine epididymitis[14,15,17], seminal vesiculitis in bulls[18] and has been isolated from semen of a bull with severe epididymitis[16] and from a bull with purulent ejaculate[4]. Conjunctivitis[19] and polyarthritis[5,20] have also been described. The condition is reported to be commoner in feedlot than in dairy cattle on pasture[1,3,21,22,23] and is more common in winter than in summer[24].

The organism is a small pleomorphic Gram-negative coccobacillus which is a commensal of the bovine urogenital tract[4,25].

This paper describes clinical, gross and histopathological changes in four calves from two farms in Zimbabwe.

Case Report

Farm 1 was a dairy farm consisting mainly of the Jersey breed of cattle. Eleven calves were kept in an approximately

10 x 10 metre calf pen in which the dung and litter had not been removed for a considerable period of time. Younger calves were fed on fresh milk and concentrates while the older ones were on both concentrates and hay. Sporadic calf mortality was reported to have occurred previously for the last year.

At the time of the visit to the farm, the presenting clinical signs included pyrexia (39.8-40.5°C), anorexia, depression and recumbency. Some calves had limb stiffness and were reluctant to move while others showed knuckling at the fetlocks and later went down with paralysis. Although some calves were recumbent and depressed, others were recumbent but appeared bright while some had a sleepy appearance. Five of the calves had very rapid respiratory sounds with pleuritic rub. Odontoprisis associated with abdominal pain was a common finding. Nearly all of the affected calves had diarrhoea. The faeces was whitish-grey and foul smelling.

Three of the calves were taken to Large Animal Hospital of the Clinical Veterinary Studies Department, University of Zimbabwe. Because of the severe abdominal pain, an exploratory laporatomy was performed on one of the calves. This revealed a very severe, generalised fibrinous peritonitis with subserosal petechiae and ecchymoses along the walls of the intestines. She was later euthanised and autopsy performed immediately. The second one died the following day while the third one remained paralysed for about one month when she developed severe decubitous sores on her hind quaters. She could not stand on her own unless helped by an attendant (fig 1). The condition of this animal deteriorated and was euthanized and autopsied. It was later reported that four calves recovered following parenteral antibiotic treatment, and one had since died in the farm without autopsy being performed.

Farm 2 was another dairy farm 40 km away from farm 1 and had no managerial connection with the first one. The calf management practice were comparatively better here than in Farm 1. They were kept in separate, clean and dry pens with straw beddings. They were fed on milk and concentrates consisting of a mixture of ground maize and cotton seeds. A two-month-old Guernsey heifer calf had died earlier after a short illness. She had been described as having respiratory embarrassment and died despite parenteral antibiotic treatment. A week later, a 3-month-old calf died with the same clinical signs except that she had limb weakness. This ocurred despite antibiotic treatment and it was autopsied like the others.

Pathological findings

Post-mortem examination was carried out on all calves. Selected tissues were fixed in 10% formal saline, processed to paraffin blocks, cut 5μ thick and stained with Haematoxylin and Eosin. Pieces of liver and lung and swabs taken from the fibrinous exudate were plated on blood agar and MacConkey medium and incubated in O_2 and CO_2.

Pathological findings were similar in the 4 calves and these are summarised in the table 1. There was a fibrinous polyserositis with deposition of fibrinous exudate within abdominal and pleural cavities. Fibrinous exudate also occurred in the pericardial cavities of case 1, 2 and 4. Case 3 had a constrictive pericarditis characterised by diffuse fibrinous adhesions between the pericardium and epicardium. In case 4 fibrinous exudate was within the capsules of major limb joints. Meninges in cases 1, 2 and 4 were congested. Pneumonia occurred in cases 3 and 4, but the lungs in cases 1 and 2 were also congested and oedematous.

Histological sections of the brain showed meningitis and encephalitis characterised by infiltration of lymphocytes, plasma cells and polymorphonuclear neutrophilic leucocytes (Fig 2). Some blood vessels were thrombosed (Fig 3). Spinal cord was examined only in case 3 and it showed inflammatory reaction within the grey matter similar to that in the brain parenchyma. Heavy growth of *Escherichia coli* was obtained from most tissues and the inflammatory exudate. The cerebrospinal fluid had a marked

Fig. 1: Calf No. 3, with TEME paralysed in the hind quarters and showing decubitous sores (arrow).

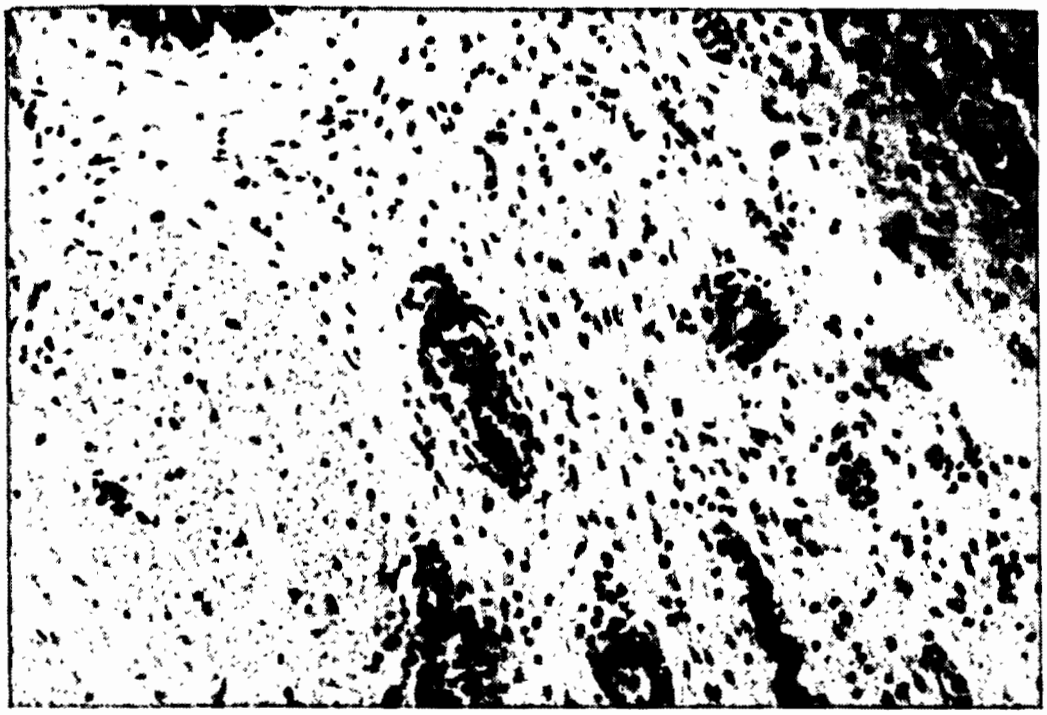

Fig. 2: A case of TEME. Section of brain showing inflammatory reaction characterised by infiltration of lymphocytes, plasma cells and neutrophyls. H & E X 360.

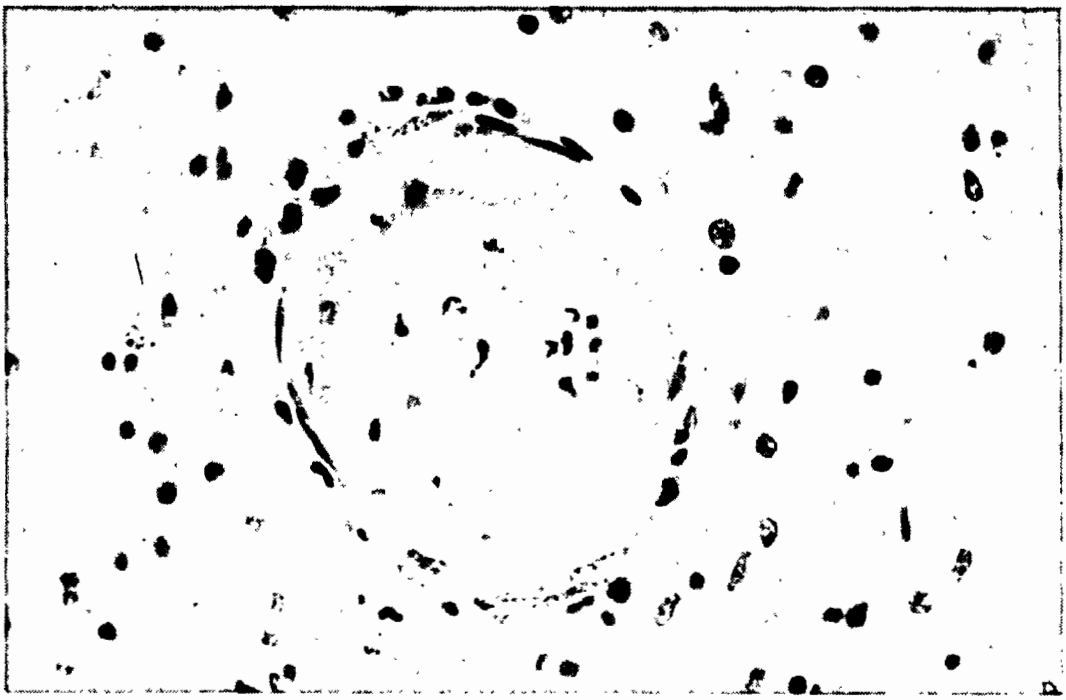

Fig. 3: A case of TEME. Section of the brain showing a thrombosed blood vessel. H & E X 720.

Table 1: Bovine Thromboembolic Meningoencephalitis. Principal lesions in 4 cases

Type of lesion	Presence (+) or absence (−) in cases 1 to 4			
	1	2	3	4
Peritonitis	+	+	+	+
Pleurisy	+	+	+	+
Pericarditis	+	+	+	+
Polyarthritis	−	−	−	+
Meningoencephalitis	+	+	+	+
Hydrocephalus	−	+	−	−
Pneumonia	−	−	+	+

increase in total protein and an increase in the number of neutrophils and glucose values were low.

Discussion

The clinical syndrome of fever, depression, anorexia, respiratory distress, polyarthritis, paralysis and sleepy appearance were similar to those described by Panciera et al[2], while post-mortem findings of haemorrhage, polyserositis, septicaemia, fibrinous peritonitis and pericarditis, pleurisy and fibrinopurulent polyarthritis and the histological changes of neutrophilic vasculitis with perivascular cuffing, meningitis and encephalitis with vascular thrombosis and infarction were similar to those described[2,7,8,26,27,28,29,30,31]. The increase in total protein and neutrophils with a decrease in glucose levels in the CSF were similar to those reported by others[32,33,34]. The clinicopathological findings in the cases suggest that these were cases of TEME presumably caused by *Haemophilus somnus*.

Although no *Haemophilus somnus* organism was isolated from samples submitted to the laboratory, William *et al.*[35] and Stephen *et al*[21] reported that failure to isolate the organism does not necessarily rule out the existence of the disease. It is also reported[22,31] that chances of isolating the organism are minimised after antibiotic treatment since the organism is very susceptible to

the commonly used antibiotics[24,36]. Our findings concur with the above authors as we were unable to isolate the organism from all the cases presented to us after antibiotic treatment. Little and Sorensen[25] recorn that limited success has been achieved in treating clinical cases of TEME; recovery, however, has been reported[1,35,37] in uncomplicated cases treated with antimicrobials. However, once neurological signs are advanced, treatment of such animals is generally unsuccessful[2,8]. Saunders et al[3] reported that TEME may affect animals between 7-9 months of age but that more cases were seen in calves less than 4-months-old. In our case, the calves were between 3 to 6 months old.

Recent transport and overcrowding have been incriminated as predisposing factors in the development of the disease[2,23,25,28] and that, Saunders et al[3] herds consisting of individuals from various sources may act as carriers and introduce the disease among a susceptible group. However, in our case, there was no history of recent transport except for the overcrowding in Farm 1. The farm is known to constantly swop adult animals with a sister farm belonging to the daughter and among which more than four cases of WCS have been diagnosed clinically. The source of the infection in these cases remain a mistery. Urinary excreation is known to be another method of disseminating the organism[3,29,35] and may represent a mode of transmission.

References

1. Kennedy, P.C., Biberstein, E.L., Howarth, J.A., Frazier, L.M. and Dungworth, D.L. (1960): *Am. J. Vet.* **21**:403-409.
2. Panciera, R.J., Dahlgren, R.R. and Rinker, H.B. (1968): *Path. Vet.* **5**:212-226.
3. Saunders, J.R. Thiessen, W.A. and Janzen, E.D. (1980): *Can Vet. J.* **21**:112-123.
4. Corboz, L. and Wild, P. (1981): *Elekrophorese. Schweizer Arch. Tierheilk* **123**:79-88.
5. Waldhalm, D.G., Hall, R.F., Meinershagen, W.A., Card, C.S. and Frank, F.W. (1974): *Am. J. Vet. Res.* **35**:1401-1403.
6. Ward, J. (1974): *Am. Ass. Bov. Pract. Proc.* **6**: 97-104.
7. Card, C.S. Spencer, G.R., Stauber, E.H., Frank, F.W., Hall, R.F. and Ward, A.C.S. (1975): *The Proceed. 77th Ann. Meet., U.S., Anim. Health, A,* **1973 (1974)**:67-72.
8. Baile, W.E., Anthony, H.D. and Weide, K.D. (1966): *J.A.V.M.A.* **148**:162-166.
9. Firehammer, B.D. (1959): *J.A.V.M.A.* **135**:421-422.
10. Chladek, D.W. (1975): *Am. J. Vet. Res.* **36**:1041.
11. Corboz, L. and Nicolet, J. (1975): *Schweizer, Arch, Tierheilk* **117**:493-502.
12. Van Dreumel, A.A. and Kiestead, M. (1975): *Can. Vet. J.* **16**:367-370.
13. Luginbuhl, A., Kupper, K. and Nicolet, J. (1981): *Schweizer Arch Tierheilk* **123**:629-637.
14. Ekdahl, M.O., Money, D.F.L. and Martin, C.A. (1968): *N.Z. Vet. J.* **16**:81-82.
15. Van Tonder, E.M. and Balton, T.W.F. (1968): *J.S. Afr. Vet. Med. Ass.* **39 (2)**:87-90.
16. Van Tonder, E.M. and Balton, T.W.F. (1970): *J.S. Afr. Vet. Med. Ass.* **41**:287-288.
17. Bulgin, M.S. and Anderson, B.C. (1983): *J. Am. Vet. Med. Ass.* **182**:372-374.
18. Jones, T.H., Barret, K.J., Greenham, L.W., Osborne, A.D. and Ashdown, R.R.C. (1964): *Vet. Rec.* **76**:24-38.
19. Lomont, H.H. and Hunt, B.W. (1982): *Vet. Rec.* **111**:21.
20. Stephens, L.R., Little, P.B., Wilkie, B.N. and Barnum, D.A. (1981a): *J. Am. Vet. Med. Ass.* **178**: 378-384.
21. Stephens, L.R., Little, P.B., Wilkie, B.N. and Barnum, D.A. (1981b): *Am. J. Vet. Res.* **42**:468-473.
22. Martin, S.W., Meek, A.H., Davis, D.G., Thomson, R.G., Johnson, J.A., Lopez, A., Stephens, L., Curtis, R.A., Prescott, J.F., Rosendal, S., Savin, M., Zubaidy, A.J., and Balton, M.R. (1980): *Can J. Comp. Med.* **44**:1-10.
23. Martin, S.W., Meek, A.H., Davis, D.G., Johnson, J.A. AND Curtis, R.A. (1981): *Can J. Comp. Med.* **45**:103-112.
24. Martin, S.W., Meek, A.H., Davis, D.G., Johnson, J.A. and Curtis, R.A. (1982): *Can. J. Comp. Med.* **46**:341-349.
25. Little, P.B. and Sorensen, D.K. (1969): *J. Am. Vet. Med. Ass.* **155**:1892-1903.
26. Humphrey, J.D., Little, P.B., Stephens, L.R., Barnum, D.A., Doig, P.A. and Thorsen, J. (1982a): *Am. J. Vet. Res.* **43**:791-795.
27. Griner, L.A., Jensen, R. and Brown, W.W. (1956). *J. Am. Vet. Med. Ass.* **129**:417-421.
28. Olander, H.J., Gallina, A.M., Beckwith, D. and Marrow, M. (1970): *Proc. U.S. Anim. Hlth. Ass.* **74**:589-600.
29. Brown, L.N., Dillman, R.C. and Dierks, R.E. (1970a): *Proc. U.S. Anim. Hlth. Ass.* **74**:94-108.
30. Smith, P.B. and Biberstein, E.L. (1977): *Cornell. Vet.* **67**:300-305.
31. Keister, D.M. (1981): *The Compend. Contin. Educ. Pract. Vet.* **3**: **(7)**, S260-S267.
32. Clausen, H.H., Eigenmann, U.J.E., Amtsberg, G., Martens, J. and Meier, K. (1980): *Schweizer Arch. Tierheilk* **122**:661-672.
33. Howard, J. and Fawcett, K. (1966): *Iowa St. Univ. Vet.* **28**:101-105.

34. Eigenmann, U.J., Clausen, H.H., Martens, J. and Stober, M. (1980): *Dt. Tierarztl. Wschr.* **87**: 117-120.
35. Williams, J.M., Smith, G.L. and Murdock, F.M. (1978): *Am. J. Vet. Res.* **39**:1756-1762.
36. Garcia-Delgado, G.A., Little, P.B. and Barnum, D.A. (1977): *Can. J. Comp. Med.* **41**:380-388.
37. Case, M.T., Raubabaugh, W.G. and Finnell, J.H. (1965): *Illinois Vet.* **8 (2)**:31-32.

Received for publication on 22nd April 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 39-43

A CASE OF BOVINE HEMORRHAGIC SEPTICAEMIA IN KENYA

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UN CAS DE SEPTICEMIE HEMORRAGIQUE BOVINE AU KENYA

Résumé

Une enquête à été conduite pour déterminer la cause de mort subite d'un veau Frison de souche dans une ferme à Limuru dans le centre du Kenya. Les conclusions macroscopiques de la nécropsie ont révélé une hémorragie pétéchiale grave sur la muqueuse trachéale au niveau du troisième anneau supérieur et une hémorragie qui baissait en intensité à l'embranchement de la trachée. On a observé à l'aide d'examens macroscopiques et microscopiques un oedème étendu des poumons et des ganglions lymphatiques, et une hyperémie des poumons. Du point de vue histologique, il y avait une dégénérescence nécrotique grave sur la muqueuse de la trachée. On a également remarqué l'oedème dans le foie, les reins et le cerveau. L'examen bactériologique de la trachée et les prélèvements de ganglion lymphathique des bronches ont révélé la présence de *Pasteurella multocida*. Les examens virologiques des prélèvements obtenus des animaux morts et des animaux en bonne santé vivant à la ferme n'ont pas confirmé la présence du virus.

Summary

An investigation was carried out into the cause of sudden death of a high grade Friesian calf on a farm located in Limuru area of central Kenya. The gross post-mortem findings included marked petechial hemorrhages on the upper third of the tracheal mucosa, decreasing in intensity down to the tracheal bifurcation. Extensive oedema of the lungs and lymph nodes, and hyperaemia of the lungs were observed both grossly and microscopically. Histologically, there was severe necrotic degeneration on the mucosa of the trachea. Oedema was observed in the liver, kidneys and the brain. Bacteriological examination of the trachea and bronchial lymph node samples revealed the presence of *Pasteurella multocida*. Virological tests on samples obtained from the dead animal and from other healthy animals on the farm did not show evidence of the presence of virus.

INTRODUCTION

Hemorrhagic septicaemia is a form of Pasteurellosis caused by *Pasteurella multocida* and charactrized by acute septicaemia and high mortality in cattle, buffalo and pigs[1]. It is a disease of particular importance in the tropics. In Nigeria, the disease is prevalent during the rainy season especially in marshland and swampy places, and is often associated with heavy rainfall and marked drop in atmospheric temperature[2,3]. There exists scanty in formation on the pathology of the disease in the veterinary literature[1]. And information concerning the histological changes on the tracheal mucosa of affected cattle could not be found in the available literature. In Kenya, some protozoan diseases may give a pathological picture similar to hemorrhagic septicaemia[4]. In this report, a record is made of the results of the pathological and microbiological investigations on a calf which died suddenly on a cold rainy night.

Materials and Methods

A post-mortem examination was carried out on a ten months old female Friesian calf, found dead on a cold wet morning. The animal had been observed healthy, active and eating well the previous evening. There had been considerably heavy rainfall in the night after a long dry period. Blood in EDTA, ocular and nasal swabbs were taken from 16 cattle

on the farm for virological examination. Two of the 16 cattle had previously been treated with antibiotics for clinical pneumonia and had recovered.

Results

Macroscopic lesions

The animal which was in very good body condition was found lying on sternal recumbency with a protruding rectum and hind limbs flexed as if it had died while attempting to stand up. Clear froth was oozing from the nostrils but on turning the animal, this became blood-tinged fluid. The other body orifices appeared to be normal. The carcass appeared fresh at the time of post-mortem examination.

An area of hemorrhage and oedema, measuring approximately 10 x 15 cm was found in the sternal region affecting the subcutaneous tissues and extending deep into the underlying muscles.

The thoracic cavity contained approximately 200 mls of blood-tinged fluid with fibrinous flecks. Fibrous tags, some of which were hemorrhagic, formed adhesions between the parietal and visceral pleurae. The lungs were heavy, characterized by oedema and hyperaemia. Marked petechial hemorrhages were observed on the tracheal mucosa affecting more extensively the upper part of the trachea, especially around the larynx and upper third of the tracheal mucosa and becoming less towards the tracheal bifurcation where congestion became more marked. A large amount of blood-tingered fluid was found in the lumen of the trachea. There was an increase in blood-tinged fluid in the pericardial sac. Petechial hemorrhages were present on the epicardium.

Prescapular and bronchial lymph nodes were hemorrhagic and oedematous. The spleen was marshy in consistency and enlarged. The liver appeared to have a uniform greyish colour and the gall bladder contained very little yellowish bile. The brain was softer in consistency and the gyri appeared wider. No other gross lesions were observed.

Microscopic lesions

Tissues were fixed in 10% formalin and Bouin's fixatives and stained with H & E. Hyperaemia, oedema and hemorrhage were observed in the lung, affecting all the lobes. The alveoli were filled with fluid and occasionally, macrophages with large eosinophilic cytoplasm were observed immersed in the protein rich fluid (Fig. 1). The visceral pleura was infiltrated with mononuclear cells mixed with fibrous tissue. Coagulative necrosis affected the mucosa of the trachea which also showed patches of mononuclear cell infiltration (Fig. 2). Oedema was observed in the kidneys, liver, and brain (Fig. 3) characterized by fluid in the Bowman's space, between hepatocytes and sinusoids and in the perivascular spaces (the Virchow-Robin spaces) respectively.

The lymph nodes and spleen showed lack of cellular density in both the cortex and paracortex of the lymph nodes and periarterial lymphoid sheaths of the spleen. There was an apparent lack of the red pulp in the spleen. Fibrinous fluid was present in some parts while in others, mononuclear cells appeared scattered in between clear spaces. Most of the mononuclear cells were lymphocytes and plasma cells. In the heart, some myocardial fibres contained sarcocystis cysts.

Bacteriological examination

Bacteriological examination was carried out by inoculation onto blood agar, samples of trachea, spleen, bronchial and prescapular lymph nodes and fluid content of the trachea. There was no bacterial growth from lung and spleen samples. Small pin-point, non-hemolytic bacterial colonies with smooth surface were observed on blood agar, 24 hours following inoculation with tracheal and bronchial lymph node samples and aerobic incubation at 37°C. With gram's stain, the smears prepared were gram negative and with methylene blue stains, they showed bipolar staining. Biochemical tests[1] characterized the organism as *Pasterurella multocida*. Staphylococci and B-hemolytic streptococci were isolated from the fluid contents of the trachea.

Glass slide smears

Glass slide smears prepared from heart blood and impression smears prepared

Fig. 1: Lung x 100. This shows oedema fluid in the alveoli and respiratory bronchioles (arrows).

Fig. 3: Brain x 100. Oedema fluid can be seen in the distended virchow-Robin spaces (arrow).

Fig. 2: Tracheal Mucosa showing necrotizing tracheitis x 100. The mucosa shows necrosis (arrow) and mononuclear cell infiltration (double arrows).

from bronchial and prescapular lymph nodes, brain and spleen did not show any changes associated with parasitic disease.

Virological examination

A wide range of samples which was collected from the dead animal and including blood in EDTA, nasal, vaginal and ocular secretions from 14 healthy cows on the farm were examined virologically by inoculation onto primary bovine kidney cell monolayer cultures. The cultures were incubated at 37°C in roller tubes and examined for 14 days for evidence of virus cytopathic effects. No virus was demonstrated in any of the samples examined.

Discussion

The occurrence of hemorrhagic septiceamia in Kenya was reported by Shirlaw,[4], affecting animals of less than 2 years of age and of breeds that have been imported into the country. The case reported here was observed in the Limuru area and falls within the age limit and definition of the type of animals which according to Shirlaw[4] are affected by this disease.

The same author also reported the relative absence of epizootics of hemorrhagic septicamia in Kenyan cattle and observed that the few outbreaks did not appear to be influenced by climatic variation. This is in contrast to reports from other tropical countries. In Nigeria for example, where the disease is prevalent in marshy areas during the rainy season, outbreaks were often associated with heavy rainfall and a marked drop in atmospheric temperature[2]. The animal reported here died on a night of heavy rainfall which occurred following a long dry period.

This would appear to agree with the observations made in Nigeria, implying that in Kenya, like in other tropical countries, the occurrence of hemorrhagic septicaemia may be influenced by climatic variations.

The lesions observed in this study, mainly consisting of petechiation of the epicardium and the mucosa of the upper respiratory system, excess blood-tinged fluid in the pericardial sac and thoracic cavity, oedema and hyperaemia of the lungs are similar to the lesions described by Losos[1] for hemorrhagic septicaemia. The widely distributed signs of vascular disturbance in the form of oedema, hyperaemia and hemorrhages are similar to the lesions observed in a calf which died following admistration of endotoxin prepared from *pasteurella multocida* reported by Rhoades, Heddlestone and Rebers[5]. These lesions are compatible with changes associated with shock. The calf administered with endotoxin prepared from *Pasteurella multocida* died 4½ hours later[6]. Endotoxin shock would therefore explain the sudden death of the calf reported here which was seen to be healthy in the evening and found dead the following morning. Interstitial pneumonia, which was observed in calves with septicaemic hemorrhagic pasteurellosis[5] was not found in the present case. This suggest that endotoxin rather than septicaemia caused the pathological changes observed in the present case. Careful examination of the carcass assisted in eliminating lightning as a possible cause of the sudden death.

Hemorrhagic septicaemia may show lesions similar to those of some protozoan infections[4] and unless careful and detailed examination is carried out, there is a possibility of a misdiagnosis. Blood smears and glasslide impression smears of the lymph node and spleen were found negative for protozoan infections. The possibility of viral infection was investigated and found negative. IBR-virus as well as P1-3- virus infections may help facilitate the establishment of *Pasteurella hemolytica* in the lung in appropriate amounts to induce fibrinous pneumonia[7]. In the absence of viral infection, the necrotising tracheitis was considered to have been caused by the bacteria which were isolated.

Tissue cyst-forming Eimerid coccidia, of which Sarcocystis is one, are known to cause disease in cattle in the acute stage of the infection (Tadros and Laarman)[8]. The Sarcocystis cyst found in the myocardium of the present case represents the

chronic stage and can thus be removed from the discussion.

Pasteurellosis is common among goats and sheep in Kenya[9,10] and vaccination is practiced on some farms. Recommendation for vaccination of cattle breeds introduced into Kenya in the recent times would appear appropriate, especially in those areas where hemorrhagic septicaemia has been reported.

Acknowledgements

We are grateful to Proffessor H-J. Hansen of SIDA for useful advice in drafting this paper and to B. Oduor, H. Ogutu and J. Ngaho for technical assistance. This paper is published with the kind permission of the Director, National Veterinary Research Centre, Muguga, Kenya Agricultural Research Institute.

References

1. Losos, G.J. 1986. In *Infectious tropical diseases of domestic animals* 1st. Ed. Longman.
2. Kasali, O.B. 1972. *Bull. epiz. Dis. Afri.* **20**, 203-204.
3. Anosa N.O. and T.T. Isoun. 1975 *Bull. Anim. Hlth Prodl Afr.* **23**, 337-340.
4. Shirlaw, J.F. 1957 *Br. Vet. J.* **113**, 35-46.
5. Rhoades, K.R., K.L. Heddlestone and P.A. Rebers
6. Rebers. P.A.; K.L. Heddlestone and K.R. Rhoades. 1967. *J. Bact.* **93**, 7-14.
7. Rehmutulla, A.J. and R.G. Thomson. 1981 *Canadian Vet. Journal.* **22 (1)**, 1-18.
8. Tadros, W. and J.J. Laarman. 1982. Advances in *Parasitology.*, **20.**
9. Mugera, G.M. and T.T. Kramer. 1967. *Bull epizoot. Dis. Afri.* **15**, 125-131.
10. Wandera, J.G. 1967. *Bull epizoot. Dis. Afri.* **15**, 245-258.

Received for publication on 21st April 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 45-49

ROLE OF SOME PASSERIFORMES BIRDS IN TRANSMISSION OF NEWCASTLE DISEASE. I. SUSCEPTIBILITY OF SOME WILD BIRDS OF SUDAN TO NEWCASTLE DISEASE VIRUS*

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ROLE DE CERTAINS OISEAUX PASSEREAUX DANS LA TRANSMISSION DE LA MALADIE DE NEWCASTLE I. SENSIBILITE DE CERTAINS OISEAUX SAUVAGES DU SOUDAN AU VIRUS DE LA MALADIE DE NEWCASTLE

Résumé

On n'a détecté ni le virus de la maladie de Newcastle ni des anticorps chez des oiseaux sauvages apparemment en bonne santé. Cinq espèces d'oiseaux sauvages ont été inoculées au virus de la maladie de Newcastle vélogène et viscérotropique. Lea alouettes *(Euplectes orix)* et les pinsons *(Lagonostica senegala)* étaient les plus sensibles, avec un taux de mortalité de 100%. Les moineaux francs du Soudan *(Passer domesticus arborius)* et les moineaux dorés du Soudan *(Auripasser Lutea)* avaient un taux de mortalité de 62,5%. Les tisserins *(Ploceus spp.)* étaient infectés mais avaient de légers symptômes cliniques et un taux de mortalité de 12,5%. Les signes cliniques révélaient que le système nerveux et l'appareil digestif étaient atteints. Des lésions hémorragiques dans le proventricule ont été observées chez les cinq espèces d'oiseaux. Certains oiseaux infectés puis guéris avaient des niveaux élevés d'anticorps qui entrainaient l'inhibition de l'hémagglutination.

Summary

Neither Newcastle disease virus was nor antibodies were detected in apparently healthy wild birds. Five species of wild birds were inoculated with velogenic viscerotropic newcastle disease virus. Durra birds *(Euplectes orix)* and rosy fire finches *(Lagonostica senegala)* were the most susceptible with a 100% mortality. Sudan house sparrows *(Passer domestic arborius)* and Sudan golden sparrows *(Auripasser Lutea)* suffered a mortality of 62.5%. Weaver birds *(Ploceus spp.)* became infected with relatively mild clinical symptoms and had a mortality rate of 12.5%. Clinical symptoms indicated the involvement of the nervous and digestive systems. Haemorrhagic lesions in the proventriculus were seen inbirds of the five species. Some infected and recovered birds had high levels of serum haemagglutination inhibiting antibodies.

INTRODUCTION

The isolation from wild birds of paramyxoviruses, Influenza A, Herpes, Reo, Pox and Adeno viruses, has given evidence to the suspicion that birds act as reservoirs of viral agents more than was previously suggested[1,2,3,4,5,6].

Newcastle disease virus (NDV) has a wide host range of birds. In addition to domestic fowls, turkeys and guinea fowls, numerous other avian species were reported to be either naturally or experimantally susceptible[7,8,9,10].

In Sudan, besides domestic fowls, NDV has only been isolated from naturally infected pigeons[11]. In Khartoum area with the methods of poultry husbandry where open houses are used, wild birds are usually seen incontact with domestic fowls. This observation stimulated this study with the aim to investigate the susceptibility of five passeriformes species birds to NDV in order to determine the possible role they may play in the transmission of the disease.

Materials and Methods

Newcastle Disease Virus

The Kh. S-1 was isolated from 28-day-old chicks showing nervous signs, diarrhoea and haemorrhages in the proventriculus. The agent haemaggluti-

*Part of a thesis submitted by the first author for M.V.Sc. degree in 1987 to the University of Khartoum.

nated chicken erythrocytes and its haemagglutinability was inhibited by antisera prepared against komarov strain of NDV. Kh. S-1 isolate grew readily in embryonating chicken eggs to a litre of $10^{9.5}$CELD 50% and infected embryos died with haemorrhages in various organs.

The pathogenic properties of Kh. S-1 including $CELD_{50}$, mean death time (MDT), intracerebral pathogencity to 8-week-old chickens were previously determined and were compared to Herts 33/56 strain[12].

*Wild Birds**

Five passeriformes species of Sudan wild birds (Sudan house sparrows *Passer domesticus arborius),* Sudan golden sparrows *(Auripasser lutea),* Durra birds *(Euplectes orix),* Rosy fire finches *(Lagonostica senegala)* and Weaver birds *(Ploceus spp.)* were selected because they were the most frequent visitors to poultry houses. They were also observed to be very gregarious and often mixed in intraspecies groupings. Sudan house sparrows and rosy fire finches were frequently seen inside poultry houses in large groups, while Sudan golden sparrows, durra birds, and weaver birds were rarely seen inside poultry houses.

Wild birds were collected from the premises of three farms, namely University of Khartoum farm, Almassara poultry farm and a small private farm near the Faculty of Veterinary Science.

All wild birds used in this study were captured by mistnets in the period between *May 1985* and *August 1985.* Mistnets were placed at the side of the poultry houses opposite to partitions between pens, where food was usually present in large quantities and attracted wild birds.

Birds were sorted out and grouped into species. Each group was transferred to a small cage (1 cubic meter) and supplied with commercial poultry feed and leftovers of vegetables and fruits and were allowed water ad. lib. Birds were kept under observation for 15 days before use. Wild birds were used for two purposes.

*Identified by Dr. Dawi Musa Hamed, Sudan Natural History Museum, University of Khartoum.

1. Detection of NDV or Antibodies.

A total of 160 wild birds were used in an attempt to isolate NDV from spleens and also to detect the presence of antibodies against NDV in sera of these birds. Birds were killed by severing the jugular vein with a pair of scissors. Spleens (pooled samples of each three spleens) were aseptically collected. Phosphate buffered saline containing penicillin (200 i.u/ml) was added and samples were then homogenized and a few drops of PBS were added to make approximately 10% suspension. The suspensions were centrifuged and the supernatent fluids used for inoculation of embryonated eggs via the allantoic cavity. Each embryo received 0.2 ml of inoculum. Embryonated eggs were candled daily and Amnioallantoic fluids (AAF) from dead embryos and those still alive 5 days post-inoculation were collected and examined for the virus by haemagglutination (HA) and haemagglutination inhibition (HI) tests using standard techniques[13]. For antibody detection, pooled blood samples from groups of three birds were collected in petri dishes and left for half an hour on the bench to clot. The clot was then cut by a scalpel and samples were refrigerated at 4°C overnight. Sera were then separated, inactivated for 30 minutes at 56°C and stored at -20°C.

2. Determination of Susceptibility.

Five species groups were inoculated intra-nasally (I/N) with Kh. S-1 isolate of NDV to study their susceptibility as is shown in Table 1. Two birds were separately caged as controls for each species group. Wild birds of each group were separately caged, supplied with food and water and observed daily. Birds that died or survived the duration of the experiment and sacrificed were autopsied and examined for macroscopic lesions. Brains, lungs, spleens, and livers were aseptically collected. Phosphate buffered saline containing penicillin, streptomycin and gentamycin was added and samples were stored at -20°C. Prior to inoculation of embryonated eggs, tissue samples were homogenized, few drops of PBS were added to make approximately 10%

Table 1: Species of Birds used in the Determination of Susceptibility of Wild Birds to NDV, No. and Dose of Exposure

Species	No. of Birds Used	Dose (CELD 50%)
Sudan house sparrows	10	$10^{7.5}$
Weaver spp.	9	$10^{7.5}$
Rosy fire finches	6	$5 \times 10^{6.5}$
Durra birds	8	$5 \times 10^{6.5}$

suspensions, centrifuged and the supernatant fluids used for inoculation. Each sample was inoculated into two embryonated eggs via the allantoic cavity. Each embryo received 0.2 ml of inoculum. Embryonated eggs were candled daily and AAF from dead embryos and those still alive 5 days (p.i.) were collected and examined for the presence of the virus using HA and HI tests.

Blood samples were also collected from birds that survived and were killed at the end of the experiment, sera were separated, inactivated, stored at -20°C and examined using HI test.

Results

Isolation Attempts and Detection of Antibodies in Wild Birds

Newcastle disease virus was not isolated from spleens of the 160 wild birds during the period of study. Haemagglutination inhibiting antibodies against NDV were not demonstrated in the sera of these birds.

Susceptibility of Wild Bird Species to Experimental NDV Infection

Four birds (2 house sparrows and 2 golden sparrows) died 1-3 days p.i. They did not show any clinical symptoms prior to death.

Sudan house sparrows, Sudan golden sparrows, weaver birds, rosy fire finches and durra birds, experimentally inoculated with Kh. S-1 isolate, developed clinical symptoms 5-15 days p.i. Clinical symptoms were first observed as depression and weakness of legs and wings. Except for weaver birds, clinical symptoms progressed to incoordination, paralysis of various degrees and diarrhoea in most of the sick birds. Paralysis was either unilateral or bilateral as was indicated by wing drop and/or leg paralysis and by inability of birds to fly or walk. Table 2 shows the effect of Kh. S-1 isolate on the five species of wild birds. Birds with leg or wing paralysis died within 1-16 days after the onset of symptoms. One Sudan house sparrow and 2 Sudan golden sparrows, survived the duration of the experiment with unilateral leg paralysis. One Sudan golden sparrow and one weaver recovered by days 21 and 29 postinoculation, respectively.

The prominent gross pathological lesions observed on necropsy were haemorrhages in the proventriculus of some birds of the five wild bird species. Newcastle disease virus was reisolated from five Sudan house sparrows, four

Table 2: Effect of Kh.S-1 Isolate of NDV on Five Species of Wild Birds

Birds	Incubation period in days	Morbidity rate infected/ exposed	Crude death died/ exposed	Cause – specific death/rate died/ exposed
Sudan house sparrows	11-13	6/10	7/10	5/10
Sudan golden sparrows	14-15	8/10	7/10	5/10
Weaver birds	14-15	8/9	2/9	1/9
Rosy fire finches	5-6	6/6	6/6	6/6
Durra birds	7-8	8/8	8/8	8/8

*Crude death rate: Deaths occuring in the duration of the experiment/total No. exposed.
**Cause-specific death: Deaths related to Newcastle disease infection/total No. exposed.

Table 3: Results of NDV Reisolations from 5 Species of Wild Birds

Birds	No. of birds	Days until death or killing of birds	Brain (S)	Spleen-liver (pool)	Lung (S)
Sudan house sparrows	2	2	–	–	–
	1	13	+	+	++
	2	14	+	+	+
	2	16	+	–	–
	3	47*	–	–	–
Sudan golden sparrows	1	1	ND.	ND	ND
	1	3	–	–	–
	1	15	+	–	+
	3	16	+	–	+
	1	18	ND	ND	ND
	3	35*	–	–	–
Weaver birds	1	8	ND	ND	ND
	1	27	–	–	–
	3	54*	–	–	–
	4	65*	–	–	–
Rosy fire finches	2	6	+	+	+
	3	7	+	+	+
	1	8	+	–	+
Dura birds	1	6	+	+	+
	2	8	+	–	+
	1	10	+	–	–
	1	13	–	–	–
	3	15	–	–	–

*Killed
ND Not done

Sudan golden sparrows, six rosy fire finches, four durra birds, but not from weaver birds. Reisolations were made from the brains, lungs and pools of spleen and liver (Table 3).

Antibodies against NDV were demonstrated in pooled sera of three Sudan house sparrows, three golden sparrows and seven weaver birds that survived till the end of the experiment and gave HI titres of 6, 7 and 7 ($\log_2$), respectively.

Controls did not show any signs of infection nor was the virus isolated from them. Surviving controls had no detectable anti-NDV antibodies.

Discussion

The Kh. S-1 isolate of NDV produced an infection in all the five wild bird species comparable to that in chickens. There were however, minor variations within the species in susceptibility to infection. Durra birds and rosy fire finches seemed to be the most susceptible. They became infected within 5-8 days after inoculation and exhibited the highest mortality rate. Sudan house sparrows and Sudan golden sparrows became infected within two weeks and suffered a lesser mortality. Weaver birds became infected with relatively milder clinical symptoms and had a mortality rate of 12.5%.

Clinical responses varied and consisted of nervous involvement with diarrhoea in some birds. Nervous signs appeared to be the marked feature of infection with velogenic viscerotropic NDV in wild birds.

Haemorrhagic visceral lesions that were observed in birds of the five species differed from those seen in chickens. These lesions were restricted to the proventriculus in wild birds, whereas haemorrhages in chickens were usually

present in the proventiculus and ventriculus and in the upper and lower intestinal tract.

Experimental susceptibility of house sparrows to NDV obtained in this study corroborated the findings of Chelidze[14] and Gustafson and Moses[15,16] who succeeded in infecting sparrows by aerosolization or through intramuscular (I/M) and intranasal (I/N) inoculation and recovered the virus from the liver, lung, brain and intestinal contents of the experimentally infected birds.

However, these results disagree with those of Abdel Magid, Abdel Aziz and El Nassri[17], and Popvic[18], who failed to infect sparrows with NDV. This discripancy may be due to the virus strains used, the dose or the experimental circumstances. Gustafson and Moses[15,16] used a viscerotropic velogenic strain of NDV, while others did not mention the type of the virus used in their experiments. Abdel Magid *et al.*[17] could not detect HI-antibodies in sera of orally inoculated sparrows.

Susceptibility of weaver birds to experimental infection with NDV had been reported by Vickers and Hanson 1979[19] while susceptibility of Sudan golden sparrows, durra birds and rosy fire finches is reported in this study for the first time.

The failure to isolate NDV and to detect antibodies against it in wild birds during this study is probably because no outbreak of NDV occured in the vicinity before or during the investigation.

The ability of the Kh. S-1 isolate of NDV to establish infection in 5 species of Sudan wild birds suggests that these birds may be naturally susceptible to the virus and hence constitute an imprtant reservoir or vehicle of the virus.

References

1. Bahl, A.K., Pomeroy, B.S., Madigunedimedjo, S. and Easterday, B.C. (1977). *J. Am. Vet. Med. Ass.* **171**, 949.
2. Annuar, B.O., Mackenzie, J.S. and Lalor, P.A. (1983). *Arch. Virol.* **76**, 217.
3. Senne, D.A., Pearson, J.E., Miller, I.D. and Gustafson, G.A. (1983). *Avian Dis.* **27**, 731.
4. Tumova, B., Turek, R., Kubinova, I., Stumpa, A. and Clampor, F. (1984). *Acta Virol.* **28**, 114.
5. Mackenzie, J.S., Edwards, E.C., Holmes, R.M. and Hinshaw, V.S. (1984). *Auot. J. Exp. Biol. Med. Sci.*, **62**, 89.
6. Otsuki, K., Kariya, H., Matsuo, K., Sugiyama, S., Hoshina, K., Yoshikane, T., Matsumoto, A. and Tsubokura, M. (1987). *Jap. J. Vet. Sci.* **4**, 721.
7. Lancaster, J.E. and Alexander, D.J. (1975). Newcastle disease. Virus and spread. Ottawa, Canada Department of Agriculture. Monograph No. II.
8. Chu, H.P., Trow, E.W., Greenwood, A.G., Jenning, A.R. and Keymer, I.F. (1976). *Avian Path.* **3**, 227.
9. Shortridge, K.F., Alexander, D.J., Hu, L.Y. and Kam, S.L. (1978). *J. Comp. Path.* **88**, 633.
10. Manjunath, L.H. and Mallick, B.B. (1981). *Indian J. Anim. Sci.* **11**, 1116.
11. Eisa, Mand Omer, E.A. (1984). *Vet. Rec.* **114**, 297.
12. Ismail, M.H. (1987). M.V.Sc. Thesis. University of Khartoum.
13. Allan, W.H., Lancaster, J.E. and Toth, B. (1978). Newcastle disease vaccines. Their production and use. Rome, Food and Agricultural Organization. Rome.
14. Chelidze, G.T. (1964). *Vopr. Vet. Virus. 1, 395. (Cit. Vet. Bull.* **34**, 597, 1964).
15. Gustafson, D.P. and Moses, H.E. (1952). *Am. J. Vet. Res.* **13**, 566.
16. Gustafson, D.P. and Moses, H.E. (1953). *Am. J. Vet. Res.* **14**, 581.
17. Abdel Magid, I.M., Abdel Aziz, M.A. and El Nassri, B.B. (1965). *J. Arab. Vet. Med.* **25**, 193.
18. Popvic, B. (1951). *Acta Vet. Belgr. 1, 168 (Cit. Vet. Bull.* **23**, 414, 1953).
19. Vickers, M.L. and Hanson, R.P. (1979). *Avian. Dis.* **1**, 70.

Received for publication on 30th May 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 51-54

ROLE OF SOME PASSERIFORMES BIRDS IN TRANSMISSION OF NEWCASTLE DISEASE. II. PATHOGENESIS OF NEWCASTLE DISEASE VIRUS IN SUDAN HOUSE SPARROWS *(PASSER DOMESTICUS ARBORIUS)*

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ROLE DE CERTAINS OISEAUX PASSEREAUX DANS LA TRANSMISSION DE LA MALADIE DE NEWCASTLE II. PATHOGENESE DU VIRUS DE LA MALADIE DE NEWCASTLE CHEZ LES MOINEAUX FRANCS DU SOUDAN *(PASSER DOMESTICUS ARBORIUS)*

Résumé

Des moineaux francs du Soudan *(Passer domesticus arborius)* furent inoculés au VMN (Kh.S-1) vélogène et viscérotropique. Ils avaient des symptômes cliniques permettant d'observer l'infection du système nerveux et de l'appareil digestif 11 à 13 jours après l'inoculation (p.i.). Le virus fut d'abord détecté dans les organes des moineaux infectés 9 jours p.i. puis il y est resté pendant 8 jours; il fut ensuite évacué par la cavité orale 5 à 10 jours p.i. Les sérums prélevés des oiseaux survivants avaient des niveaux élevés d'anticorps provoquant l'inhibition de l'hémagglutination qui furent discernables jusqu'à 88 jours p.i.

Summary

Sudan house sparrows *(Passer domesticus arborius)* were inoculated with a velogenic viscerotropic NDV (Kh.S-1). They developed clinical symptoms indicative of the involvement of nervous and digestive systems 11-13 days post-inoculation. The virus was first detected in organs of infected sparrows 9 days postinoculation and persisted for up to 8 days. The virus was shed through the oral cavity within 5-10 days post-inoculation. Sera from surviving birds had high levels of HI antibodies which remained detectable upto day 88 post-inoculation.

INTRODUCTION

Information on the duration of NDV infection, virus shedding and immune response for many species of wild birds is needed to determine whether or not there is a carrier state and the potential of each species as possible spreaders of NDV[1,2].

Sudan house sparrow *(Passer domesticus arborius)* was chosen for further studies on the pathogenesis of NDV infection because of its presence in large numbers in the vicinity of poultry farms and because of the pattern of experimantal NDV infection in it[3].

Materials and Methods

The Kh. S-1 isolate of NDV and wild birds used in this study have previously been described[3].

Distribution of the Virus in Tissues of Infected Birds

Eighty eight Sudan house sparrows were inoculated intranasally (I/N) with Kh. S-1 isolate of NDV. Each bird received $10^{7.5}$CELD 50%. Three birds were killed every 48 hours till 40 days postinoculation (p.i). Brains spleens, lungs, and tracheas were separately collected aseptically into bijou bottles. Phosphate buffered saline containing penicillin, streptomycin and gentamycin was added and samples were stored at -20°C until used. Tissue samples were then homogenized, a few drops of PBS were added to obtain approximately 10% suspensions which were then centrifuged and the supernatent fluids used for inoculation of embryonated eggs via the allantoic cavity.

Each embryo received 0.2 ml of inoculum. The embryonated eggs were candled daily and amnioallantoic fluids from dead embryos and those still alive 5 days p.i. were collected and examined for the presence of the virus using haemagglutination (HA) inhibition (HI) tests according to standard techniques[4].

Virus Shedding and Antibody Response

Sixteen Sudan house sparrows were inoculated intranasally with Kh. S-1 isolate of NDV. Each bird received $10^{7.5}$ CELD 50%. Oral and cloacal swabs were collected daily at random from 4 birds for 13 days p.i. and then every three days for 40 days p.i. Swabs were placed in tubes containing 1-2 ml of brain heart infusion broth (BHIB: 33.3 gm BHI powder, 900 ml distilled water, 100 ml bovine serum, 10 million units penicillin, and 10 gm streptomycine sulfate). They were then frozen at -20°C until they were used to inoculate embryonated eggs via the allantoic cavity for reisolation of virus. Swab specimens were thawed at room temperature, centrifuged at 1500 r.p.m for 5 minutes and 1-2 ml of supernatent fluid was filtered through a 0.4 mμ milipore filter fitted to a syringe and each filterate was used to inoculate two 11-day-old embryonated eggs.

Each embryo received 0.2 ml of inoculum. The embryonated eggs were candled daily and AAF from dead embryos and those still alive 5 days p.i. were collected and examined for the virus using HA and HI tests.

Blood samples from three randomly selected birds were collected from the jugular vein 3, 7, 11, 15, 19 days p.i. and at weekly intervals thereafter for 88 days. Sera were separated, inactivated at 56°C for 30 minutes and then frozen at -20°C till examined for the presence of NDV haemegglutination inhibiting antibodies.

Results

Classical ND clinical signs as previously described[3] were observed in 9 birds out of 16.

1. *Virus Distribution in Tissues of Infected Birds*

Reisolation of NDV from organs of experimentally inoculated sparrows are shown in Table 1. Newcastle disease virus was first detected 9 days p.i. in the brains, spleens and lung-trachea (POOLs), and was not detected in any of these organs later than 17 days p.i. The virus persisted longest in the brain (8 days), next in spleen (6 days), and then in lung-trachea (4 days).

Virus Shedding from Infected Birds: Table 1 also shows the results of NDV reisolation from oral and cloacal swabs. The virus was reisolated from oral swabs 5-10 days p.i., except at day 6, and was not reisolated later than day 10 p.i. The virus was not reisolated from any of the cloacal swabs till the end of the experiment. No reisolation was made from any of the oral or cloacal swabs from control birds.

2. *Humoral Antibody Response of Infected Birds*

Fig. 1 shows the mean HI-antibody titre in surviving birds.

Haemogglutination inhibiting antibodies were first detected on day 7 p.i. Mean antibody titre at day 19 p.i. was 6.33 $(\log)_2$ and at day 39 p.i. was 2 $(\log)_2$. Sera from surviving birds showed detectable antibodies till day 88 p.i., when the experiment was terminated.

Control birds did not show detectable antibodies in sera collected at the same time.

Discussion

Sudan house sparrows inoculated with Kh. S-1 isolate of NDV developed clinical symptoms indicative of the involvement of nervous and digestive systems 11-13 days p.i.

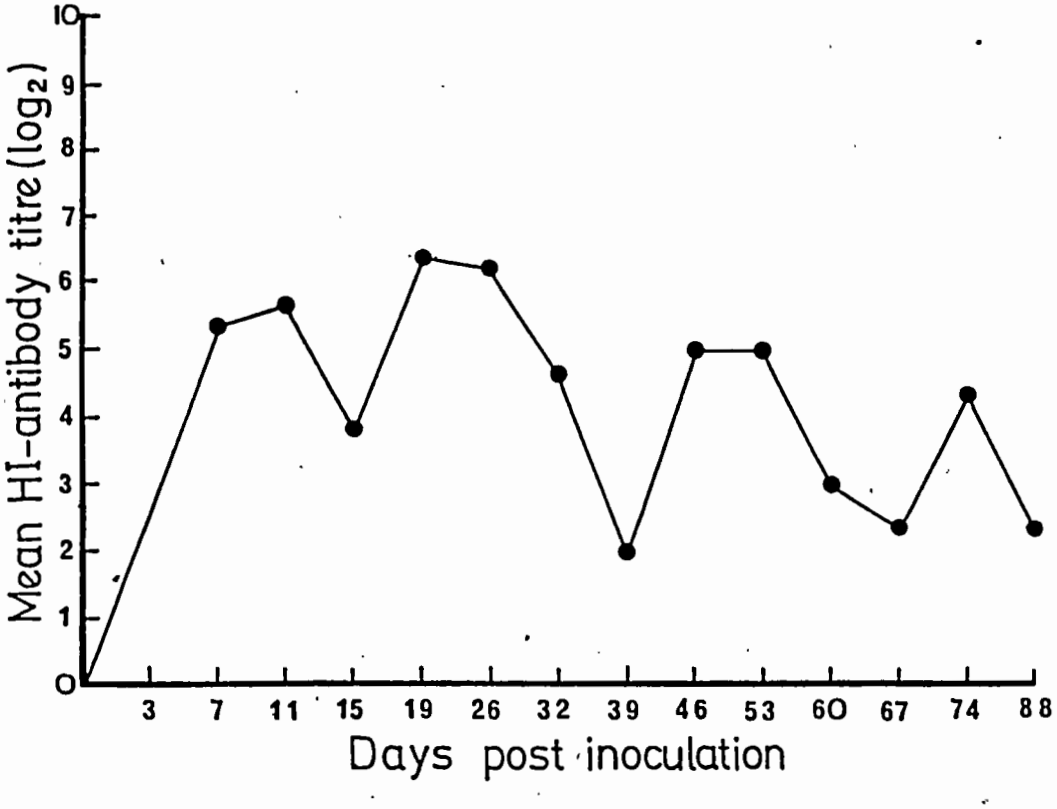
Newcastle disease virus was first detected 9 days p.i. in the brains, spleens, and lung-trachea and was not detected in any of the organs later than day 17 post-inoculation.

Recovery of the virus from more than one organ of infected birds suggested the pantropic nature of the Kh. S-1 isolate of NDV in Sudan house sparrows. It seems

Table 1: Detection of NDV in Organs and Swabs of Sudan House Sparrows Inoculated with Kh.S-1

Day post inoculation	Brain	Spleen	Lung-trachea	Oral swabs	Cloacal swabs
5	–	–	–	+	–
6				–	–
7	–	–	–	+	–
8				+	–
9	+	+	+	+	–
10				+	–
11	+	+	+	–	–
12				–	–
13	+	+	+	–	–
14				–	–
15	+	+	–	–	–
16				–	–
17	+	–	–	–	–
18				–	–
19	–	–	–	–	–
20				–	–
21	–	–	–	–	–

Fig.1. Mean HI-antibody titre (log_2) of house sparrows inoculated with Kh.S-1 isolate of NDV.



that in Sudan house sparrows Newcastle disease virus takes longer time (9 days) to reach its predilection sites and propagate to produce clinical infection.

In chickens, however, following subcutaneous, intramuscular, and intratracheal inoculations and in mallard inoculated intravenously with NDV, the virus was recovered from different organs 1-7 days p.i.[5,6,7,8].

The isolation of the virus 9-17 days p.i. suggests that sparrows may be potential spreaders of the velogenicviscerotropic NDV.

Virus shedding through the oral cavity was found to be between 5-10 days p.i. and the virus was not isolated from cloacal swabs. it is apparent from this result that Sudan house sparrow sheds NDV for a relatively shorter period of time as compared to psittacine and other pet birds which were found to be capable of shedding the virus for longer periods varying between 32 to 402 days[1,9]. This observation supported the concept that passeriformes shed NDV for a short period[9]. Difficulties in the reisolation of NDV from passeriformes had been reported before[10,11] as had the failure to isolate NDV from cloacal swabs of intranasally inoculated African weaver finches[9].

References

1. Erickson, G.A., Mare, C.J., Gustafson, G.A., Miller, L.D., Proctor, S.J. and Carbrey, E.A. (1977). *Avian Dis.* **4,** 642.
2. McDaniel, H.A., and Orsborn, J.S. (1973). *J. Am. Vet. Med. Assoc.* **103,** 1075.
3. Khalafalla, A.I., Nayil, A.A., Nimir, A.H. and Hajer, I. (1988) *Bull. Anim. Hlth. & Prod. (impress).*
4. Allan, W.H., Lancaster, J.E. and Toth, B. (1978). Newcastle disease vaccines. Their production and use. Food and Agriculture Organization, Rome.
5. Miller, B.R. and Miller, R.E. (1950). *J. Am. Vet. Med. Ass.* **117,** 229.
6. Hofstad, M.S. (1951). *Am. J. Vet. Res.* **12,** 334.
7. Asdel, M.K. and Hanson, R.P. (1960). *Am. J. Vet. Res.* **21,** 128.
8. Friend, M. and Trainer, D.O. (1972). *Avian Dis.* **4, 700.**
9. *Vickers, M.L. and Hanson, R.P. (1978). Avian Dis.* **1,** 70.
10. Gustafson, D.P. and Moses, H.E. (1953). *Am. J. Vet. Res.* **14,** 581.
11. Pearson, G.L. and McCann, M.K. (1975). *J. Am. Vet. Med. Ass.* **7,** 610.

Received for publication on 30th May 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 55-58

ROLE OF SOME PASSERIFORMES BIRDS IN TRANSMISSION OF NEWCASTLE DISEASE. III: TRANSMISSIBILITY OF NEWCASTLE DISEASE VIRUS BY SUDAN HOUSE SPARROWS *(PASSER DOMESTICUS ARBORIUS)*

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ROLE DE CERTAINS OISEAUX PASSEREAUX DANS LA TRANSMISSION DE LA MALADIE DE NEWCASTLE III. TRANSMISSIBILITE DU VIRUS DE LA MALADIE DE NEWCASTLE PAR LES MOINEAUX FRANCS DU SOUDAN *(PASSER DOMESTICUS ARBORIUS)*

Résumé

Il a été constaté que les moineaux francs du Soudan *(Passer domesticus arborius)* expérimentalement infectés pouvaient transmettre le virus de la maladie de Newcastle vélogène et viscérotropique aux poulets et vice versa.

Summary

It was possible to transmit velogenic viscerotropic Newcastle virus from experimentally infected Sudan house sparrows *(Passer domesticus arborius)* to chickens and vise versa.

INTRODUCTION

Appearance of Newcastle disease (ND) in relatively isolated poultry farms gave impetus to the opinion that free-flying birds might be associated with dissemination of the disease among domestic fowls.

Many authors[1,2,3,4] presented evidences that wild birds were one of the major factors in the spread of NDV.

After confirmation of experimental susceptibility of Sudan house sparrows to velogenic viscerotropic NDV[5], experiments were designed to determine whether Sudan house sparrow *(Passer domesticus arborius)* might become infected as a result of cohabitation with infected chickens or whether, if infected, they may transmit ND to susceptible chickens.

Materials and Methods

The Kh. S-1 isolate of NDV and wild birds used in this study have previously been described[5].

Transmission of NDV from Infected Chickens to Incontact Sudan House Sparrows

Sixteen Sudan house sparrows and ten 7-week-old chickens were used in this experiment. All chickens were screened for haemagglutination inhibiting (HI) antibodies in their sera and were found to have no antibodies.

Seven chickens (Group A) were inoculated intransally with $10^{7.5}CELD_{50}$ of Kh. S-1 isolate of NDV and were cohabitated in a large cage (4.62 cubic meters) with 16 sparrows. Three more uninoculated chickens (Group B) were also cohabitated to maintain and prolong the infection. Cohabitating chickens and sparrows shared food and water.

Transmission of NDV from Infected Sparrows to Incontact chickens

Sixteen sparrows and ten 7-week-old chickens were used in this experiment. Eight sparrows (Group A) were each inoculated intransasally with $10^{7.5}CELD_{50}$ of Kh. S-1 isolate of NDV and were cohabitated in a large cage with chickens. Four days later, a second group of 8 sparrows (Group B) were inoculated and mixed with the first group to maintain and prolong infection. Cohabitating chickens and sparrows shared food and water.

Chickens and sparrows were observed

daily for signs of infection.

Birds that died, killed or survived the duration of the experiments were necropsied and examined for gross lesions. Brains, lungs, livers and spleens were aspectically collected. Phosphate buffered saline containing antibiotics was added and samples were then frozen at -20°C.

For virus isolation, the tissue samples were homogenized, and a few drops of PBS added to make approximately 10% suspension. Samples were inoculated via the allantoic cavity into 11-day-old embryonating eggs in duplicate. Each embryo received 0.2 ml of inoculum. The embryonating eggs were candled daily, and amnioallantoic fluids (AAF) from dead embryos and those still alive 5 days p.i. were collected and examined for NDV using haemagglutination (HA) and haemagglutination inhibition (HI) tests according to standard techniques[6]

Blood samples were collected from surviving birds at different times. Sera were separated, inactivated at 56°C for 30 minutes and then stored at -20°C till examined using HI test.

Results

Transmission from Infected Chickens to Incontact Sparrows

All inoculated chickens became clinically ill. Obvious clinical symptoms of infection began on days 4-5 p.i. Symptoms included depression, inappetance, closed eyes, stretched neck and diarrhoea. Clinically sick chickens died within 5-10 days p.i.

All the three incontact chickens which were mixed to prolong the infection, developed clinical symptoms 8 days after cohabitation. They showed nervous signs, diarrhoea and death which occurred 10-14 days after cohabitation.

Two sparrows died 41 days post-cohabitation and no necropsy was made because they were decomposed. Two sparrows showed depression and incoordination on the same day and another sparrow on day 42 post-cohabitation.

Necropsy revealed haemorrhages in the proventriculus and intestine of all inoculated and incontact chickens for prolongation of infection. Haemorrhages were also observed in the proventriculus of three sparrows that were killed on day 42 postcohabitation.

Newcastle disease virus was isolated from organs of inoculated and incontact chickens and was not isolated from incontact sparrows (Table 1).

Pooled sera from clinically sick sparrows killed on day 42 postinoculation, yielded HI antibody titre of 3 ($\log_2$). Titres of 2 and 3 ($\log_2$) were also detected in pooled sera (2 birds each) obtained from birds sacrificed on day 51 after cohabitation.

Transmission of NDV from Infected Sparrows to Incontact Chickens

One inoculated house sparrow died 3 days postinoculation. Out of the 16 inoculated sparrows, 9 showed clinical signs suggestive of Newcastle disease 11-14 days p.i. Eleven died 13-18 days p.i. All the ten-incontact chickens showed signs of depression, inappetance incoordina-

Table 1: Transmission of NDV from Infected Chickens to Incontact House Sparrows

Chickens			Sparrows		
No. of birds	Days until death	Virus reisolation	No. of birds	Days until death	Virus reisolation
3[A]	5	ND	2	41	ND
4[A]	6	+	3	42	–
1[B]	10	ND	4	51	–
1[B]	13	+			
1[B]	14	+			

A=Experimentally inoculated birds.
B=Uninoculated-incontact birds.
ND=Not done.

Table 2: Transmission of NDV from Infected House Sparrows to Incontact Chickens

Sparrows			Chickens		
No. of birds	Days until death	Virus reisolation	No. of birds	Days until death	Virus reisolation
1[A]	3	ND	3	10	+
3[A]	13	+	2	13	+
1[A]	17	–	3	15	+
1[B]	7	ND	1	17	–
3[B]	18	+	1	18	–
2[B]	21	–			

A=Experimentally inoculated birds.
B=Birds inoculated 4 days later to prolong the infection.
ND=Not done.

tion, paralysis and diarrhoea. These chickens died 10-16 days after cohabitation. Four birds died with no obvious clinical signs.

Haemorrhages in the proventriculus were observed in 7 out of 11 necropsied sparrows and also in 4 out of 10 incontact chickens. In chickens, haemorrhages were also observed in intestine.

Newcastle disease virus was isolated from 6 sparrows and 8 chickens (Table 2).

Antibodies against NDV were detected in pooled sera of 4 sparrows that survived and were killed on day 35 postinoculation at the termination of the experiment.

Controls did not show clinical signs and at the termination of the experiment no pathological lesions were observed, nor did their sera yield detectable antibodies.

Discussion

Transmission of Kh. S-1 isolate of NDV from experimentally infected Sudan house sparrows to incontact chicken was evidenced by the appearance, in chickens, of classical symptoms of ND with haemorrhages in the gut, and by the isolation of the virus from different organs.

Transmission of, at least inapparent infection to sparrows from experimentally inoculated chicks by direct contact was indicated by the demonstration of HI-antibodies in pooled sera of sparrows.

Results of transmission of NDV between sparrows and chickens agree with that of Gustafson and Moses[7] who found that, chickens cohabitating with aerosol-infected sparrows became infected with NDV as evidenced by the development of clinical signs and isolation of the virus from tracheal washings. In that study, Newcastle disease virus was isolated from the lungs, livers, or brains of 3 out of 23 sparrows that died at 21 and 26 days of cohabitation with infected chickens and antibodies against NDV were demonstrated in sera of surviving sparrows.

These results also agree with those of Hartwigk and Nitsch[8], who stated that two 10-week-old chickens developed ND after being incontact with orally infected sparrows, but they disagree with the results of Maglione[9], who observed that sparrows did not contract the disease by direct contact with infected chickens, or from infected premises nor did they act as carriers of the virus.

These studies showed that five passeriformes species from Sudan wild birds (Sudan house sparrows, Sudan golden sparrow, Durra birds, Rosy fire finches and weaver birds) which frequently visit poultry houses and mix with poultry were susceptible to NDV. When infected, these birds shed the virus for a limited period of time. It was also possible to transmit NDV from experimentally infected Sudan house sparrow to chickens and vise versa. It, therefore, became extremely important to prevent these wild birds from entering poultry houses.

References

1. Brandly, C.A., Moses, H.E., Jones E. Elizabeth and Jungherr, E.L. (1946). *Am. J. Vet. Res.* **7,** 243.
2. Skoda, R. and Zuffa, A. (1956). Vet. Capsopies 5, 441 *(Cit. Vet. Bull.* **9,** 466, 1957).
3. Zarzuelow, E. and Galiano, G.F. (1969). *Rev. Patr. Anim.* 13, 49 *(Cit. Vet. Bull.* **7,** 40, 1970).
4. Alexander, D.J., Wilson, G.W.C., Russel, P.H., Lister, S.A. and Parson's, G. (1985). *Vet. Rec.* **17,** 429.
5. Khalafalla, A.I., Nayil, A.A., Nimir, A.H. and Hajer, I. (1988). *Bull. Anim. Hlth. Prod.* (in press).
6. Allan, W.H., Lancaster, J.E. and Toth, B. (1978). Newcastle disease vaccines. Their production and use. Food and Agriculture Organization, Rome.
7. Gustafson, D.P. and Moses, H.E. (1953). *Am. J. Vet. Res.* **14,** 581.
8. Hartwigk, H. and Nitsch, G. (1957). Tierztle. Wochschr. 70, 285 *(Cit. Vet. Bull.* **10,** 523, 1957).
9. Maglione, E. (1957). *Ann. Fac. Med. Vet.* Torine. 6, 63 *(Cit. Vet. Bull.***10,** 577, 1957).

Received for publication on 30th May 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 59-64

HAEMATOLOGICAL STUDIES IN APPARENTLY NORMAL FIVE INDIGENOUS BREEDS OF GOATS IN NIGERIA

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ETUDES HEMATOLOGIQUES MENEES AVEC CINQ RACES INDIGENES DE CHEVRE APPAREMMENT NORMALES AU NIGERIA

Résumé

Onze paramètres hématologiques ont été étudiés chez 210 chèvres apparemment normales composées de cinq races indigènes du Nigeria. On a comparé les valeurs hématologiques des cinq races.

Dans l'ensemble, la chèvre naine de l'Afrique de l'Ouest (WAD) avait le nombre de globules rouges (GR) le plus petit (7,09 x 16⁶/cc) et la chèvre rousse de Kano (KB) le nombre de GR le plus élevé. Pour les valeurs moyennes de l'hématocrite, la valeur la plus élevée a été observée chez la race Salla (29,31%) et la plus faible chez WAD (23,67%). La concentration d'hémoglobine chez les quatre races était très similaire (9,21-9,99 mg/dl); en revanche chez WAD, elle était de 7,73 mg/dl. Les valeurs moyennes se rapportant à trois érythrocytes (volume corpusculaire moyen: VCM; hémoglobine corpusculaire moyenne: HCM; concentration corpusculaire moyenne de l'hémoglobine: CCMH) chez les 5 races étaient similaires. Les valeurs de CCMH, de l'hématocrite et de GR chez toutes les races étudiées ont tendance à baisser lorsque les animaux avaient cinq ans et plus, mais la baisse n'était pas très significative (P>0,05). Aucun des onze paramètres n'était très affecté par la race. En général, le nombre total de leucocytes et la numération différentielle de leucocytes n'étaient affectés ni par la race ni par l'âge. Borono White (BW), Salla et KB avaient apparemment une numération élevée de leucocytes neutrophiles. La numération moyenne la plus élevée de lymphocytes a été observée chez WAD et la plus faible chez BW.

Summary

Eleven haematological parameters were studied in apparently normal 210 goats comprising five breeds from Nigeria. Haematological values were compared and contrasted for the five breeds.

The mean RBC counts were least (7.09 x 10^6/cc) for the West African Dwarf (WAD) goats and highest (8.79 x 10^6/cc) for the Kano brown. The mean PCV values were highest for Salla (29.31%) and lowest for WAD 23.6%). The haemoglobin concentrations for four breeds were very similar (9.21-9.99mg/dl) but the WAD had a value of 7.73mg/dl. The mean values for three erythrocytic indices (MCV, MCH and MCHC) in the 5 breeds were similar. The MCHC, PVC and RBC values in all breeds studied tend to decrease in goats aged five years and above. The decrease was not found very significant (P>0.05). None of the eleven parameters is significantly affected by breed. Total leucocytes and differential leucocyte counts were generally not affected by breed or age. The Borno white, Salla and Kano brown had apparently high neutrophil counts. The highest mean for lymphocytes was observed in the WAD while the Borono white goats had the lowest count.

INTRODUCTION

Investigations of haematological values in apparently normal domestic animals have received increasing attention in recent year[1]. There are reasons of both scientific interest and economics for attempting to establish normal haematological values for tropical animals. Haematological values are of importance in diagnosing many haemoparasitic infections in food animals[2,3]. The usefulness of normal haematological values in assessing the health status of ruminants has been established[4,5]. In goats anaemia is not a primary disease symptom but some defects in feeding or parasitic infestation[5,6]. It is often difficult to assess the correct health status of an animal without recourse to an examination of its blood[6]. Haematological values are also of great help to the veterinarian in diagnosis, treatment and prognosis in many dis-

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eases in the tropics[7].

There are relatively few reports on normal blood values for goats in Africa. Baseline data on the haematological values of Nigerian goats were investigated by Oduye[5]. More recently Amakiri[8] compared the blood values of some breeds of Nigerian goats with those of exotic breeds. The present study was carried out to determine the normal haematological values in five breeds of Nigerian goats and to compare the effect of breed and age on such values among the breeds.

Materials and Methods

210 goats comprising of five phenotypically different breeds were sampled for this investigation. These are the Red Sokoto (Maradi), the Kano brown, the Desert and Sahel (Salla), the Borono white and the West African Dwarf (WAD). The majority of these goats were those passed for slaughter at a government abattoir at Ibadan. The animals were observed for any gross bodily abnormalities were not selected for the study.

Each animal was bled from the jugular vein before being slaughtered; about 4 cc. of blood was collected in bijou bottles containing either 40 I.U of heparin or ethylene-diamine-tetra-acetic acid (EDTA). Samples were labelled and the sex, breed and age of goat indicated on the bottle.

Estimation of haematological values

Each blood sample was first examined for haemoparasites using to approaches. A thin smear was made and after air-drying and fixing in alcohol, it was stained with Giemsa stain. Slides were examined under the high power of the microscpoe using x100 objective and x10 aye piece for the presence of parasites. Also the Buffy coat examination technique was used[9] to screen for trypanosome infection.

Estimation of total erythrocytes and leucocytes

The total red blood cells (RBC) and white blood cells (WBC) values were determined by the use of Coulter Dual Diluter III and an automatic counting device as previously described by Oduye[5]. The packed cell volume (PCV) and the haemoglobin value were determined as previously described by Cole[10].

Determination of erythrocytic indices

The mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were calculated mathematically from values of PCV, Hb, and RBC as described by Schalm[11].

Statistical analysis

Statistical examination of the various erythrocytic values for the five breeds was carried out using the analysis of variance (ANOVA). Values for different age groups were also analysed by the variance technique[12].

Results

Both Giemsa stained thin blood film microscopy and buffy coat technique were used for parasite detection; no blood parasites were detected by any of the two methods.

Clinical observation on the goats

Generally, most of the bucks and does appeared to be clinically normal as there were no gross abnormalities observed in any one of them.

Erythrocytic values

The haematological values obtained from this study are shown in Table 1, 3, 4, 5 and 6. The mean total RBC counts ranged from 7.09 x 10^6/cc for West African Dwarf goats to 8.79 x 10^6/cc for Kano Brown breeds (Table 1). The mean PCV values recorded were highest for Salla (29.31%) and lowest for the WAD (23.67%). The haemoglobin concentration values for the various breeds were very similar (9.21-9.95 mg/dl) except for the West African goats which had a mean value of 7.73 mg/dl. The mean values for the different erythrocytic indices (MCV, MCH and MCHC) in the five breeds of goats were similar. (Tables 1 & 2)

Table 1: Mean erythrocytic values for five breeds of Nigerian goats

Breeds of goats	Number of blood samples examined	PCV (%)	Hb (mg/dl)	RBC ($\times 10^6$)	MCV (fl)	MCH (pg)	MCHC (g/dl)
Breeds of goats	**Number of blood samples examined**	**PCV (%)**	**Hb (mg/dl)**	**RBC ($\times 10^6$)**	**MCV (fl)**	**MCH (pg)**	**MCHC (g/dl)**
Sokoto Red	124	28.14[a] ±5.48	9.95[b] ±1.56	8.17[d] ±1.73	35.77[e] ±7.18	11.75[f] ±1.98	32.55[c] ±5.16
Kano Brown	45	28.86[a] ±4.25	9.68[b] ±1.56	8.79[d] ±1.73	33.26[e] ±7.18	10.77[f] ±1.98	33.39[c] ±3.99
Salla	28	29.31[a] ±4.8	9.21[b] ±1.41	8.01[d] ±1.72	38.38[e] ±9.18	11.63[f]	31.31[c] ±4.92
Borono White	7	28.33[a] ±4.56	9.23[b] ±1.68	7.72[d] ±1.8	36.37[e] ±6.37	12.17[f] ±2.69	32.33[c] ±3.37
West African Dwarf (WAD)	6	23.67[a] ±5.8	7.73[b] ±1.88	7.09[d] ±2.06	34.33[e] ±5.88	11.0[f] ±2.3	32.67[c] ±3.25

± =Standard deviation.
Figures with the same superscript indicate no statistically significant difference; i.e. P>0.05.

Table 2: Mean Leucocytic values for five breeds of goats

Breeds of goats	Number of blood samples examined	WBC ($\times 10^3$)	Differential leucocyte counts in percentages			
			Neutrophils	Lymphocytes	Eosinophils	Monocytes
Sokoto Red	124	9.219 ±4.56	48.01[h] ±14.9	50.01[i] ±15.50	1.1[j] ±0.32	1.3[k] ±0.62
Kano Brown	45	10.69 ±4.56	53.83[h] ±13.37	44.83[i] ±13.02	2.0[j] ±0.43	1.19[k] ±0.41
Salla	28	8.419 ±3.06	56.69[h] ±19.56	47.2[i] ±20.17	1.67[j] ±1.6	1.25[k] ±0.45
Borono White	7	6.999 ±2.4	56.33[h] ±10.75	41.5[i] ±10.03	3.5[j] ±2.0	1.0[k]
West African Dwarf (WAD)	6	13.489 ±6.37	46.17[h] ±7.92	53.00[i] ±7.2	N.D[j]	1.25[k] ±0.47

N.D=Not done
±=Standard deviation
Figures with the same supercript indicate no statistically significant difference; P>0.05.

Effect of breed on haematological values

The mean values of each set of haematological values for the 5 breeds of goats examined were compared by analysis of variance technique[12]. There was no significant difference between the means of any of these haematological values. None of the eleven haematological values is therefore significantly affected by breed.

Effect of age on erythrocytic values

Mean values of each haematological parameter in 3 age groups i.e. 0-3 years, 4

Table 3: Mean haematological values in various age ranges for Sokoto Red Goats

Age range (years)	Number of goats	PCV (%)	Hb (mg/dl)	WBC ($\times 10^3$)	RBC ($\times 10^6$)	MCV (fl)	MCH (pg)	MCHC (g/dl)
0–3	63	28.52 ±2.38	9.26 ±0.89	7.92 ±7.38	8.54 ±1.09	34.13 ±5.56	10.72 ±1.71	31.64 ±3.1
4	45	29.4 ±4.25	7.79 ±1.96	9.78 ±3.96	7.96 ±2.00	37.24 ±7.21	13.68 ±3.38	34.21 ±5.41
5 and above	16	25.91 ±5.6	8.78 ±2.1	7.31 ±6.74	7.96 ±2.07	27.13 ±10.46	9.38 ±4.61	34.38 ±4.43
		P>0.05	P>0.05	P>0.05	P>0.05	P>0.05	P>0.05	P>0.05

±Standard deviation

Table 4: Mean haematological values in various age ranges for Kano Brown Goats

Age range (years)	Number of goats	PCV (%)	Hb (mg/dl)	WBC ($\times 10^3$)	RBC ($\times 10^6$)	MCV (fl)	MCH (pg)	MCHC (g/dl)
0–3	25	30.11 ±5.2	9.62 ±2.65	10.08 ±3.3	8.77 ±3.8	35.18 ±14.04	10.83 ±4.04	31.619 ±4.04
4	14	27.0 ±4.52	9.7 ±1.49	11.38 ±4.31	8.88 ±1.78	31.2 ±7.23	10.44 ±2.04	34.5[f] ±4.1
5 and above	6	26.5 ±3.34	9.95 ±1.6	11.26 ±3.04	8.03 ±1.36	33.5 ±4.63	13.5 ±1.69	37.25[f] ±3.74

Figures with different superscript indicate significant diffarence (P<0.05)

± =Standard deviation

Table 5: Mean haematological values in various age ranges for Salla goats

Age range (years)	Number of goats	PCV (%)	Hb (mg/dl)	WBC ($\times 10^3$	RBC ($\times 10^6$	MCV (fl)	MCH (pg)	MCHC (g/dl)
0–3	13	30.0 ±3.54	9.27 ±1.4	10.31 ±3.82	8.00 ±1.77	35.33 ±9.56	10.33 ±2.76	30.67 ±5.37
4	9	31.25 ±5.63	9.2 ±1.28	8.79 ±2.46	8.00 ±1.81	40.00 ±10.28	11.5 ±1.64	29.0 ±4.32
5 and above	6	29.33 ±5.63	8.8 ±1.28	8.14 ±2.46	7.52 ±1.81	40.38 ±10.28	11.38 ±1.64	30.0 ±4.32
		P>0.05	P>0.05	P>0.05	P>0.05	P>0.05	P>0.05	P>0.05

± =Standard Deviation

years and 5 years and above were compared (Table 3-6). Mean values of each of the seven haematological parameters for the three age groups, in all the five breeds showed no significant difference (P>0.05). This seemed to indicate that the mean values of haematological parameters are apparently not affected by age. Within each breed, however, age was found to affect haematological values. The means of MCV, MCH values in Sokoto Red and MCHC values in Kano Brown breeds of goats for the same age differed significantly (P>0.05). There

Table 6: Mean haematological values in various age ranges for Borono White Goats and West African Dwarf Goats

Age range (years)	Number of goats	PCV (%)	Hb (mg/dl)	WBC ($\times 10^3$)	RBC ($\times 10^6$)	MCV (fl)	MCH (pg)	MCHC (g/dl)
Borono White		27.75	8.9	7.05	8.25	38.25	12.25	32.0
0–3	4	±5.66	±0.28	±1.99	±1.37	±0.71	±2.83	±5.66
4	3	28.67	10.13	7.42	8.8	32.67	11.33	35.0
		±5.57	±0.65	±1.97	±1.61	±1.17	±0.82	±11.95
WAD		21.0	6.73	12.59	6.02	37.0	12.0	N.D
0–3	3	±2.83	±1.28	±2.74	±0.78	±1.41	±2.12	
5	3	28.0	8.7	8.69	8.71	31.5	9.0	N.D
		±6.50	±2.70	±6.20	±2.10	±2.52	±4.36	

ND=Not done
±=Standard deviation

was, however, no significant difference (P>0.10) between the means of MCH values for Sokoto Red goats aged between 0-3 and 4 years. The general trend, however, suggested that the mean PCV and RBC values in the various age ranges in all breeds, decreased with advancing age (Tables 3-6).

Leucocytic values

Leucocytic values for the various breeds of goats are shown in Table 2. The highest total WBC counts (13.48 x 10^3) was recorded for the WAD. The Borono White, Salla and Kano Brown breeds were found to have high neutrophil counts whilst the WAD had the lowest count. These values were, however, not statistically significant. it was therefore concluded that the total and differential leucocyte count were generally not affected by breed or age Tables 3-6. No significant differences between the values of neutrophils, Lymphocyte, eosinophil and monocyte counts were noticed.

Discussion

The present investigation examined both the effects of breed and age on haematological values of apparently normal five breeds of goats in Nigeria. All the breeds are indigenous to the region. The erythrocytic values obtained for these goats were similar to those found for indigenous tropical goats[5,8]. The difference between the means of total RBC values in the five breeds of goats was not significant (P>0.05). Similarly age did not produce any significant difference (P>0.05) between the means of the RBC values. The results of this study have thus shown that clinically normal Nigerian goats have a range of haematological values that is independent of breed but different from those of the temperate breeds[13]. Previous study by Oduye[5] considered the effect of age and sex but did not find any significant difference; studies by Amakiri[8] on the other hand evaluated haematological values in the Red Sokoto, the West African Dwarf, the Saanen and the F_1 generation of crosses between the Saanen and the local breeds. The study did not reveal any significant difference in the haematological values. The present study has expanded the scope or two previous studies by examining five phenotypically different local breeds. In all the five breeds examined no significant difference was observed in the erythrocytic values. The mean values of RBC, PCV and MCV in the five breeds of goats are not markedly different from values of females goats previously reported by Holman and Dew[1]. The haemoglobin values in the Nigerian goats were considerably lower than those reported by Millson *et al.*[14]. Further work on the

changes in the erythrocytic values from birth to three years in the Nigerian goat breeds might be very illuminating. Previous study in temperate goats by Holman and Dew[1] showed a mean total white blood (WBC) of 8.08 x 10^3 whilst Millson *et al.*[14] reported a range of 9.00 x 10^3 to 15 x 10^3. The present investigation showed that Nigerian goats have a mean higher WBC counts than the breed in temperate climate. The findings of the present investigation therefore corroborate those of Edwards *et al*[15] who recorded a mean WBC count of 14.54 x 10^3 for West African Dwarf goats in Ghana. The highest mean WBC counts of 10^3 x 13.48 were observed in the West African Dwarf. The values recorded for the four other breeds were similar to those found by Amakiri[8] for three breeds of goats in Nigeria. It was found that factors such as breed and age had no noticeable effect on WBC counts. No significant difference ($P>0.05$) was noted between the means of WBC values recorded according to age or breed. In the overall consideration of all the five breeds studied the West African Dwarf goats have the lowest PCV and Hb values. The Salla and Borono white breeds had the highest neutrophil counts while the Red Sokoto and West African Dwarf had the lowest counts. Statistical evaluation of those values however did not reveal any significant difference between breeds for neutrophil, lymphocyte, eosinophil and monocytes. The relatively high WBC counts in the five breeds of goats deserves further investigation. It is possible that husbandry practice and environmental disposition might enhance subclinical parasitic infestation[16,17,18]; however none of the goats evaluated in this study was found to have any haemoparasitic infection.

References

1. Holman, H.H. and Dew, S.M. (1963). *Res. Vet. Sc.* **4,** 121.
2. Bennet, G.F. (1962). *Can. J. Zoo* **40,** 124.
3. Anosa, V.O. and Isoun, T.T. (1980). *J. Comp. Path.* **90,** 155.
4. Oduye, O.O. and Okunaiya (1971). *Bull. Epiz. Dis. Afr.* **19,** 213.
5. Oduye, O.O. (1976). *Trop Anim. Hlth Prod.* **8,** 131.
6. Ilemobade, A.A. (1982). Proceedings of the Third International conference on goat production and diseases. Arizona. *Dairy Goat Journal* **68-71.**
7. Clarkson, M.J. (1968). *J. Comp. Path.* **78,** 189.
8. Amakiri S.F. (1981). *Bull. Anim. Hlth Prod. Afr.* **29,** 377.
9. Woo, P.T.K. and Kauffmann, M. (1972). *ACTA. Trop.* **28,** 304.
10. Cole, E.H. (1980). *Veterinary Clinical Pathology* Publisher: W.B. Saunder Ltd Philadelphia.
11. Schalm, O.W. (1965). *Veterinary Haematology* Publisher: Lea and Fiebiger Philadelphia Pa.
12. Snedecor; C.W. and Cochran, W.G. (1980). *Statistical Methods.* The Iowa State University Press, Ames. U.S.A.
13. Lewis, J.H. (1976). *Am. J. Vet. Res.* **37,** 601.
14. Millson, G.C; West. L.D. and Dew, S.M. (1960). *J. Comp. Path.* **70,** 194.
15. Edwards, E.E., Judd; J.M. and Squire F.A. (1955) *Nature* **176,** 549.
16. Laing, J. (1980). Goat Husbandry. Publisher: Faber and Faber, London. Pg. 226.
17. Ugochuku, E.I. (1983). *Bull. Anim. Hlth Prod. Afr.* **31,** 391.
18. Kariuku, D.P. and Jacobsen, P. (1980). *Bull. Anim. Hlth Prod. Afr.* **28,** 7.

Received for publication on 24th May 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 65-67

ANTIBIOTIC RESIDUES IN MILK FOLLOWING TREATMENT OF BOVINE MASTITIS

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RESIDUS D'ANTIBIOTIQUES DANS LE LAIT APRES LE TRAITEMENT DE LA MASTITE BOVINE

Résumé

Des échantillons de lait de vaches ayant subi un traitement de la mastite ont été examinés pour déterminer leur qualité et les résidus d'antibiotiques qu'ils contiennent, à des intervalles de 24 h, 72 h et 120 h après l'interruption du traitement. Le lait des vaches traitées avec une association d'antibiotiques, y compris l'ampicilline, la cloxacilline ou la pénicilline ne contenait pas de résidus 72 h après l'arrêt du traitement. En revanche, le lait des vaches traitées avec une combinaison d'antibiotiques contenant de l'oxytetracycline ou de la tetracycline avait des résidus pendant longtemps, même 72 heures après l'interruption du traitement.

Summary

Milk samples from treated mastitic cows were qualitatively tested for antibiotic residues at intervals of 24hr., 72hr. and 120hr. after withholding treatment. Cows that received ampicillin, cloxacillin or penicillin combination were found to be clear from residues in milk 72hr. Whereas those received combinations of oxytetracycline or tetracycline showed long persistancy of residues in milk till 72hr., after withholding treatment.

INTRODUCTION

Antibiotics are important medicinal components in veterinary practice.

The wide spread use and misuse of antibiotics in dairy industry has created potential residues problem in dairy products specially milk, which is heavily consumed by general public[1].

The presence of antibiotics in milk even in small quantities is not advisable for human consumption and has retarding effects in dairy industry[2].

The purpose of the present study was to determine the presence of antibiotics in milk after its systemic and/or local administration in lactating cows affected with mastitis.

Materials and Methods

Animals

All cows used in this study were from a commercial dairy farm. Forty lactating cows, affected with mastitis were divided into seven groups. Receiving different combination of antibiotics, administered through intramuscular and/or intrammary routes (Attached table).

Samples

Composite milk samples were collected per quarter from the affected cows, 24, 72, and 120hr., after withholding treatment. Tha samples were directly used for the determination of antibiotic residues.

Determination of antibiotics

This is determined qualitatively by microbiological method utilizing *Staph. aureus* disc assay (manual of veterinary/ investigation, 1984). The *Staph. Aureus* used was oxford strain NCTC No. 8178.

Inhibition, of *Staph. Aureus* growth round the disc indicates presence of bacterial inhibitors(s) in the milk samples. The inhibition zone a round the disc defines the presence of antibiotics in the milk sample whether positive or negative. The wider the inhibition zone the more concentration of antibiotic residues in the milk sample tested.

Results

Various antibiotics were used to treat cows during lactation, details of results of this study were given in the table attached. Antibiotic residues in milk following treatment of mastitis during lactation decreased, as the time post treatment increased. The frequency of antibiotic residues in milk after treatment through intramammary, intramuscular or multiple route were similar for groups A, C, D, and G. They were all found to be clear from antibiotic residues 120hr., after withholding treatment. Groups B.E. and F. were all negative for antibiotic residues in milk 72hr., post withholding treatment.

Discussion

About 57% of the milk samples were positive for residues 72hr., after treatment, this exceeded withdrawal times noted for several of the antibiotics preparations used. Marcinak[3] reported 72hr. as time limit for withholding milk from treated cows for human consumption. However, we found that 120hr. is optimum and safest period for withholding milk from treated cows. Antibiotic combinations which contain ampicillin, Penicillin and cloxacillin were found to be the least persistant. They were eliminated from milk 72hr. post withholding treatment. In contrast antibiotic combinations that contain tetracycline or oxytetracyline persisted longer no matter the route of administration was. However, this results were in agreement with that reported by Brauning[4]. Moreover, the persistance of antibiotics in mastatic cows found to be more longer than in normal ones[5]. Also the milk production at time of treatment, frequency of milking, antibiotic used and route of administration were found to affect the persistancy of antibiotic residues in milk[6].

From the above stated results it is felt that if this information is made available to milk producer, it will contribute to improvement of milk quality and protection of consumer health.

Table 1: Residues in Milk after Antibiotic Treatment in Lactating Cows

Group	No. of Animal	No. of Quarters	Antibiotics Administered		Antibiotic residues after withholding treatment at intervals		
			IM*	IMaM**	24h	72h	120h
A	5	10	Procainepenicillin	Tetracyclin/neomycin/ Bacitracin	+	+	–
B	6	10	Chloramphinicol	Ampicillin/ Cloxacillin	+	–	–
C	5	10	Oxytetracycline	Chloramphinicol/ neomycin/penicillin	+	+	–
D	4	10	Oxytetracycline	Ampicillin/ cloxacillin	+	+	–
E	7	10		Ampicillin/ cloxacillin	+	–	–
F	6	10		Chloramphinicol/ neomycin/penicillin	+	–	–
G	7	10		Tetracycline/ neomycin/Bacitracin	+	+	–

*IM: Intramuscular injection
**IMaM: Intramammary infusion
+:Positive
–:Negative detection of antibiotic residues milk samples.

References

1. Allison, J.R.D. (1985). *British Veterinary Journal* pp. 141.
2. Marth, E.H. & Ellickson, B.E. (1959). *Journal of milk & Food Technology* pp. 22.
3. Marciniak, T. (1982). Medycyna Weterynaryjna, 37 pp. 141.
4. Brauning, I. (1980). Pharmacokinetics of inhibitors in milk after intramammary administration of Commercial mastitis preparation containing antibiotics and sulfonamides, Inaugural Dissertation. Ludwing Maximilians Universitat, Munchen, pp. 116.
5. Nouw, J.F.M. & Ziv., G. (1977). *Journal of Food protection*, 41 pp. 8.
6. Mercer, H.D., Geleta, J.N., Schultz, E.J. & Wright, W.W. (1970). *American Journal of Veterinary Research*, 31 pp. 1549.

Received for publication on 4th July 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 69-76

PERFORMANCE OF DIFFERENT GENOTYPES OF BROILER CHICKS FED VARYING PROTEIN LEVELS IN THEIR STARTER AND FINISHER DIETS

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PERFORMANCE DE DIVERS GENOTYPES DE POULETS DE GRIL NOURRIS DE DIFFERENTS NIVEAUX DE PROTEINE CONTENUS DANS LEURS REGIMES DE DEMARRAGE ET DE FINITION

Résumé

A l'âge de 0 à 6 semaines, 135 poussins asexués de un jour dont 45 de chacun des génotypes ci-après: Anak, Hubbard et Starbro ont été regroupés à trois par cage et nourris de trois régimes isothermes (3 Mcal EM/kg) contenant 23; 25 ou 27% de protéine brute (PB) selon un modèle factoriel 3 x 3. Pendant la période de finition (7-11 semaines), chaque poulet est mis dans une cage puis nourri de trois régimes isothermes (3,2 Mcal EM/kg) contenant 17; 19 ou 21% PB selon un modèle factoriel 3 x 3 x 3 (3 niveaux PB de régimes de démarrage, 3 niveaux PB de régime de finition et 3 génotypes). Pour les poulets Anak, au cours de la phase pour le régime de démarrage, on a obtenu les taux de croissance les plus élevés et les meilleurs indices de consommation (IC) avec le régime contenant 27% PB. Pour les variétés Hubbard et Starbro, il n'y a pas eu de différence significative, quant aux gains pondéraux et aux indices de consommation, entre les poulets nourris de régimes contenant 25% PB et 27% PB respectivement. A l'exception de la consommation alimentaire, on n'a pas observé aussi d'effets notables du régime de démarrage sur la performance des poulets pendant la période de finition. La protéine brute dans le régime de démarrage n'a pas beaucoup affecté la consommation alimentaire, les gains pondéraux et les indices de consommation, et il n'y avait pas d'interactions significatives régime de finition x génotype. En revanche, il y avait des effets importants dus au génotype ($P<0,01$) dans tous les paramètres mesurés; en effet, la performance des poulets Anak et Hubbard étaient en général meilleure que celle des poulets Starbro.

Summary

From 0-6 weeks of age, 135 unsexed day-old chicks, 45 of each of Anak, Hubbard and Starbro genotypes were housed three per cage and given three isocaloric (3.0 Mcal ME/kg) diets containing 23, 25 or 27% crude protein (CP) in a 3 x 3 factorial design. During the finishing phase (7-11 weeks), the chicks were housed one per cage and fed three isocaloric (3.2 Mcal ME/kg) diets containing 17, 19 or 21% CP in a 3 x 3 x 3 factorial design (3 levels of previous dietary CP, 3 levels of finishing dietary CP and 3 genotypes). For Anak chicks, during the starting phase, the fastest growth rates and best feed conversion ratios (FCR) were obtained with the 27% CP diet. For Hubbard and Starbro chicks, weight gains and FCR of chicks given the 25% CP diet did not differ significantly from those of chicks given the 27% CP diet. There were no significant carryover effects of starter diet on chick performance during the finishing phase, except in feed intake. Crude protein in finisher diet did not significantly affect feed intake, weight gains and FCR and there were no significant finisher diet x genotype interactions. But there were significant ($P<0.01$) genotype effects in all parameterrs measured, with Anak and Hubbard chicks performing consistently better than Starbro chicks.

INTRODUCTION

Crude protein (CP) requirements for starting and finishing broiler chicks have been well established for temperate[1,2] and tropical[3,4,5,6] environments and there are indications that broiler chicks reared in tropical environments have higher CP requirements than similar birds reared in temperate environments[3,4,5,6]. There is also some evidence, not only for differences among broiler genotypes in their response to dietary CP levels, but also for genotype x diet interaction[7,8]. These differences might be related to inherent differences in their requirement for individual amino acids[2,9,10,11] or to differences among broiler strains in their efficiency of metabolism of essential amino acids[12].

There is paucity of information on the comparative efficiencies of growth and feed utilization under identical feeding and management regime, of the many commercial broiler strains available in

Nigeria. It was the objective of the present study, therefore, to compare the response of three commercial strains of broiler chicks to different levels of CP in their starter and finisher diets and to examine the possible existence of two factor and three factor interactions in the response of the different genotypes to different CP levels in their starter and finisher diets.

Materials and Methods

Experiment 1.

In experiment 1 (the starter phase), 135 unsexed day-old chicks, 45 of each of three commercial hybrid broiler genotypes, namely, Anak, Hubbard and Starbro, were used in a 3 x 3 factorial design with three isocaloric (3.0 Mcal ME/kg) diets containing 23, 25 or 27% CP. Each genotype was randomly divided into three groups of fifteen chicks each and housed three per cage in 75 x 62.5 x 75 cm cages. Thus there were five replications per treatment. All the chicks were given the 25% CP diet during the first week in order to stabilize them. At the end of the first week, the groups were randomly assigned to the three diets (Table I) and fed for another five weeks. Heat was provided by means of 100 watt electric bulbs suspended about 0.45 metres above the cages. Feed and water were provided *ad libitum*. Feed consumption was recorded daily and the birds were weighed weekly. Feed conversion ratio (FCR) was calculated as the number of grams of feed consumed per gram of weight gained. Protein efficiency ratio (PER) was calculated as the number of grams of weight gained per gram of protein consumed.

The data obtained were subjected to a 3 x 3 factorial analysis of variance[13].

Experiment 2.

At the end of experiment 1, each genotype group previously assigned to each starter diet was further divided into three subgroups of five birds per subgroup. The birds were housed one per cage, according to genotype and previous dietary treatment, in 30 x 30 cm cages. Three isocaloric (3.2 Mcal ME/kg) diets containing 17, 19 or 21% CP (Table II) were given to these chicks in a 3 x 3 x 3 factorial arrangement (3 levels of previous dietary CP, 3 levels of finishing dietary CP and 3 genotypes) for another five weeks. Thus there were 27 treatments, each replicated five times. Feed and water were provided *ad libitum*. Feed consumption was recorded daily and the birds were weighed weekly. FCR and PER were calculated as in experiment 1.

The data obtained were analyzed using a 3 x 3 x 3 factorial analysis of variance[13].

Table I: Composition (%) of starter diets used in experiment 1.

Ingredients	Crude protein in diet (%)		
	23	25	27
Yellow maize	58.09	53.70	49.32
Groundnut cake	24.34	28.0	31.65
Fish meal	8.12	9.35	10.55
Wheat bran	6.45	5.95	5.48
Bone ash	1.75	1.75	1.75
Common salt	0.25	0.25	0.25
Vitamin-mineral premix[1]	0.50	0.50	0.50
Methionine	0.50	0.50	0.50
Total	100.0	100.0	100.0
Analyzed composition (%):			
Moisture	9.60	8.90	8.50
Crude protein	23.00	24.8	26.7
Ether extract	3.90	4.00	4.00
Crude fibre	6.81	6.60	6.40
Ash	7.62	7.70	7.35
Nitrogen-free extract	49.07	48.00	47.05
Calculated metabolizable energy (Mcal/kg)	3.00	3.00	3.00

[1]Supplied the following nutrients per kg of diet: vitamin A, 10,000 I.U.; vitamin D3, 2,000 I.C.U.; vitamin E, 5 I.U.; vitamin K, 2.24 mg; riboflavin, 5.5 mg; vitamin B12, 10 μg, calcium pantothenate, 10 mg; niacin, 25 mg; choline chloride, 350 mg; folic acid 1.0 mg; manganese, 56 mg; zinc, 50mg; copper, 10 mg; iron, 20 mg; cobalt, 1.25 mg; amprolium 125 mg; terramycin, 100 mg.

Results

Experiment 1

Average live weights of the experimental chicks at day-old were: Anak, 42.6 g; Hubbard, 40.8 g; and Starbro, 41.3 g. At the beginning of the experiment, that is,

Table II: Composition (%) of finisher diets used in experiment 2.

Ingredients	Crude protein in diet (%) 17	19	21
Yellow maize	76.69	72.34	68.00
Groundnut cake	10.76	13.46	16.15
Fish meal	7.18	8.97	10.75
Wheat bran	2.37	2.23	2.09
Bone ash	1.75	1.75	1.75
Common salt	0.25	0.25	0.25
Vitamin-mineral premix[1]	0.50	0.50	0.50
Methionine	0.50	0.50	0.50
Total	100.0	100.0	100.0
Analyzed composition (%):			
Moisture	8.90	9.10	9.40
Crude protein	17.04	18.90	20.90
Ether extract	3.10	3.20	3.80
Crude fibre	8.50	7.73	7.00
Ash	7.20	7.06	7.15
Nitrogen-free extract	55.26	54.07	51.75
Calculated metabolizable energy (Mcal/kg)	3.20	3.20	3.20

[1]Same as in Table I.

after one week of stabilization on the 25% CP diet, the average liveweights (± standard errors of the means) of the genotypes were: Anak 86.2 ± 13.5 g; Hubbard, 84.8 ± 8.9 g and Starbro, 83.6 ± 7.0 g. There were no significant ($P>0.05$) differences in the liveweights of the genotypes either at day-old or at one week of age. But at the end of the starter phase, Anak chicks weighed significantly ($P<0.05$) more than Hubbard chicks which, in turn, weighed more than Strabro chicks. Average weights of Anak, Hubbard and Starbro chicks at six weeks of age were 833.7, 680.8 and 515.9 g, respectively.

The response of the different genotypes to varying protein levels in their starter diets is shown in Table III. There were significant ($P<0.01$) genotype x diet interactions in daily feed consumption. Anak chicks fed the 25% CP diet had the highest feed intake which was significantly ($P<0.05$) different from the feed intake of Anak chicks fed the 23 or 27% CP whose feed intake values were identical. But for Hubbard chicks, feed intake progressively increased as the level of CP in the diet increased whereas for Starbro chicks feed intake of chicks given the 25 and 27% CP diets was similar but significantly ($P<0.05$) higher than that of chicks given the 23% CP diet.

There were also significant ($P<0.01$) genotype x diet interactions in daily weight gains. Daily weight gains of Anak chicks progressively ($P<0.05$) increased with increases in dietary CP level. For Hubbard and Starbro chicks, daily weight gains of chicks given the 23% CP diet were inferior ($P<0.05$) to those of chicks given the 25 and 27% CP diets. The 25 and 27% CP diets produced identical weight gains in Hubbard chicks as well as in Starbro chicks.

There were also significant ($P<0.01$) genotype x diet interactions in FCR. Among Anak chicks, the best efficiency of feed utilization was obtained in the chicks fed the 27% CP diet. Among Hubbard chicks, however, the best FCR was obtained in the chicks fed the 25% CP diet. Among Starbro chicks, the 25 and 27% CP diets produced better FCR than the 23% CP diet.

There were significant ($P<0.01$) genotype x diet interactions in PER. Among Anak and Starbro chicks, there were no significant differences in PER among chicks fed the various CP levels but among Hubbard chicks, the 23 and 25% CP diets produced significantly ($P<0.05$) better PER than the 27% CP diet.

Experiment 2

Table IV gives the F values obtained from full model analysis of variance of chick performance data in experiment 2. The interactive effects of starter diet, finisher diet and genotype on performance of the chicks during the finishing phase are shown in Table V. There were significant ($P<0.01$) starter diet x finisher diet x genotype interactions in daily feed intake but there were no starter diet x finisher diet x genotype interactions in daily weight gains, FCR and PER.

Table VI gives the effects of starter diets and finisher diets on performance of the chicks in experiment 2. There was a significant ($P<0.01$) starter diet x finisher

Table III: Response of different genotypes of broiler chicks to varying protein levels in their starter diets (experiment 1)

Response variable	Genotype	Crude protein in diet (%) 23	25	27	Mean	S.E.
Daily feed intake (g)	Anak	60.4	69.9	61.1	63.8	
	Hubbard	44.0	47.3	52.7	48.0	
	Starbro	45.2	51.2	52.4	49.6	
	Mean	49.9	56.1	55.4		4.16[a]
Daily weight gain (g)	Anak	20.1	24.3	27.1	23.8	
	Hubbard	15.8	21.0	21.6	19.5	
	Starbro	12.4	15.6	16.2	14.7	
	Mean	16.1	20.3	21.6		1.76[a]
Feed conversion ratio (g feed/g gain)	Anak	3.17	2.94	2.27	2.79	
	Hubbard	3.58	3.03	3.39	3.33	
	Starbro	2.82	2.49	2.51	2.61	
	Mean	3.19	2.82	2.72		0.33[a]
Protein efficiency ratio (g weight gained/ g protein consumed	Anak	1.57	1.56	1.66	1.60	
	Hubbard	1.72	1.79	1.16	1.56	
	Starbro	1.66	1.77	1.48	1.64	
	Mean	1.65	1.71	1.43		0.17[a]

[a]Significant ($P<0.01$) genotype x diet interaction and Significant ($P<0.01$) genotype and diet effects.
[b]Significant ($P<0.01$) genotype x diet interaction but no significant ($P<0.05$) genotype or diet effect.

Table IV: F values from full model analysis of variance of chick performance data in experiment 2

Source of variation	Degrees of freedom	F values for Daily feed intake	Daily weight gain	Feed conversion ratio	Protein efficiency ratio
Replications	4				
Starter diet (S)	2	0.60	0.17	2.28	1.89
Finisher diet (F)	2	2.18	2.59	1.04	30.0**
Genotype (G)	2	56.0**	60.4**	33.7**	25.6**
S x F	4	5.59**	2.39	1.76	1.67
S x G	4	0.69	0.28	0.72	0.86
F x G	4	0.05	0.42	0.96	1.44
S x F x G	8	6.74**	1.42	0.52	0.78
Residual	104				
Total	134				

**($P<0.01$)

diet interaction in feed intake. Among birds started on 27% CP, finisher diet had no effect on their feed consumption. But chicks started on 25% CP and finished on 17 or 19% CP consumed significantly ($P<0.01$) more feed than chicks started on the same 25% CP but finished on 21% CP. Chicks started on 23% CP consumed significantly ($P<0.01$) more feed when they were finished on 21% CP than when they were finished on 17 or 19% CP.

There were no significant starter diet x finisher diet interactions in daily weight gains, FCR and PER, Neither starter diet

Table V: Starter diet x finisher diet x genotype effects on performance of broiler chicks during the finishing phase

Starter diet (% CP)	Finisher diet (% CP)	Genotype	Treatment number	Treatment means for: Daily feed intake (g)[a]	Daily weight gain (g)[b]	Feed conversion ration (g feed/g gain)[b]	Protein efficiency ratio (g weight gained/g protein consumed)[b]
	17	Anak	1	103.1	36.0	2.98	2.0
		Hubbard	2	94.3	38.5	2.48	2.52
		Starbro	3	92.3	26.5	3.67	1.71
23	19	Anak	4	96.8	34.1	2.80	1.96
		Hubbard	5	95.2	41.1	2.56	2.24
		Starbro	6	72.5	20.6	3.61	1.61
	21	Anak	7	107.8	36.6	3.13	1.60
		Hubbard	8	120.3	44.7	2.91	1.72
		Starbro	9	81.8	24.7	3.54	1.43
	17	Anak	10	118.3	36.5	3.43	1.80
		Hubbard	11	110.4	44.1	2.58	2.34
		Starbro	12	84.4	26.0	3.49	1.82
25	19	Anak	13	106.2	36.0	3.01	1.79
		Hubbard	14	116.5	42.3	2.81	1.88
		Starbro	15	76.1	25.9	3.15	1.84
	21	Anak	16	84.5	27.4	3.22	1.54
		Hubbard	17	95.0	35.4	2.97	1.82
		Starbro	18	78.0	21.8	3.75	1.34
	17	Anak	19	101.2	33.2	3.25	1.96
		Hubbard20	124.6	51.0	2.61	2.35	
		Starbro	21	75.8	24.7	3.48	1.95
27	19	Anak	22	106.8	34.2	3.26	1.69
		Hubbard	23	104.0	35.2	3.11	1.80
		Starbro	24	88.9	22.5	4.15	1.35
	21	Anak	25	114.9	37.9	3.17	1.56
		Hubbard	26	101.8	37.1	2.84	1.71
		Starbro	27	71.9	20.1	3.80	1.32

[a]Significant ($P<0.01$) starter diet x finisher diet x genotype interaction in feed intake
[b]No significant ($P>0.05$) starter diet x finisher diet x genotype interaction in daily weight gain, FCR and PER.

nor finisher diet significantly affected feed intake, weight gains and FCR, but finisher diet had a significant ($P<0.01$) effect on PER. Chicks finished on the 17% CP diet had a significantly ($P<0.01$) better efficiency of converting protein consumed to body weight than those finished on 19 or 21% CP.

Table VII summarizes the effects of starter diet and genotype on performance of the chicks during the finishing phase. There were no significant starter diet x genotype interactions and no significant starter diet effects on feed intake, weight gains, FCR and PER. But genotype had significant ($P<0.01$) effects on all the parameters measured. Anak and Hubbard chicks consumed significantly ($P<0.01$) more feed, gained significantly ($P<0.01$) more weight and had superior ($P<0.05$) FCR than Starbro chicks. Hubbard also had superior ($P<0.01$) PER than

Table VI: Starter diet x finisher diet effects on performance of broiler chicks in experiment 2

	Starter diet (% CP)	Finisher diet (% CP)				
		17	19	21	Mean	S.E.
Daily feed intake (g)	23	96.1	88.1	103.3	96.0	
	25	104.4	99.6	85.8	96.6	
	27	100.5	99.9	96.2	98.9	
	Mean	100.5	95.9	95.1		0.59**
Daily weight (gain (g)	23	33.7	31.9	35.3	33.6	
	25	35.6	34.7	28.2	32.8	
	27	36.3	30.6	31.7	32.9	
	Mean	35.2	32.4	31.7		3.39NS
Feed conversion ratio (g feed/g gain)	23	3.04	2.99	3.19	3.07	
	25	3.17	2.99	3.31	3.16	
	27	3.11	3.51	3.27	3.30	
	Mean	3.11	3.16	3.26		0.22NS
Protein efficiency ratio (g weight gained/g protein consumed	23	2.08	1.94	1.58	1.86	
	25	1.99	1.84	1.57	1.80	
	27	2.09	1.61	1.53	1.74	
	Mean	2.05	1.80	1.56		0.13a

**Significant (P<0.01) starter diet x finisher diet interaction but no significant starter diet or finisher diet effect.
NSNo significant starter diet x finisher diet interaction and no significant starter diet or finisher diet effect.
aSignificant (P<0.01) finisher diet effect but no significant starter diet effect and no significant starter diet x finisher diet interaction.

Table VII: Starter diet x genotype effects on performance of broiler chicks (experiment 2)

	Starter diet (% CP)	Genotype				
		Anak	Hubbard	Starbro	Mean	S.E.
Daily feed intake (g)	23	102.5	103.3	82.2	96.0	
	25	104.4	103.0	107.3	79.5	96.6
	27	107.7	110.1	78.9	98.9	
	Mean	104.4	106.9	80.2		0.59a
Daily weight (gain (g)	23	35.6	41.4	24.0	33.6	
	25	33.3	40.0	24.5	32.8	
	27	35.1	41.1	22.5		32.9
	Mean	34.6	41.0	23.7		3.39a
Feed conversion ratio	23	2.97	2.65	3.60	3.07	
	25	3.22	2.79	3.46	3.16	
	27	3.23	2.85	3.81	3.30	
	Mean	3.14	2.76	3.62		0.22a
Protein efficiency ratio	23	1.85	2.16	1.58	1.86	
	25	1.71	2.01	1.67	1.80	
	27	1.74	1.96	1.54	1.74	
	Mean	1.77	2.04	1.60		0.13a

aSignificant (P<0.01) genotype effect but no significant starter diet and no significant starter diet x genotype interaction.

Table VIII: Finisher diet x genotype effects on performance of broiler chicks (experiment 2)

	Finisher diet (% CP)	Genotype				
		Anak	Hubbard	Starbro	Mean	S.E.
Daily feed intake (g)	17	107.6	109.8	84.2	100.5	
	19	103.2	105.2	79.2	75.9	
	21	102.4	105.7	77.2	95.1	
	Mean	104.4	106.9	80.2		0.59[a]
Daily weight (gain (g)	17	35.2	44.5	25.8	35.2	
	19	34.8	39.5	23.0	32.4	
	21	33.9	39.0	22.2	31.7	
	Mean	34.6	41.0	23.7		3.39[a]
Feed conversion ratio	17	3.22	2.56	3.55	3.11	
	19	3.02	2.83	3.64	3.16	
	21	3.18	2.91	3.70	3.26	
	Mean	3.14	2.76	3.63		0.22[a]
Protein efficiency ratio	17	1.92	2.41	1.83	2.05	
	19	1.82	1.97	1.60	1.80	
	21	1.57	1.75	1.36	1.56	
	Mean	1.77	2.04	1.60		0.13[a]

[a]Significant ($P<0.01$) genotype effect but no significant finisher diet and no significant genotype x finisher diet interaction.
[b]Significant ($P<0.01$) finisher diet and genotype effects but no significant finisher diet x genotype interaction.

Anak and Starbro chicks. At 11 weeks of age when the experiment was terminated, Anak and Hubbard chicks weighed significantly ($P<0.01$) more than Starbro chicks. Average weights of Anak, Hubbard and Starbro chicks at 11 weeks of age were 2.05, 2.12 and 1.34 kg, respectively.

Table VIII summarizes the interactive effects of genotype and finisher diet on performance of the chicks. There were no significant genotype x finisher diet interactions in feed intake, weight gains, FCR and PER.

Discussion

The results of experiment 1 suggest that Anak chicks require up to 27% CP in their starter diets while Hubbard and Starbro chicks require not more than 25% CP in their starter diets. This difference in CP requirement among different genotypes of broiler chicks might be related to inherent differences in their abilities for body weight gain and body protein accretion. Anak chicks grew faster and had heavier weights at six weeks of age than Hubbard and Starbro chicks. It had been recommended that diets of starting broiler chicks in the tropics should contain not less than 24% CP[3] while other workers[6] had recommended a range of 24.5 to 26% CP. The results of the present study suggest that for most broiler genotypes, diets containing up to 25% CP would be adequate but that very rapidly growing strains such as Anak would require up to 27% CP in their starter diets.

The non-significant effect of dietary CP ranging from 17 to 21% on feed intake, weight gains and FCR of finishing broiler chicks in experiment 2 supports the results of a previous study[6] in which no significant correlations were found between CP levels of 18, 20 and 22% and feed intake, weight gains and FCR of finishing broiler chicks. A diet containing 17% CP and 3.2 Mcal ME/kg was

adequate during the finishing phase for the strains of broiler chicks used. This CP level is lower than the 20 to 22% CP previously recommended[6] in the same environment, although the previous workers used Shaver x Starbro chicks.

There were no significant carryover effects of CP level in starter diet on growth rate and FCR during the finishing phase. But there were significant genotype effects on all parameters measured, with Anak and Hubbard chicks performing consistently better than Starbro. Differences among broiler genotypes in their response to dietary CP levels had been reported previously[7,8]. These differences have been attributed to inherent differences in their requirement for individual amino acids[2,9,10,11] or in their efficiency of metabolism of essential amino acids[12].

References

1. National Research Council (1977). *Nutrient requirements of poultry,* National Academy of Science Washington D.C. p. 29.
2. Scott, M.L., Nesheim, M.C. and Young, R.J. (1976). *Nutrition of the chicken* 2nd ed. Scott and Associates, Ithaca, New York. p. 79.
3. Babatunde, G.M. and Fetuga, B.L. (1976). *Niger. J. Anim. Prod.* **3 (1)**:126.
4. Olomu, J.M. (1976). *Niger. J. Anim. Prod.* **3 (1)**:177.
5. Olomu, J.M. and Offiong, S.A. (1980). *Poult. Sci.* **59 (4)**:828.
6. Okorie, A.U., Anugwa, F.O.I. and Osakwe, I.I (1988). *Arch. Geflugelk* **52 (2)**:55.
7. Hullan, H.W., Proudfoot, F.G., Ramey, D. and McRae, K.B. (1980). *Poult. Sci.*, **59**:748.
8. Hullan, H.W. and Proudfoot, F.G. (1981). *Poult. Sci.* **60(1)**:172.
9. Nesheim, M.C. (1968). *Federation Proc.* **27**: 1210.
10. Nesheim, M.C. and Hutt, F.B. (1962). *Science* **137**:691.
11. McDonald, M.W. (1957). *Austr. J. Agric. Res.* **8**:587.
12. Sorenson, P. (1977). *Br. Poult. Sci.* **18**:625.
13. Snedecor, G.W. and Cochran, W.G. (1974). *Statistical Methods* 6th ed. Iowa State University Press, Ames, Iowa.

Received for publication on 24th March 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 77-88

A STUDY OF THE GROWTH AND NUTRITIVE VALUE OF THE MAIZE PLANT

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UNE ETUDE SUR LA CROISSANCE ET LA VALEUR NUTRITIVE DU MAÏS

Résumé

On a mené une expérience pour examiner les changements relatifs au rendement, à la valeur nutritive et à l'apport des diverses parties de la tige de maïs comme résidus de récolte.

La variété de maïs utilisée était le "Legon composite 4". Les grains étaient récoltés six fois à trois semaines d'intervalles, la première récolte étant faite trois semaines après avoir planté le maïs. Lors de la récolte, les diverses parties de la tige étaient séparées, séchées à 70°C pendant 48 heures en vue d'évaluer le rendement en matière sèche (RMS), passées au tamis de 1mm puis analysées pour déterminer la teneur en protéine brute, les éléments constitutifs de la paroi cellulaire et la digestibilité *in vitro* de la matière sèche (DIVMS).

RMS de toute la plante a augmenté ($P<0,01$) à mesure que la plante grandissait et il a atteint un maximum de 11,8 mt/ha^{-1} quinze semaines après avoir planté le grain. Le rendement en résidus de récolte a aussi atteint le maximum de 9 mt ha^{-1} après 12 semaines, puis il a baissé à 6,9 mt ha^{-1} après 18 semaines. Les rendements en grain étaient respectivement de 2,8 et 4,7 mt ha^{-1}. RMS était composé de grains, de feuilles, d'enveloppes, de tiges, d'aigrettes, d'épis, de cosse et de faisceaux de soies qui constituaient 40; 6; 9; 22; 2 et 11% respectivement à la récolte finale. Le rapport résidu de récolte/grain était de 1,5:1.

La teneur en protéine brute des résidus de récolte et des différentes parties de la tige a diminué ($P<0,05$) à mesure que la plante grandissait et elle variait entre 1 et 4,9% à la récolte finale.

Les élements constitutifs de la paroi cellulaire des résidus de récolte et ceux des différentes parties de la tige ont augmenté ($P<0,01$) à mesure que la plante grandissait.

DIVMS des résidus de récolte et des diverses parties de la tige ont diminué également ($P<0,01$) à mesure que la plante grandissait. La valeur DIVMS des résidus de récolte était de 56%, tandis que celles des diverses parties de la tige oscillaient entre 49 et 65% après 18 semaines.

Summary

An experiment was conducted to examine the changes in yield and nutritive value as well as the contribution of the various maize plant fractions to that of whole maize crop residue with growth.

The maize variety used was the "Legon Composite 4". The plants were harvested six times at three week intervals beginning at three weeks post-planting. At harvest, the plants were separated into the various plant fractions, dried at 70°C for 48hrs. for dry matter yield (DMY) determination, ground through a 1mm sieve and analyzed for crude protein, cell wall constituents and *in vitro* dry matter digestibility (IVDMD).

DMY of the whole plant increased ($P<0.01$) with advancing age reaching a maximum of 11.8mt ha^{-1} at the eighteenth week. Grain yields were 2.8 and 4.7mt ha^{-1} respectively. DMY was made up of grains, leaves, leaf sheaths, stems, tassel, cobs, husk and silk which constituted 40, 6, 9, 22, 2 and 11% respectively at the final harvest. The ratio of the crop residue to grain was 1.5:1.

Crude protein content in the whole crop residue and the various morphological fractions decreased ($P<0.05$) with advancing growth, and ranged between 1.0 and 4.9% at the final harvest.

Cell wall constituents of the whole crop residue and those of the various morphological fractions increased ($P<0.01$) with advancing growth period.

IVDMD of the whole crop residue and the morphological fractions also decreased ($P<0.01$) with advancing growth. The IVDMD value of the whole crop residue was 56% while those of the various morphological fractions ranged between 49 and 65% at the eighteenth week.

INTRODUCTION

Maize is one of the major crops grown by most of the peasant farmers in the West African sub-region. As a cereal crop, it is also a good forage crop, exceeding some of the conventional forage crops[23]. However, the importance of maize as a

grain crop has tended to overshadow its value for processing as fodder and silage[3].

The total maize production levels in the West African Sub-region in 1985 is reported to be between one thousand and three million metric tons[8,15]. Powell[24] had also estimated the straw; grain ratio of maize to be 2:1, which means that twice as much straw as grain that could be used as animal feed was produced.

In the West African Sub-region it is not known how much of the crop residues are used as livestock feed. Nevertheless, it is acknowledged that some of the crop residues are used for animal feeding but the bulk of them is burned or left to waste[13,14].

The maize crop residue consists of various plant fractions which have different digestibilities[9,12,18], and hence different nutrient contents. Furthermore, grazing animals select the most palatable fractions of the total crop residue[24]. Thus, a more efficient utilization of the crop residues could be made if one has a knowledge of the nutritive value and the contribution of the various plant fractions that make up the crop residue.

A study was therefore carried out to examine the contribution and changes in the nutritive value of the various maize plant fractions and the maize crop residue.

Materials and Methods

Location, Climate and Soils

The experiment was carried out at the Department of Animal Science, University of Ghana, Legon, Ghana. The area has a sub-humid climate. The total annual rainfall is 934.2mm and it is bi-modal in character. The major rainy season being in March/April and ends in July, while the minor rainy season lasts between September and November. Temperatures are fairly uniform with a maximum and minimum of 32.5 ± 1.7°C and 27.7 ± 1.1°C, respectively. Relative humidities are high during the rainy season being 90 - 100% but may drop to about 40% or below during the dry season. Potential evaporation is about 1800mm p.a.

The soil is part of the Nyibenya-Hacho Complex, which is light textured clay and free draining[2,19].

Cultivation and Harvesting

The field was ploughed, harrowed and divided into three plots each of 4.5m x 3.50m. Seeds of maize (Zea mays) of variety "Legon Composite 4" (obtained from the Crop Science Department, University of Ghana, Legon) were sown at the rate of three seeds per hill. Planting distances were 0.75m between rows and 0.25m within rows. Two weeks after planting, the seedlings were thinned to one plant per hill, and a compound fertilizer (20kg each of N, P_2O_5 and K_2O applied. Six weeks after planting, the field was top-dressed with sulphate of ammonia (to provide 10.5kg N). The field was hand irrigated at regular intervals, and weed clearing was done manually using hoe and cutlass.

Harvesting of plants began at three weeks after planting and was done at three weeks intervals. At harvest, six plants from each plot were randomly selected, cut at ground level and separated into leaves, leaf sheaths, stem, tassel, husk and silk, cobs and grains. These plant fractions were then dried in the oven at 70°C for more than 48hrs and weighed. The dried samples were then bulked for the respective plant fractions, ground to pass through a 1mm sieve using a Wiley Mill and stored until analysis.

Chemical Analysis and Digestibility

The ground samples were analyzed for crude protein content[1], neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin[16] and also *in vitro* dry matter digestibility (IVDMD)[21].

Statistical Analysis

The data was subjected to Analysis of Variance test[26]. Duncan's Multiple Range Test was used to separate the means.

Results

The dry matter yield of the whole maize plant increased significantly ($P<0.01$)

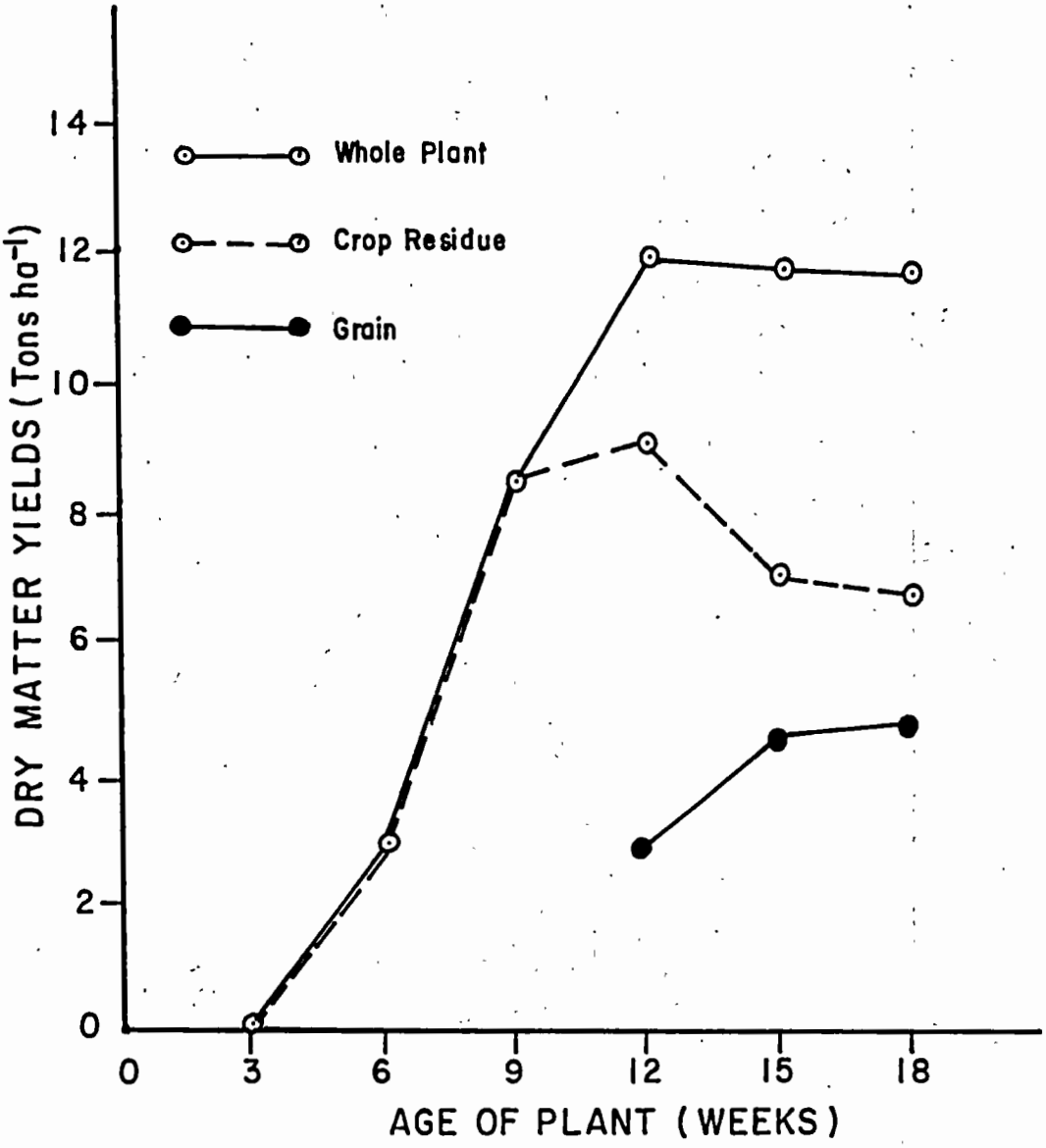

Fig.1 Changes in Dry Matter Yield of whole Maize Plant, Grain and Crop Residue with Age.

with advancing growth period (Fig. 1). It reached a maximum of 11.8mt ha^{-1} both at the fifteenth and the eighteenth week.

Crop residue i.e. whole maize without the grain, was 9.0mt ha^{-1} at the twelfth week but decreased thereafter to 7.1 and 6.9mt ha^{-1} at the fifteenth and eighteenth week respectively. Grain yield on the contrary increased from 2.8mt ha^{-1} at the twelfth week to 4.5 and later 4.7mt ha^{-1} at the fifteenth and eighteenth week respectively. Consequently the crop residue: grain ratio varied from 2.3:1 at the twelfth week to 1.47:1 at the eighteenth week.

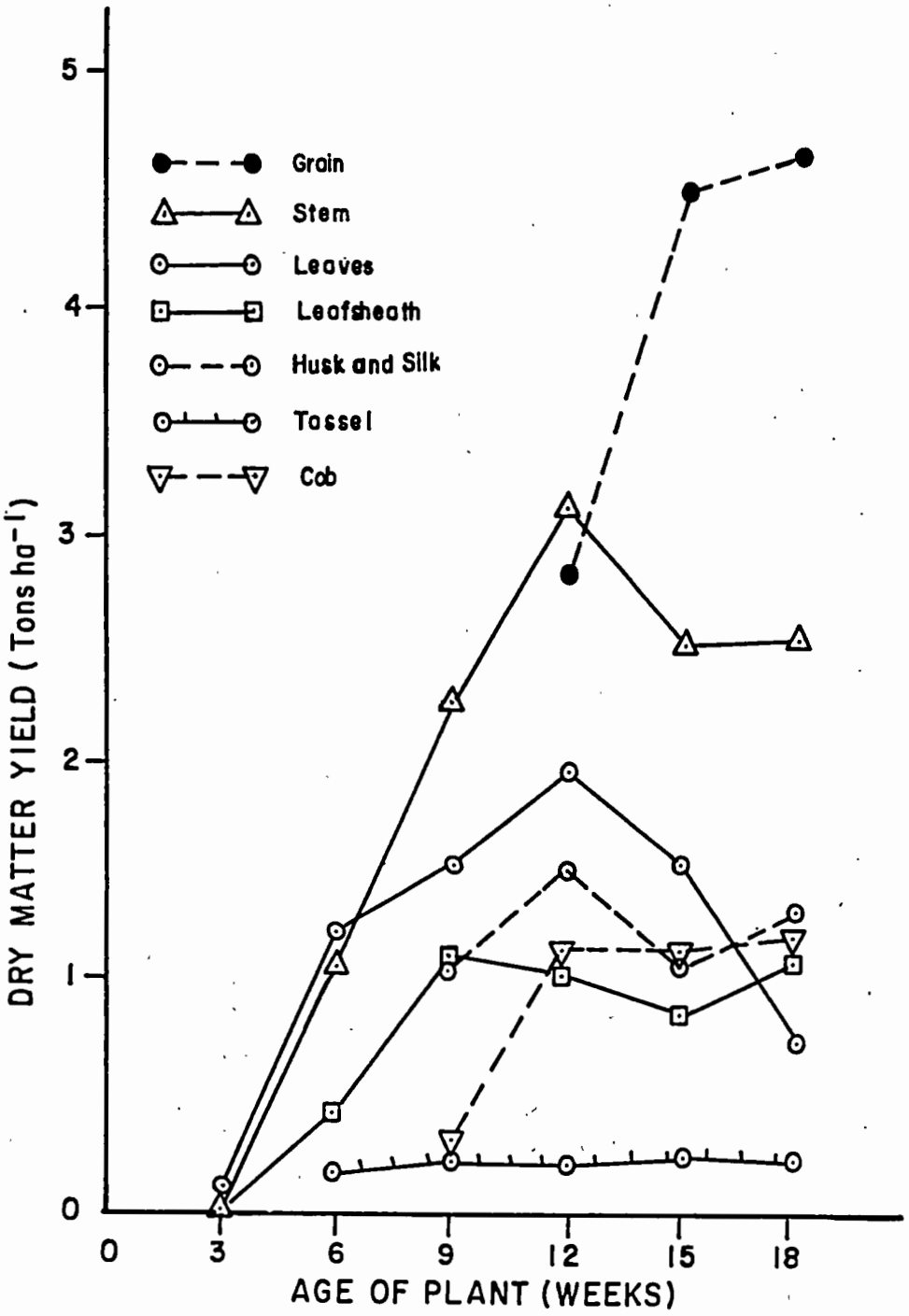

Fig. 2 Changes in Dry Matter Yield of Maize Plant Morphological Fractions with Age.

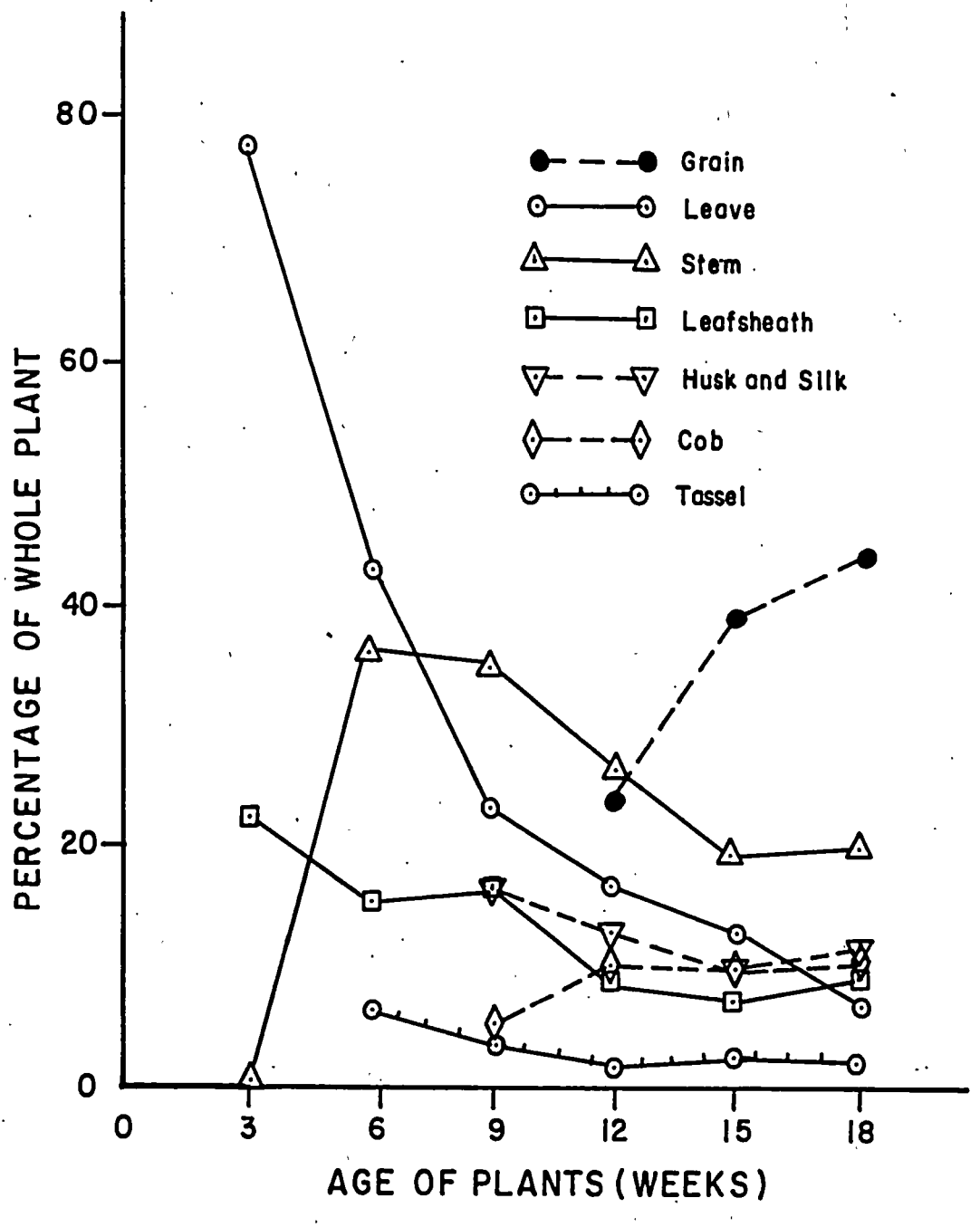

Fig. 3 Percentage Distribution of the Maize Plant Fractions with Age

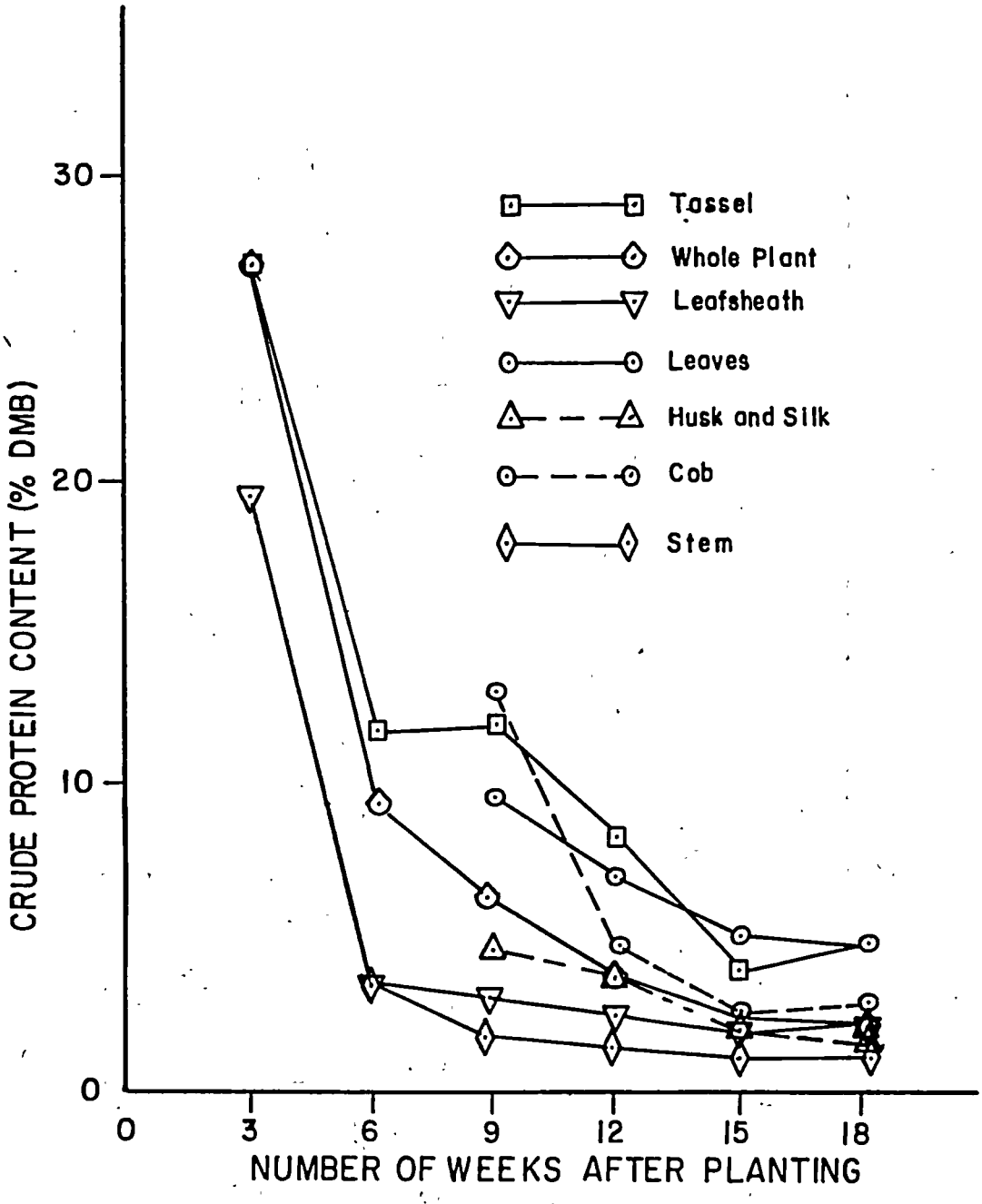

Fig. 4 Changes in Crude Protein Content of Whole Maize and its Botanical Fractions with Age

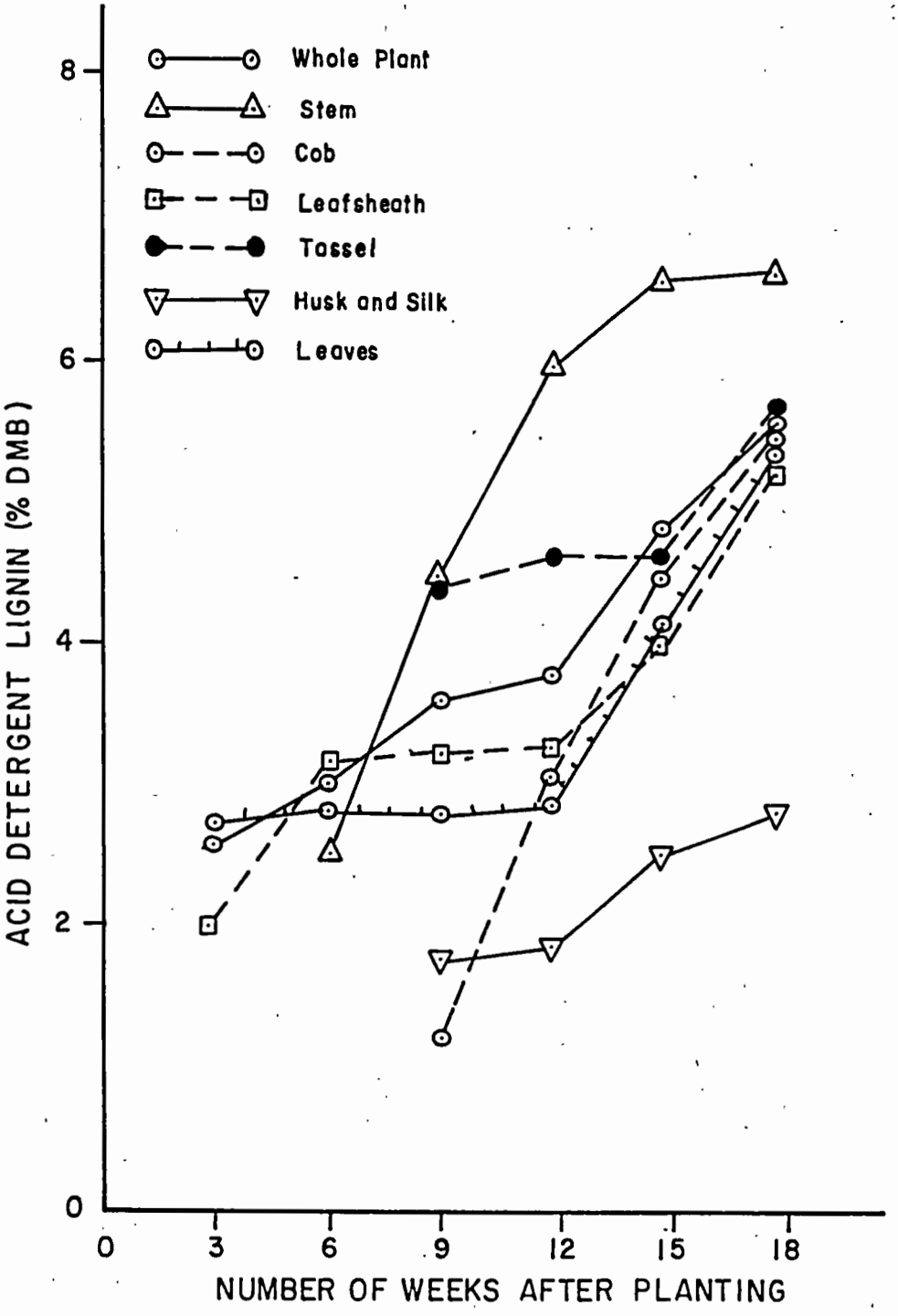

Fig.5 Changes in Lignin Content of whole Maize Crop Residue and its Botanical Fractions with Age

The dry matter yield of the morphological fractions of the maize plant are shown in Fig. 2. Leaves increased continuously from 0.125mt ha^{-1} three weeks post-planting to a peak of 1.95mt ha^{-1} at the twelfth week and then decreased to 0.75mt ha^{-1} at the eighteenth week. Leaf sheaths increased from 0.04mt ha^{-1} at three weeks post-planting to 1.11mt ha^{-1} at the nineth week thereafter remaining somewhat stable. Stems increased from 0.001mt ha^{-1} at the initial harvest to a peak of 3.1mt ha^{-1} at the nineth week. It then declined to a stable level of 2.5mt ha^{-1} at the fiftenth and the eighteenth week. The yield of the tassel increased slightly from 0.175mt ha^{-1} at the sixth week to 0.225mt ha^{-1} at the eighteenth week. The cob also increased from 0.325mt ha^{-1} at the nineth week to a stable peak of 1.175mt ha^{-1} at the twelfth week. The husk and silk vacillated in yield but on the average was about 1.26mt ha^{-1}.

The proportional distribution of these morphological fractions are shown in Fig. 3. At three weeks post-planting the plant consisted of 77% leaves, 22% leaf sheaths and 1% stems. With advancing growth period, the proportion of leaves and leaf sheaths decreased while that of stems increased. Thus, by the sixth week, the leaves, leaf sheath and stems were 43%, 14% and 37% respectively.

The tassel, which has appeared by this time, formed 6% of the total dry matter. The cobs, husk and silk appeared from the nineth week onwards while the grain appeared from the twelfth week. Thus, by the eighteenth week the grains, leaves, leaf sheaths, stems tassel, cobs, husk and silk constituted 40,6,9,22,2,10 and 11%, respectively.

Crude Protein Content

Crude protein contents of the whole maize plant and that of the various botanical fractions are shown in Fig. 4. Crude protein content decreased ($P<0.05$) with increasing growth period. For example, the crude protein content of the whole maize plant which was 27.2% at three weeks post-planting decreased to 2.3% by the eighteenth week.

There were also differences in the crude protein contents of the various botanical fractions. The leaves had the highest crude protein content at the early stages of growth (three and six weeks post-planting) but was later superceeded by that of the cob at nine weeks post-planting and then the grains (data not shown) at twelve weeks post-planting. The stem had the lowest crude protein content. The crude protein content of the grains was greater than that of the whole maize crop residue or any of the botanical fractions at any harvest. By the eighteenth week the crude protein contents were as follows: grain, 10.2%; tassel, 4.9%; leaves, 4.9%; leaf sheath, 1.4%; stems, 1.0%; cob, 3.0%; husk and silk, 2.4% and the whole maize plant, 2.4%.

Neutral Detergent Fibre

Neutral detergent fibre content of the whole maize crop residue and the various plant morphological fractions are shown in Table 1. There was a significant ($P<0.01$) increase in the various morphological fractions with advancing growth period.

Even though some differences were observed among the various plant morphological fractions, these differences were not significant ($P<0.05$). By the eighteenth week however, leaves had the lowest NDF content (66%) followed by those of stems, leaf shealth and tassel (75-78%) with the husk together with the silk and the cob having the highest NDF content (85% and 88%) respectively.

Acid Detergent Fibre

Table 2 shows the acid detergent fibre content of whole maize crop residue and those of the various plant morphological fractions. There was a significant ($P<0.01$) increase in the ADF contents with advancing growth period.

On the contrary, while some variations were observed among the different plant fractions, particularly at the initial growth phase, these differences narrowed up considerably with advancing growth period. Consequently no significant differences were observed among the various morphological fractions by the eighteenth week by the hust together with

Table 1: Neutral Detergent Fibre Content of Maize Crop Residue and its Botanical Fractions

PLANT PART	WEEKS AFTER PLANTING							
	3	6	9	12	15	18	$\bar{X}^a$	S.E.[b]
Whole Crop Residue	53.1	57.8	59.8	69.1	80.2	78.7	66.5	11.3
Leaves	53.1	60.9	68.6	71.1	72.9	66.5	65.5	7.4
Leaf Sheath	56.5	66.9	65.7	70.4	78.9	75.2	68.9	7.9
Stem	–	51.3	52.1	61.8	78.1	77.6	64.2	13.1
Tassel	–	–	65.3	80.2	81.6	77.4	76.1	7.4
Husk and Silk	–	–	61.0	68.6	88.1	85.3	75.8	13.1
Cob	–	–	44.8	69.6	86.9	88.2	72.4	20.2

a=Mean of all harvest
b=Standard error of the mean.

Table 2: Acid Detergent Fibre Content of Maize Crop Residue and its Botanical Fractions

PLANT PART	WEEKS AFTER PLANTING							
	3	6	9	12	15	18	$\bar{X}^a$	S.E.[b]
Whole Crop Residue	25.2	32.8	35.8	37.0	45.4	49.6	37.7	8.6
Leaves	24.7	33.3	35.1	36.2	44.2	48.6	37.0	8.4
Leaf Sheath	30.2	36.2	39.7	39.5	45.2	49.3	40.0	6.7
Stem	–	36.2	37.1	38.7	49.6	51.0	42.5	7.2
Tassel	–	–	35.1	44.4	44.7	54.9	44.8	8.1
Husk and Silk	–	–	24.7	28.8	39.8	39.8	33.3	7.7
Cob	–	–	13.2	32.4	40.1	40.5	31.6	12.8

a=Mean of all harvest
b=Standard error of the mean.

Table 3: In Vitro Dry Matter Digestibility of whole crop Residue and those of the Morphological Plant Fraction

PLANT PART	WEEKS AFTER PLANTING							
	3	6	9	12	15	18	$\bar{X}^a$	S.E.[b]
Whole Crop Residue	76.8	75.9	71.0	65.9	58.1	55.7	67.2	8.9
Leaves	76.1	72.1	71.3	67.2	67.6	65.1	69.9	4.0
Leaf Sheath	81.9	70.3	70.7	64.0	61.3	59.9	68.0	8.2
Stem	–	79.6	64.2	61.8	53.0	52.0	62.1	11.1
Tassel	–	84.0	57.4	51.5	54.5	48.6	59.2	14.2
Husk and Silk	–	–	78.7	79.9	72.3	66.0	74.2	6.4
Cob	–	–	72.5	75.9	53.8	51.9	63.5	12.4

a=Mean of all harvest
b=Standard error of the mean.

the silk and the cobs had considerably lower ADF content (<40%) than the rest.

Acid Detergent Lignin

Acid detergent lignin content of the whole maize crop residues and those of the various morphological fractions are shown in Fig. 5. There was a significant ($P<0.01$) increase in the ADL contents of both the whole crop residue and in all the various morphological fractions.

The ADL contents of the leaves, leaf shealth, stem, tassel and cob did not differ significantly ($P>0.05$) from each other, but they were significantly higher ($P<0.05$) than that of the husk with silk. The stem generally had the highest ADL content, being 6.7% by the eighteenth

week after planting. This was followed by those of tassel. Cobs, leaves and leaf sheaths which were about the same (5.4 - 5.7%) and the least of all was that of the husk with silk (2.5%).

In vitro Dry Matter Digestibility

In vitro dry matter digestibility (IVDMD) of the whole crop residue and those of the various plant morphological fractions are shown in Table 3. IVDMD of the whole crop residue and the morphological fractions significantly ($P<0.01$) decreased with advancing growth period.

Except for the initial harvest of leave sheaths (at three weeks after planting), stems and tassel (at six weeks post-planting) which were higher in IVDMD than that of leaves, the latter had higher IVDMD than these plant fractions at all other times. The cob had higher IVDMD than the leaves, leaf sheaths, stems and tassel at the nineth and twelfth weeks but decreased to the level of the stems and tassel later. The husk together with the silk also had higher IVDMD than the leaves between the nineth and the fifteenth week but was about the same by the eighteenth week.

Discussion

The dry matter yield increase with increasing growth period has also been reported by others[10,20]. The total above-ground dry matter yield at the twelfth and eighteenth weeks were similar to that reported by Fleischer *et al.*[10], and just as was observed in the present experiment, Fleischer *et al.*[10], also observed some decrease in the total dry matter yields in later harvests. Similar losses in plant tops had been reported by others in the literature[31].

In the present experiment, the crop residue yield were similar to, but the grain yields were higher than, that reported by Fleischer *et al.*[10]. The apparent difference in grain yield was however not immediately clear. However, the higher grain yield of 4.7mt ha^{-1} was found to be lower than the potential maximum of 6.5mt ha^{-1} indicated by Dadson[4]. Also the crop residue: grain ratio at the eighteenth week was comparable to that obtained by Fleischer[10] but lower than that reported by Powell[24].

The dry matter yields of the various morphological fractions were comparable to those reported by Fleischer *et al*[10]. Under the present traditional farming systems where most crop residues are left to rot on the farm, the farmer may harvest and transport some of these crop residue parts (at least cobs, husk and silk which totals 2.4mt ha^{-1} or some 35% of the total crop residue) as incidental to grain harvesting without any extra cost.

The changes observed in the proportional distribution of the morphological fractions followed a pattern similar to that in the other species of the grass family[9,12]. These changes were due to the shift in the source-sink relations in the distribution of photosynthates[28]. Consequently even though an increase in the total dry matter yield is observed with increasing growth period (Fig. 1), this was accompanied by an increase in the yield of the various plant fractions (Fig. 2) and hence changes in the proportions of these morphological fractions (Fig. 3).

That there was a decrease in the crude protein content with advancing growth period in both the whole crop residue and also in the various morphological fractions is in agreement with published results[9,10,25,32]. This decrease in protein content was due partly to the increase in size of the plant during which photosynthates are mainly diverted into the production of low-nitrogen containing fractions[17,21] of which the stems, leaf sheaths and cobs form the bulk.

Furthermore, the difference in the various plant fractions is similar to the results of Fleischer[9] with green panic and rhodes grass and it is due to the functional differences of these plant fractions[7].

There was an increase in cell wall constituents with advancing growth period. Such increases have also been reported by other workers[10,26] and are due to changes in the morphological composition of the whole crop residue the bulk of which is made up of stems, leaf sheaths and cobs.

The variation in the different plant fractions has also been observed in other grass species[9]. These variations are due to the functional differences in the various plant morphological fractions[7].

The decline in digestibility with advancing growth period is in accordance with much of the published literature[5,10,26,29]. This decrease in IVDMD was due not only to the increase in cell wall constituents but also to the gross changes in the proportion of the various plant fractions.

Furthermore, the variations among the IVDMD of the different plant fractions also followed trends observed with other grasses[9,18]. These variations were due to differences in the cell wall constituents of the various plant fractions[18] as a result of differences in functional role in the whole plant[7]. On the contrary the observation that in the early stages leaf sheaths had higher IVDMD than the leaves was probably due to the environmental temperature effects[5,11] since at this period the leaves while shading the leaf sheaths were exposed to the direct influence of the sun.

Conclusion

Dry matter yield of maize crop about ground increased with advancing growth period up to 11.8mt ha^{-1} at the eighteenth week. This was made up of 6.9mt ha^{-1} of crop residue and 4.7mt ha^{-1} of grains ratio varied from 3.2:1 at the twelfth week to 1.47:1 at the eighteenth week. There were variations in the pattern of changes in the dry matter yield of the various morphological fractions of the maize crop.

Crude protein content decreased with advancing growth period in the whole maize crop residue and also the various plant morphological fractions. The highest crude protein content of about 5% at the eighteenth week was found in the leaves and the tassel.

Cell wall constituents however increased in the whole maize crop residue and the morphological fractions with advancing growth period. There was variation in the cell walls constituents of the various morphological fractions.

In vitro dry matter digestibility decreased with advancing growth period in both the whole maize crop residue the various morphological fractions. At the final harvest however, it was highest in the leaves and husk, and lowest in the tassel and stem.

Acknowledgement

The authors are greatly indebted to Professor R.K.G. Assoku of the Department of Animal Science for his constructive criticism during the writting up of the paper. They are also very grateful to Mr. Abubakari Yacubu for helping with the chemical analyses and *in vitro* dry matter digestibility trials.

They are very grateful again to the University of Ghana Research and Conferences Committee which made available funds for the research.

References

1. Association of Official Analytical Chemists (1975) *Official Methods of Analysis,* Washington D.C.
2. Brammer, H. (1967). *Soils of the Accra-Plains. Memoir,* Soil Res. Inst., Kumasi, **3**:1.
3. Bunting, E.S. (1966) *Outl. Agric.* **5**:104.
4. Dadson, R.B. (1975) *Universitas,* University of Ghana **4**:162.
5. Deinum, B. (1976). *In Carbohydrate Research in Plant and Animals.* Miscellaneous Paper No. 12, Agricultural University, Wageningen, 29.
6. Deinum, B. and J.G.P. Dirven (1971) *Neth. J. Agric. Sci.,* **19**:264.
7. Esau, K. (1965) *Plant Anatomy,* John Wiley and Sons, Lond., N.Y. 767 pp.
8. Ewusie, K. (1986) *Statistical Tables on the Economy of Ghana (1950-1985).* Inst. of Statistical, Social and Economic Research, University of Ghana, Legon, Ghana.
9. Fleischer, J.E. (1987) *Bull. Anim. Hlth. Prod. Afri.* **35**:229.
10. Fleischer, J.E., Anna R. Barnes,; B. Awumbila; K. Amaning-Kwarteng and C.K. Tieku (1987). Paper presented at the 4th ARNAB Workshop, Bamenda, Cameroon, 20-27/10/87.
11. Fleischer, J.E.; Y. Masuda and I. Goto (1983). *J. Fac. Agric.,* Kyushu University **28** (2,3): 83.
12. Fleischer, J.E.; Y. Masuda and I. Goto (1984) *J. Japan Grassl. Sci., 30 (1):20.*
13. *Food and Agriculture Organisation (1983). Intergrating Crops and Livestocks in West Africa.* F.A.O. Production and Health Paper 41, F.A.O., Rome, 112 pp.
14. Food and Agriculture Organisation (1986a). *Better utilization of crop Residues and By-products in Animal Feeding: Research Guidelines 2. A*

practical Manual for Research Workers. F.A.O. Animal Production and Health Paper 50/2. 154 pp.
15. Food and Agriculture Organisation (1986b). *Production year Book. Vol. 40,* F.A.O., Rome.
16. Goering, H.K. and P.J. Van Soest (1970). *Forage Fibre Analysis:* U.S.D.A. Husbandry, Jacket No. 379.
17. Greenwood, E.A.N.; D.W. Goodall and Z.V. Titmanis (1965). *Plant and Soil* **23**:97.
18. Hacker, J.B. and D.J. Minson (1981). *Herb. Abstr.,* **51**:459.
19. Jenik, J. and J.B. Hall (1976). *Folia Geobot. Phytotax.,* Praha, **11**:163.
20. Lutz, J.A.; Camper and G.D. Jones (1971). *J. Agron.* **63**:12.
21. Minson D.J. and M.N. McLeod (1972). The *In vitro* Technique: Its Modification for estimating digestibility for large number of tropical pasture samples. Australian C.S.I.R.O., Div. of Trop. Past. Tech. Pap. 8.
22. Lyttleton, J.W. (1973) In *"Chemistry and Biochemistry of Herbages"* (ed. by G.W. Butler und R.W. Bailey) Acad. Press, Lond., New York. Vol. **1**:63.
23. Morrison, F.B. (1961). *Feeds and Feeding.* Abridged 9th Edition., The Morrison Publishing Co., Ontario.
24. Powell, M. (1984). *Trop. Agric.* **62** (1):77.
25. Raymond, W.F. (1969). *Adv. in Agron.,* **21**:1.
26. Snedecor, G.W. and Cochran, W.G. (1976). *Statistical Methods.* The Iowa University Press. Ames, Iowa, U.S.A.
27. Struick, P.C. (1985). *Neth. J. of Agric. Sci.,* **33**:56.
28. Warreing, P.F. and J. Patrick (1975) *In "Photosynthesis and Productivity In Different Environments.* (Ed. by J.P. Cooper), Acad. Press, Lond.; New York, pp. 481.
29. Weber, D.W.; R.L. Velter and N. Gray (1970). *J. Anim. Sci.,* **31**:10.
30. Westselaar, R. and A.D. Farquhaar (1980) *Adv. in Agron.,* **33**:263.
31. Wilson, J.R. (1982) *In Nutritional Limits to Animal Production From Pastures.* (Ed. by J.B. Hacker), C.A.B., Farnham Royal, U.K., pp. 117.

Received for publication on 10th May 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 89-95

LONGEVITY, LIFE TIME PRODUCTION AND HEALTH OF CANADIAN HOLSTEIN-FRIESIAN CATTLE IN THE HUMID FOREST ZONE OF GHANA

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LONGEVITE, RENDEMENT TOTAL ET ETAT SANITAIRE DES BOVINS CANADIENS HOLSTEIN/FRISONS DANS LA ZONE FORESTIERE HUMIDE DU GHANA

Résumé

La longévité, l'état sanitaire et le rendement total de 35 génisses Holstein/Frisonnes importées du Canada pour une expérience d'élevage dans la zone forestière humide du Ghana ont fait l'objet d'études.

Les animaux avaient en moyenne une durée de vie de 2.680 jours pendant laquelle ils avaient 3,89 vêlages et produisaient 15.996 kg de lait en 1.562 jours. Le poids moyen était de 576 kg à l'âge moyen de 75 mois. Seuls 26% des 42 génisses que les animaux avaient produites leur ont survécu.

Tous les animaux souffraient de grippe des bovins à leur arrivée. En six mois, les maladies les plus prévalentes observées étaient la conjonctivite, l'infection généraliséo, la trypanosomiase et la pododermatite infectieuse. Six animaux sont morts au cours de cette période. Tout au long de leur vie, les maladies les plus fréquentes qui affectaient les animaux étaient les suivantes dans l'ordre: l'infection généralisée, la streptothricose, la mastite, la conjonctivite, la pododermatite infectieuse et la trypanosomiase.La streptothricose (40%) et la cowdriose (23%) étaient les causes courantes de mortalité.

Ces résultats montrent que les Holstein/Frisons sont très sensibles aux maladies bovines tropicales; par exemple, la streptothricose et la cowdriose dans les conditions d'élevage en pâturage. Des soins sanitaires intensifs doivent être dispensés à ces animaux pour leur permettre de survivre sous les tropiques humides.

Summary

The longevity, health and life time production of 35 Holstein-Friesian heifers imported from Canada into the humid forest zone of Ghana were studied.

The animals had an average life span of 2680 days during which they had 3.89 calvings and produced 15,996 km milk in 1562 days. A mean mature weight of 576 kg was attained at an average age of 75 months. Only 26% of 42 heifers produced by the animals survived them.

All animals suffered from shipping fever on arrival. Within 6 months the most frequent occuring diseases were conjunctivitis, systemic infection, trypanosomiasis and foul in the foot. Six animals died within this period. In the life span of the animals the order of frequent occuring diseases were: systemic infections, streptothricosis, mastitis, conjunctivitis, foul in the foot and trypanosomiasis. Streptothricos (40%) and heart water (23% were the frequent causes of death.

The results show that Holstein-Friesians are very susceptible to tropical cattle diseases e.g. streptothricosis and heart-water under grazing-conditions. Very good health care is required for these animals to survive in the humid tropics.

INTRODUCTION

Many tropical countries lacking local dairy breeds of cattle, but wishing to reduce their dependence on imported milk and milk products have imported exotic dairy cattle to set up farms[2,9,21]. The production of some of these imported dairy breeds have been reported[1,8,15,17]. There is, however, paucity of information on the control and incidence of diseases and longevity of these imported animals.

In 1974, the University of Science and Technology, Kumasi, Ghana with the assistance of the Canadian International Development Agency (CIDA) imported 35 Holstein-Friesian heifers into the humid forest zone of Ghana. The aim of the pro-

ject was to determine if exotic dairy cattle can survice and produce satisfactorily in this area. Preliminary reports on reproductive performance[10] and milk production[11] were reported.

The aim in this study is to provide further information on the longevity, lifetime production and health of these imported cattle which have all died.

Materials and Methods

Description of the data

Lifetime milk production and reproductive data on 27 of 35 heifers imported from Canada and which calved into one or more lactations were used in the study. Health, mortality and post-mortem data were based on all 35 heifers from 27th July, 1974, on arrival, to 19th December, 1983 when the last animal was disposed of.

Measurement on individual cows were milk production, live-weight, days open, days dry, disease incidence, treatments for diseases during the first six months, date of death or disposal and postmortem report.

The following define trait codes used in the tables:

Longevity	—days from birth to death
Herdlife	—days from first calving to end of last lactation
Milk yield-life	—Milk produced during life span of the animal
Mature weight	—The highest live weight immediately post-calving in the 4th and subsequent calvings.
Days open (%)	—Total days after first calving when animal was not in calf ÷ herd life.
Days dry (%)	—Total days when animal was not lactating after first calving ÷ herd life.
An Incidence of disease	—Was from diagnosis until end of infection
Systemic Infection	—Any infection which caused a rise in body temperature (fever) but which could not be diagnosed.

Management, Feeding, Housing and Health Care:

The management, feeding, housing and health care of the lactating and non-lactating animals were previously provided[10,11]. Below is provided the management, feeding, housing and health care of the animals on arrival.

The heifers arrived at the Boadi Cattle Project site on 26th July, 1974 and were confined in a cattle barn for the first two weeks. They were then moved to a yard adjacent to the barn where they were confined for another two weeks before being moved to a paddock of giant star grass (Cenchrus cilliaris).

It was observed that the animals were worried by dipterous flies resulting in reduced appetite. The cattle were therefore transferred back to the cattle barn after two had died of trypanosomiasis. Fly proofing and ceiling electric fans were provided at this stage in the barn. The fans were provided to improve the microclimate in the barn when it was realised that the heifers were heat stressed in the afternoons, resulting in profuse panting and salvation.

Since November, 1974 depending on health of the animals, both zero grazing and the turning out of the cattle to pasture over night have been practised. The animals were fed legume-grass forage in stanchions during zero grazing periods. This was supplemented, daily, with 4.5 kg concentrate/animal (made in different proportions from maize, rice bran, groundnut cake, brewers spent grain, common salt, bone meal, dicalcium phosphate and trace mineral premix). The concentrate had a total digestible nutrients (TDN) content of 73% and 16% crude protein. The concentrate supplement was gradually reduced to 2.7 kg/animal in April, 1975, then to 1.35/animal in early May and 0.45 kg/animal in early June, 1975. The heifers began to calve in June, 1975 and the feeding management

was changed to meet their lactation requirements[11].

The heifers were-inspected daily for signs of disease infection. This consisted of monitoring rectal temperatures, respiration rates and pulse rates. They were given Berenil (Farbwek Hoechst, AG Frankfurt, Germany) as prophylaxis against trypanosomiasis ten days after arrival and monthly after. After two weeks they were vaccinated against anthrax, black quarter and rinderpest. Routine haematological examination was carried out on all cattle monthly.

Results

Health

In Table 1 is provided incidence of diseases and treatments given in the first six months of arrival of the cattle. Table 2 indicates incidence of diseases during the life span of the animals.

As expected many disease problems were encounted within the first 6 months attributable to biting flies and ticks. All the animals suffered from shipping fever on arrival. The most frequent occuring disease was, however, conjunctivitis with lacrimation followed by systemic infection, trypanosomiasis and foul in the foot. All the heifers suffered from conjunctivitis, systemic infection and trypanosomiasis, but only 50% suffered from foul in the foot within the first six months. Antrycide sulphate and Hostamycin injections were used to treat trypanosomiasis infection when diagnosed. Systemic infection, conjunctivitis and shipping fever were ably treated with antibiotics (Hostamycin, Terramycin and combiotic) injections. There were 2 cases of abortions and 6 deaths occurred from trypanosomiasis, heart water, acute liver cirrhosis and plant poisoning during this period.

The incidence of diseases over the life span of the animals revealed the following order of frequency of occurrence of major ailments (Table 2): systemic infection[325], streptothricosis[121], mastitis[111], conjunctivitis with lacrimation[109], foul in the foot[72], trypanosomiasis[60]. There were also 15 cases of retained placenta,

Table 1: Incidence of diseases, infections and treatments of the heifers in first six months of arrival

Disease	*Percent of Animals Infected	Treatment
Shipping fever	100.00	Terramycin** injection
Trypanosomiasis	100.00	Antrycide sulphate injection + Terramycin injection.
Conjunctivitis	177.14	Hostamycin** or Terramycin injection + Terramycin all-purpose powder.
Systemic infection	134.29	Combiotic or Hostamycin or Terramycine injection.
Foul in the foot	50.00	Formalin sol. or Cuso4 Sol. Foot bath + Terramycin or Hostamycin injection.
Diarrhoea	28.57	Combiotic** or Streptopen** or Oxytetracycline injection.
Wounds and sores	20.00	Hostamycin injection + Gentian violet or Sulphanilamide dressing.
Skin lesions	2.86	Ascabiol emulsion + Combiotic injection.
Dermatitis	2.86	Hostamycin injection + Gentian violet dressing.
Suspected Actinomycosis	17.14	Hostamycin or streptopen injection
Bloat	2.86	Rumen puncture
Oedema of Jowl	14.29	Terramycin injection
Suspected Calcium deficiency	25.71	Calcium gluconate injection
Emaciation	8.57	Calcium gluconate injection
Abortion	5.71	Oxytetracycline or Hostamycin injection.
Death	17.14	Post-mortem examination

*Percentage greater than 100 indicate some animals were infected more than once.
**Terramycin, Hostamycin, Combiotic and Streptopen – Pfizer Co. Ltd., U.S.A.

Table 2: Incidence of diseases and infections during the life span of animals

CONDITION	NUMBER OF CASES
Shipping fever	35
Trypanosomiasis	60
Heart water	8
Systemic infections	325
Mastitis	111
Streptothricosis	121
Conjunctivitis with lacrimation	109
Babesiosis	48
Anaplasmosis	8
Diarrhoea	20
Mycotic dermatitis	22
Foul in the foot	72
Other skin infections	18
Bloat	7
Abscess	14
Oedema	27
Liver Cirrhosis	2
Plant Poisoning (Suspected)	1
Allergy	6
Actinomycosis	1
Actinomycosis (Suspected)	6
Retained Placenta	15
Prolapse of Uterus	1
Cystic Ovaries	3
Pneumonia	1
Hepatitis	2
Milk Fever	1
Uterine Infection	10
Abortions	7
Still births	9

10 cases of uterine infection with purulent vaginal discharges, 3 cases of cystic ovaries, 7 abortions and 9 cases of still births.

Most of the diseases diagnosed responded to treatments though some animals succumbed to these infections (Table 3). Most deaths or disposals were from streptothricosis[14] and heart water[8].

Longevity and Life time Production:

Means and standard deviations of longevity, herd life and life time production traits are provided in Table 4. The animals had an average life span of 2680 days (7.3 years) of which 1562 days (4.3 years) were productive. On average 23% and 37% of herdlife were days dry and days open, respectively. The animals had 3.89 calvings (range 1-7) during which they produced, on average, 15,996 kg of milk. An average mature weight of 576 kg was attained at an average age of 75 months. Forty two heifer calves were produced during the life span of the animals but only 26.2% survived their dams.

Discussion

The results reported here of the incidence of diseases and of the steps taken to control them provide guidelines for future management of imported Holstein-Friesian cattle in the humid tropics. There was paucity of information on the management of exotic cattle just imported into the tropics during the establishment of the present herd. This resulted in errors of management decisions and thus high mortality of the imported animals. The most serious of these errors were the grazing or exposure outside the barn, of the heifers within a few weeks of arrival and the transfer of Droughtmaster cattle, to the farm, from an area known to be prevalent in Cutaneous streptothricosis infection[16]. More publications, therefore, on the successful health management of exotic dairy cattle is necessary if dairy production in the tropics is to succeed using these imported animals.

It is apparent that the first six months after arrival is the most critical period in the survival of Holstein-Friesian cattle in the humid tropics (Tables 1 and 3). This is the period when they are most susceptible to tropical diseases while adapting to the environment. The drastic change in environment results in stress, with a decline in resistance to disease infection. Shipping fever is common at this time[12 and 21] and should be treated for. A high level of nutrition in the form of concentrate supplementation is required at this time when appetite may be low.

As was expected trypanosomiasis, a major disease of economic importance in Ghana[20], was prevalent in the herd within the first six months of importation. The disease was, however, controlled by early diagnosis and treatment with antrycide sulphate and prophylaxis with Samorin (May and Baker, England) or Berenil, given every month, and bush clearing. Similar results have been reported by Yeboah[21] from the Accra

Table 3: Yearly Mortality and disposal of imported Holstein-Friesians

YEAR	NUMBER OF DEATHS OR DISPOSALS	CAUSE OF DEATH OR DISPOSAL
1974	6	[a]2 - Trypanosomiasis
		2 - Patuerellosis – heart water complex
		1 - Suspected Plant poisoning
		1 - Acute liver cirrhosis
1975	1	1 - Heartwater
1976	1	1 - Slaughtered Cystic ovaries
1977	3	1 - Heartwater
		1 - Chronic hepatitis and acute bloat
		1 – Slaughtered Cystic ovaries
1978	4	1 - Destroyed Pneumonia
		1 - Heart water
		1 - Black quarter
		1 - Undiagnosed
1979	3	2 - Heart water
		1 - Destroyed liver cirrhosis
1980	1	1 - Destroyed Streptothricosis
1981	10	1 - Suspected Ketosis complex
		2 - Destroyed Streptothricosis
		1 - Slaughtered broken Sternun
		1 - Heart water
		5 - Streptothricosis
1982	4	1 - Destroyed Streptothricosis
		3 - Streptothricosis
1983	2	2 - Destroyed Streptothricosis

[a]Numbers involved.

Table 4: Means and standard deviations (SD) of longevity and life time production traits

Trait	Mean	SD
Longevity (days)	2680.3	654.3
Herd life (days)	1561.6	486.1
Number of calvings	3.89	1.34
Days dry (%)	20.03	9.80
Days open (%)	31.88	13.06
Mature liveweight (kg)	576	43
Milk yield – life (kg)	15996	5402
Number of females both in life time	42	–
Females surviving their dams (%)	26.02	–

Plains of Ghana.

The animals were very susceptible to heartwater and cutaneous streptothricosis although sprayed weekly with Delnav (Cooper McDowall and Roberts Ltd, England) or Bacdip or Asuntol (Bayer, AG Leverkusen, Germany). Heart water proved to be very difficult to diagnose in the live animal. Even when suspected from nervous symptoms (chewing movements, protrusion of the tongue and twitching of eyelids) and high temperature, animals failed to respond to antibiotic and sulphanamide therapy. Various other studies[9,21] from the tropics have observed that heartwater was a most serious disease problem of exotic cattle undergrazing conditions despite regular dipping. This suggests the need for planned studies on the prevention and cure of this disease.

Babesiosis and anasplasmosis supris-

ingly caused no mortalities in the herd in contrast to the report of[2]. This probably was due to the weekly spraying of animals and/or early diagnosis due to the regular haemotological studies.

Cutaneous streptothricosis (Dermatophilosis) proved to be the biggest cause of mortalities in the herd, agreeing with other studies. This disease is very wasting and responds slowly, if at all, to treatments with antibiotics and tropical application of iodine solution and palm oil, following a daily scrubbing to infected parts of the body. The disease also prolonged the calving interval because during the long period of recovery isolated animals were not bred when on heat (natural service was used). It also had a depressing effect on milk yields and was responsible for 3 cases of stillbirths/abortions. The disease was introduced with Droughtmaster cattle from the Accra Plains. It is therefore advisable to prevent exotic cattle using the same facilities with local cattle.

Systemic infection and bovine conjunctivitis occurred frequently in the herd, but were not fatal. Bovine conjunctivitis appears to be a disease affecting both native[13] and exotic cattle[3] at pasture. In the present study this disease was easily treated with terramycin eye ointment or powder.

Mastitis also occurred regularly in the herd despite the use of teat dips and individual napkins for cleaning udders. There was no serious economic loss in milk production due to this infection; animals responded quickly to antibiotic therapy.

Diseases of reproductive nature diagnosed were abortions, stillbirths, retained placenta, cystic ovaries and uterine infections resulting in purulent discharges from the vagina. The uterine infections could have contributed to the long calving intervals in the herd[10]. The causitive organisms were never diagnosed. The infection was treated with antibiotics.

The rates of stillbirths (8.6%) and abortions (6.7%) observed in this study were lower than those reviewed by[18]for the tropics and subtropics. The number of abortions and retained placenta (14.3%) are far higher than the 1.4 to 5.5% and 5.5 to 17.3% respectively reported for the University of Guelph dairy herd between 1969 to 1975[5]. No causal agents could be diagnosed for the cases of abortions and stillbirths except for three cases caused by streptothricosis infection cited above and two cases of abortions which occurred during the febrile stage of trypanosomiasis.

Other diseases, not unique to the tropics[7] which caused no mortalities, but were diagnosed were foul-in-the foot, milk fever, ketosis, diarrhoea and dermatitis (Table 2).

Despite the various infections reported, the animals survived and produced 99 live calves. Their longevity and life time productions (Tables 3 and 4) are comparable if not superior to reports from other tropical and temperate areas.

A review of the literature[18] showed that the herd life of Holsteins in Kenya and Panama were 3.65 and 3.07 years, respectively. While from Ohio, U.S.A.,[6] reported a herd life of 2.9 years (first calving to last calving: 3.6 years in this study), 3.68 calvings and 16329 kg life-time milk production. The animals in the present study, however, had a poorer reproductive performance (37.0 Vs 30.3% — ratio of days open to herdlife) compared to the results of the same authors[6]

Mature skeletal size of Holsteins is reached at 5 years of age, but increase in body weight continues until about 7 years[14]. The mature weight (Table 4) recorded in this study is slightly higher than the 517 kg life weight reported by[19] for Holsteins in the Venezuelan tropics but less than the 600 kg reported by[4]. This suggests that the tropical environment had a depressing effect on mature size.

The present results also suggest that small herds of exotic dairy cattle in this environment may not generate enough female replacements. This is probable incorrect. The offspring of the present herd matured at a time when streptothricosis infections was high resulting in high mortalities. Until the advent of this disease, there were few deaths of heifers born at the farm.

Dedication and expensive health management routines are necessary if Holstein-Friesian cattle are to survive and produce satisfactorily under grazing conditions in the humid tropics.

General implications from this study on the health management of imported Holstein-Friesian cattle in the humid tropics are summarised. There is the need:

1. To treat Holstein-Friesian cattle against shipping fever on arrival and to confine the animals in well ventilated barns for the first six months, with gradual introduction to pasture.
2. To zero graze if several animals are observed showing signs of disease infection.
3. To regular monitor rectal temperatures, respiration and heart rates, during the adaptive stage, for early diagnosis of disease infection.
4. To regular dip or spray at least more than once per week, for the control of tick borne diseases.
5. To regular carryout haemotological studies, prophylaxis and bush clearing for the control of trypanosomiasis.
6. To construct facilities for only exotic cattle to prevent transmittable diseases from native cattle.

Acknowledgement

The Management of the Boadi Cattle Project for allowing this paper to be published. I thank Miss Gladys A. Ndziba for typing the manuscripts.

References

1. Adeneye, J.A. and Adebanjo, A.K. (1978). *J. Agric. Sci. Camb.* **91**:645
2. Ajayi, S.A., Fabiyi, J.P. and Umo, I (1982). *Wld. Anim. Rev.* **43**:41.
3. Aruo, S.K. (1973). *Bull. Epiz. Dis. Afri.* **21**:331.
4. Canada (1971). Department of Agriculture, Ottawa. *Dairy Husbandry in Ghana.* Publication Number 1439, p. 81.
5. Canada (1976). University of Guelph. *Guelph Dairy Industry Research Report 1975 - 1976,* p. 58.
6. Gill, G.S. and Allaire, F.R. (1976). *J. Dairy Sci.* **59** :1131.
7. Hollon, B.F. and Branton, C. (1975). *J. Dairy Sci.* **58**:93.
8. Ibeawuchi, J.A. (1984). *Bull. Anim. Hlth Prod. Afr.* **32**:123.
9. Isoun, T.T., Akpokodge, T.U., Ikede, B.O. and Fayemi, O. (1974). *Bull. Epiz. Dis. Afri.* **22**:331.
10. Kabuga, J.D. and Alhassa, W.S. (1981). *Wld. Rev. Anim. Prod.* **17**:41.
11. Kabuga, J.D. and Agyemang, K. (1984). *Trop. Anim. Hlth and Prod.* **16**:85.
12. Lloyd, D.H. (1986). Dermatophilosis. In: *Infectious Tropical Diseases of domestic animals* (Ed. G.J. Losos), Essex, England. Longman Scientific and Technical, p. 694.
13. Maina, J.A. (1986). Animal Health in Subhumid Nigeria. In: *Livestock Systems Research in Nigeria's subhumid zone* (R.V. Kaufman, S. Chater and R. Blench). Addis Ababa, Ethiopia, ILCA p. 167.
14. Mathews, C.A. and Forhrman, M.H. (1954). *Beltsville growth Standard for Holstein Cattle.* Tech. Bull No. 1099. Washington, D.C. U.S. Dept. Agric.
15. McIntyre, K.H. (1971). *Trop. Agric. Trin.* **48**:317.
16. Oppong, E.N.W. (1973). *Ghana J. Sci.* **13**:44.
17. Trail, J.C.M. and Marples, H.J.S. (1968). *Trop. Agric. Trin.* **45**:173
18. Vaccaro, de. L.P. (1974). *Anim. Breeding Abstr.* **42**:93.
19. Vaccaro, R. and Rivero, S. (1985). *Anim. Prod.* **40** :279.
20. Walker, J. (1967). *Ghana Livestock Development Handbook.* Accra. U.S. Aid Mission to Ghana, p. 9
21. Yeboah, M.A. (1977). *Proceedings of the Ninth Animal Science Symposium* held at U.S.T., Kumasi. Ghana Animal Science Association, p. 31.

Received for publication on 26th May 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 97-99

INFLUENCE OF INTERMITTENT WATERING ON MILK YIELD AND MILK COMPOSITION OF YANKASA EWES

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EFFETS DE L'ABREUVEMENT INTERMITTENT SUR LA PRODUCTION LAITIERE ET LA COMPOSITION DU LAIT DES BREBIS YANKASA

Résumé

La présente étude a été entreprise en vue de connaître les effets de l'abreuvement intermittent sur le paramètre de lactation (production laitière et composition du lait) chez les brebis Yankasa. 15 brebis allaitantes ont été soumises à trois types d'abreuvement: (1) abreuvement quotidien, (2) abreuvement tous les deux jours (48 heures), (3) abreuvement tous les trois jours (72 heures). L'expérience a duré 50 jours. La production laitière moyenne/jour des brebis était respectivement de 271,6 ml; 175,6 ml et 121,2 ml avec les abreuvements à intervalles de 24, 48 et 72 heures.

Les différences relatives à la production laitière des brebis selon les divers intervalles d'abreuvement étaient très significatives ($P<0,05$). De même, une augmentation très importante ($P<0,01$) en rapport avec la composition du lait (solides totaux, solides non gras, matière grasse, protéine et cendres) a été notée avec l'extension des intervalles de restriction d'eau. Selon les résultats de la présente étude, la baisse de la production laitière a un rapport avec la restriction d'eau chez les brebis allaitantes.

Summary

This study was conducted to find out the effect of intermittent watering on the lactation parameter (milk yield and composition) in Yankasa ewes. Fifteen (15) lactating ewes were subjected to 3 watering treatments: (1) daily watering, (2) once in 2 days (48 hours) and (3) once in 3 days (72 hours). The study lasted for 50 days. The average daily milk yield for the ewes were 271.6ml, 175.6ml and 121.2ml for 24, 48 and 72 hourly watering treatments respectively.

The differences in milk yield of the ewes on the different watering intervals were highly significant ($P<0.05$). Similarly, highly significant ($P<0.01$) increases in milk content of total solids, solids-not-fat, fat, protein and ash were obtained with increase in water restriction intervals. Results of this study indicated that milk yield decreased with a restriction of water intake to the lactating ewes.

INTRODUCTION

Water is an essential constituent of the living cell. It is a prerequisite for animal's maintenance of life. It is intimately concerned with the transportation of nutrients around the body and the excretion of materials from the cells of the different body tissues[1], reported that water is uniquely suited to become the inorganic basis of living matter. It's unusually high dielectric constant helps to bring ions into solution in range of temperatures in which water is liquid[2], reported the average composition of ewe's milk as follows: Total solids (18.4%), Protein (5.6%), Fat (7.5%) and Ash (0.87%).

The Yankasa sheep is one of the most predominant breeds of sheep in Nigeria, is found mainly in the Northern parts of Nigeria where the dry season can be as long as 8 months in a year. During the dry season, there is always a shortage of water in the region for these animals. This study was carried out to investigate what happens to the milk yield and composition of Yankasa ewes during the long dry season, when there is scarcity of drinking water.

Materials and Methods

Fifteen lactating Yankasa ewes were used for this study which lasted 50 days.

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All the ewes were nursing 2-weeks old lambs. The ewes and their lambs were fed and watered in individual pens. The ewes were given hay *(Andropogon gayanus)* and an 18% crude protein concentrate mixture consisting of maize, cottonseed cake, vitamin-mineral premix, salt and bonemeal (Tables 1 & 2). The ewes were allotted to 3 treatments.

treatment 1 — 24 hourly watering
treatment 2 — 48 hourly watering
treatment 3 — 72 hourly watering

The animals were watered using clean metal pails. The ewes were fed concentrate and roughage in the ratio of 40:60. The ewes were hand milked at fortnight intervals to determine average milk yield per day. Samples were taken from the ewes' milk for laboratory analyses. Lambs were withdrawn from the ewes a night previous to the milking day. The ewes were milked in the morning at about 8.00 a.m. After the milking, lambs were returned to their dams. Milk samples were analysed in the laboratory using the[3] method of analysis. Data was subjected to statistical analysis by the procedures of[4].

Table 1: Concentrate Composition

Ingredient	Composition (%)
Maize	60.16
Cottonseed cake	36.09
Salt	2.0
Bone meal	1.5
+Vitamin-mineral Premix	0.25

+Vitamin-mineral Premix used is ZOODRY VM 702 and it contains per kg diet, vit. A 25,000 I.U., vit. D_3 27,500 I.U., vit. E 25mg, Magnesium (mg) 250 mg, Manganese (Mn), 125mg, Iron (Fe) 200mg, Zinc (Zn) 112.5mg, Copper (Cu) 15mg, Iodine (I) 6.5mg, Cobalt (Co) 2mg.

Table 2: Composition of concentrate and hay (DM basis)

	Hay	Concentrate
Crude protein (CP)	4.10	18.0
Ash	9.0	5.8
Ether extract (E.E.)	2.0	4.98
Crude fibre (CF)	39.1	5.9
Nitrogen free extract (NFE)	37.8	59.32

Results

The results of this study indicated that water restriction affects both milk yield and composition as evident in Table 3. Animals on daily watering produced most milk. Milk production decreased with increased watering intervals ($P<0.01$). Milk production of the ewes dropped by more than 50% when watering intervals were up to 72 hours, compared with daily watering. The milk produced by ewes on the longer watering intervals was more viscous.

Consequently, the composition of the ewes' milk that is the protein, fat, ash, solid not fat (SNF) and total solids (g/100g) increased with increased water restriction.

There was a significant ($P<0.01$) decrease in milk yield with increased water restriction. These results show that the deprivation of water to lactating ewes

Table 3: Milk yield and composition

Treatments	1	2	3	SE+
Watering Intervals	24h	48h	72h	
No. of ewes	5	5	5	
Average milk yield/head/day (ml)	271.6^{c}	175.6^{c}	121.2^{a}	4.36
Total solids, %	14.97^{c}	16.51^{b}	19.21^{a}	0.51
Solid not fat (SNF), %	10.95^{c}	11.73^{b}	13.49^{a}	0.56
Fat, %	4.03^{c}	4.84^{c}	5.72^{a}	0.17
Ash, %	0.71^{b}	0.79^{b}	0.90^{a}	0.02
Crude protein (CP), %	6.19^{c}	6.67^{b}	7.02^{a}	0.84
Lactose	4.40^{a}	4.28^{ab}	4.05^{b}	0.45

+Standard error
^{abc}Means within the same variable bearing different superscripts differ ($P<0.01$).

for more than 24 hours causes marked decline in their milk yield.

Total solids of milk samples were 14.97±0.73, 16.51±0.36 and 19.21±0.89 for treatments 1, 2 and 3 respectively. These were significantly different from each other ($P<0.01$). Similarly, significant differences existed ($P<0.01$) between treatments for solid not fat, fat content, ash and crude protein.

Discussion

Milk yield of the ewes decreased with increased water deprivation[5], reported that lactating and non-lactating camels in the desert during summer differed by 27ml/kg body weight/day (44%) in water turnover rate because of the water used in milk production. Lactation is energetically expensive to a mammal but it is also a costly function in terms of water utilization[5], also reported that lactating Merino ewes produced about 1.2 litres of milk daily to feed their lambs[6], observed that milk yield decreased significantly with increasing water deprivation in lactating ewes.

For optimum milk production, water should be given to lactating ewes daily (and preferably *ad libitum* where possible) to enhance milk production.

References

1. A.O.A.C. (1970). *Methods of analysis.* 11th edition, Association of Official Agricultural Chemists. Washington, D.C.
2. Kon, S.K. and Cowie, A.T. (1961). *Milk:The Mammary Gland and its secretion.* Vol. II. London Academic Press.
3. Little, W., Sansom, B.F., Manston, R. and Allen, W.M. (1976). *Animal Prod.*, **22**, 329.
4. Macfarlane, W.V. and Howard, B. (1972). *Symp. Zool. Soc. Lond.* No. 31, 261.
5. Ronbinson, J.R. (1957). *Proc. Nutr. Soc.* **16**, 108.
6. Steel, R.G.D. and Torrie, J.H. (1960). *Principles and Procedures of statistics.* McGraw-Hill Book Co. Inc. New York.

Received for publication on 22nd March 1988

Bull. Anim. Hlth. Prod. Afr. (1990), 38, 101-102

SHORT COMMUNICATION

PREVALENCE OF *SALMONELLA* IN HEALTHY CALVES FOLLOWING TRANSPORTATION TO THE STOCKYARDS AND AT SLAUGHTER

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The prevalence of Salmonella in calves, as an indicator of the source of Salmonellosis at the production level of human food of bovine origin and the importance of calves as sources of *Salmonella* infections to other livestock and the environment following transportation, was considered. A 1% (2/200) prevalence of *S. typhimurium var copenhagen* from the calf ileo-caecal mesenteric lymph nodes was recorded. While nearly the same percentage (1%, 13/1400) of faecal samples examined, were positive for *Salmonella* involving six serotypes. The transportation and the calf-trade industry involving movement from the farmer to sales barns, the stockyards and the veal slaughter plant, did not increase the prevalence of *Salmonella* recovered from these calves, compared to observations elsewhere.

Salmonellosis has continued to be identified as the most widespread zoonotic infection in the United States of America, according to USDA 1978 report[11]; and animal products are the major sources and vehicles of Salmonellosis[2,8]. Surveillance of Salmonellosis in calves after travelling and following collection together, therefore, is an important indicator of the prevalence of this infection, since calves are more susceptible to Salmonellosis and are better amplifiers and shedders of *Salmonella*[7]. Delayed transportation has been observed to enhance the prevalence of the disease and infection in Great Britain[5,6], Australia[9] and New Zealand[7].

These studies were made using mesenteric lymph nodes of 200 calves sampled over a period of six months at a veal slaughter plant in the State of Minnesota, USA, and faecal material from 1400 calves at the stockyards in St. Paul, Minnesota. The transportation and movement of these calves was traced from their farms of origin, through sales barns to the stockyards and the veal abattoir. In all cases the calves arrived at the stockyards from all-over the State and the Western part of the State of Wisconsin within six hours, whereas those which were taken for slaughter were delivered to the abattoir within three hours. Caecal and faecal contents as well as mesenteric ileo-caecal lymph nodes were subject to routine microbiological analysis involving culture for recovery, biochemical and serological tests[1a,b,c;9].

Sample and amount	Positive	Percent	*Salmonella* serotype
A. Mesenteric lymph nodes (200)	2	1%	*S. typhimurium var Copenhagen*
B. Faecal contents (1400)	13	<1%	*S. london* (5); *S. typhimurium var Copenhagen* (3); *S. lictifield* (2); *S. heidelberg* (1); *S. bietri* (1) and *S. newport* (1)
C. Caecal contents (200)	0	0%	–

The recovery rate of 1% of *Salmonella* in mesenteric lymph nodes supports the view by Fields in 1948 as quoted by Gibson in 1965[2] and supported by others[3,6,7]

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that, Salmonellosis in calves older than 6 weeks of age, tends to persist resulting in a carrier state and unthriftiness. It has indirectly supported the view that, those calves younger than one month of age are better sources of Salmonella to the environment. *S. typhimurium var Copenhagen* has been reported[10] as the most prevalent serotype in USA. Yet the low (1%) recovery rate may be due to lack multiple samples taken from the same calves[5].

The following conclusions were drawn from this study:-

(a) Shedding and infection rates in calves of the age groups used for veal in the State (most were above one month of age) were very low.

(b) There was no difference in *Salmonella* recovery among calves which were transported from farms to the veal slaughter plant and those which were first assembled at the stockyards prior to slaughter. The quick transport gave little contact time for stress, transmission and infection to take place, especially since the calves were slaughtered soon after arrival.

(c) Veal is not a significant source of *Salmonella* in the food of bovine origin in Minnesota compared to New Zealand where large numbers of calves are slaughtered for veal and severe post-slaughter contamination is recorded[4,7].

(d) The veal slaughter plant and the stockyards did not represent the ideal points of surveillance for Salmonellosis in calves in Minnesota. Surveillance at the animal production level using calves did not show a high recovery of *Salmonella,* yet it did not rule out the fact that somewhere along the line of calf-production and trade, there may still be a high prevalence of Salmonellosis in the region of study. Hence further surveillance at different points of calf-production and trade and especially among calves less than one month of age needs to be done.

Acknowledgements

This project was sponsored by the USDA under research grant on infectious diseases, on which Professors Robinson and Pullen were major investigators in 1979/80.

References

1. *Difco Technical Information, Detroit, Michigan* (1975, 1977):
   (a) Salmonella media/enrichment (1977): 0211.
   (b) Antigenic schema for Salmonella (Kaufman-White Schema Modified) 1975:**0357**:1
   (c) Serological identification of Salmonella (1977):**0168**:1
2. Gibson, E.A. (1965). J. Dairy Science: **32**:97.
3. Gitter, M., Wray, C., Richardson, C and Pepper, R.T. (1978). *Brit. Vet. J.*; **134**:113.
4. Kallings, L.O., Laurell, A.B. and Zetterberg, B. (1959). *Acta Path. et Micro. Scand.* **45**:347.
5. Lawson, G.H.K., McPherson, E.A. and Wooding, P. (1974). *J. Hyg. Cambridge:* **72**:311.
6. Osborne, A.D., Linton, A.H. and Pethiyagoda, S. (1974). *The Vet. Record:* **94**:646.
7. Robinson, R.A. (1966). *Proceedings New Zealand Society of Animal Production;* **26**:134.
8. Robinson, R.A. and Loken, K.I. (1968). *J. Hyg. Cambridge:* **66**:207.
9. Samuel, J.L., O'Boyle, D.A., Mathers, W.J. and Frost, A.J. (1979). *Research In Veterinary Science:* **28**:238.
10. Smith, P.B. (1980). *California Veterinarian:* **4**:27.
11. USDA (1978). Proceedings of the National Salmonellosis Seminar, January 10th - 11th; USDA, 14th Independence Ave., S.W. Washington, D.C. 20250.

Received for publication on 11th February 1986

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BULLETIN OF ANIMAL HEALTH AND PRODUCTION IN AFRICA

RECOMMANDATIONS AUX AUTEURS

Objet

Le Bulletin de la Santé et de la Production animales en Afrique contient des articles de recherches originales traitant d'activités en matière de santé et de production animales visant à assurer le développement de l'industrie animale et une meilleure utilisation des ressources du bétail en Afrique. Le Bulletin est un périodique trimestriel.

Présentation des articles

Deux exemplaires des articles doivent être addressés à Monsieur le Rédacteur en Chef, Bulletin de la Santé et de la Production Animales en Afrique, Organisation de l'Unité Africaine/Bureau interafricain des Ressources animales, P.O. Box 30786, Nairobi, Kenya.

Les manuscrits doivent être rédigés en anglais ou en français clair et concis, et dactylographiés en double interligne avec de larges marges. L'orthographe doit correspondre à celle du dictionnaire *The Short Oxford English Dictionary* ou *Le Petit Robert.*

Un article ne peut être soumis pour publication que s'il n'a pas encore été proposé ailleurs; il fera l'objet de quelques modifications par le Comité de Rédaction.

Genres d'articles publiés dans le Bulletin

— des communications originales
— des brèves communications
— analyse des articles proposée par le Rédacteur
— des éditoriaux
— le courrier des lecteurs
— analyse d'ouvrages
— informations et annonces

Format des articles

Les manuscrits doivent respecter les conditions suivantes:

Le titre doit être concis et ne pas dépasser plus de 15 mots, il est suivi du (des) nom(s) de l'auteur (ou des auteurs) et des établissements où le travail a été effectué, ainsi que de l'adresse pour les correspondances si elle n'est pas la même.

Le résumé ne doit pas excéder 200 mots. Son texte bref et concis comprendra les principaux résultats et la (les) conclusion(s) de l'étude.

L'introduction expose le but de la recherche.

Le matériel et les méthodes utilisés.

Les résultats présentés brièvement.

Un débat sur l'importance de l'article.

Remerciements éventuels.

Bibliographie: les références bibliographiques doivent être numérotées dans l'ordre, telles qu'elles apparaissent dans le texte. L'identification des références dans le texte se fera à l'aide de numéros (entre parenthèses) et non pas par les noms des auteurs.
La bibliographie doit respecter la présentation suivante:

1. *Journal*
Le nom de l'auteur (ou des auteurs) suivi des initiales du ou des prénoms, l'année de parution (entre parenthèses), l'abréviation du titre du périodique suivant la *World List of Scientific Periodicals* (soulignée), le numéro de la première page. Le titre de l'article ne doit pas être inclus.

2. *Revue*
Le nom de l'auteur (ou des auteurs) suivi des initiales du ou des prénoms, l'année de parution (entre parenthèses), le titre exact (souligné), la ville où elle a été publiée, les éditeurs, le numéro de la première page.

3. *Rapport annuel*
Le nom du pays, l'année faisant l'objet du rapport, puis le nom du service ou de l'organisation, le numéro de la première page.

Si le même auteur est cité plus d'une fois, ses publications seront indiquées dans l'ordre chronologique dans la liste bibliographique et s'il y a plus d'une publication, les lettres "a, b, c," seront ajoutées aussi bien dans la liste bibliographique que dans le texte.

Illustrations
Les tableaux et les titres doivent être en nombre aussi réduit que possible. Un tableau d'une trop grande dimension est difficile à lire même s'il peut être reproduit. Les tableaux et les figures doivent être numérotés dans l'ordre, respectivement Tableau 1, etc., ou Fig. 1 etc. et joints à la fin du texte. Les références aux tableaux et aux figures dans le texte doivent être numérotées et non pas indiquées "tableau ci-dessous" ou "figure ci-dessous". Les illustrations en couleurs ne sont reproduites qu'aux frais de l'auteur (ou des auteurs).

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Une brève communication signifie que l'article ne peut pas être publié comme une communication normale. Elle ne doit pas dépasser deux pages imprimées ou 1.000 mots en incluant deux illustrations au maximum. Elle doit donc respecter les mêmes normes qu'un article habituel, sauf que le résumé et les sous-titres ne sont pas nécessaires.

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Les épreuves typographiques sont envoyées à l'auteur qui en effectue la correction des coquilles et en assure le retour rapide (dans les 3 jours).

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Photo-typeset and Printed by
Modern Lithographic (K) Limited
P.O. Box 52810, Tel: 21285/28075, Nairobi

1990

Animal Health and Production in Africa

Inter African Bureau for Animal Resources

Organization of African Unity