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GUIDANCE FOR AUTHORS

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The Bulletin of Animal Health and Production in Africa publishes articles on original research relevant to animal health and production activities which may lead to the improvement of the livestock industry in Africa and better utilisation of her animal resources. The journal is published quarterly.

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Concurrent Outbreak of Avitaminosis A and Coccioidosis in a Poultry Flock

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Eclosion Simultanée d'Avitaminose A et de Coccioidose dans un Poulailier

Résumé

Une éclosion simultanée d'avitaminose A et de coccioidose dans un poulailier de 75 oiseaux âgés de 17 semaines (45 poulettes et 30 poulets de gril) à Nsukka est décrite.

L'éclosion était caractérisée par un amaigrissement considérable et une forte baisse de la croissance, surtout prononcés chez les poulets de gril. Les autres symptômes étaient l'agglutination des paupières, le hérissement des plumes et l'ataxie. La diarrhée qui était surtout sanglante était évidente.

L'histopathologie et les examens microscopiques ont permis de découvrir des ovocytes d'Eimeria et des schizontes dans les intestins, la nécrose des cellules épithéliales, des tubules et des canaux du rein. Des sections de l'oesophage et de la base de la langue présentaient une grave kératinisation et une dilatation des glandes, tandis que les examens biochimiques faisaient apparaître un taux élevé d'acide urique dans le sang.

La pathologie, la morbidité et la mortalité étaient plus élevées chez les poulets de gril. Vingt-quatre (80%) et six (13,3%) poulets de gril et poulettes manifestaient respectivement des signes cliniques, tandis que huit (26,7%) et deux (4,4%) sont morts chez les deux catégories.

La mauvaise alimentation des oiseaux et l'absence d'une médication prophylactique appropriée, par exemple des produits anticoccidiens, en plus de l'utilisation d'une litière épaisse semblent avoir provoqué l'apparition de ces deux maladies, qui réagissaient positivement au traitement à la duocoxine (R) et aux sevenseas (R) (huile de foie de morue) en deux ou trois semaines.

Summary

A concurrent outbreak of avitaminosis A and coccidioidosis in a flock of 75, 17-week old birds (45 pullets and 30 broilers) at Nsukka is reported.

The outbreak was characterised by severe weight loss and marked decline in growth which were more pronounced in the broilers. Other signs were the gluing of the eyelids, ruffling of the feathers and ataxia. Diarrhoea which was mostly bloody was evident.

Histopathology and microscopic studies revealed numerous Eimeria oocysts and schizonts in the intestine, necrosis of the epithelial cells, tubules and ducts of the kidney. Sections from the oesophagus and base of the tongue showed severe keratinisation and gland dilatation while biochemical studies showed an elevated blood uric acid level.

Pathology, morbidity and mortality were highest in the broilers. Twenty-four (80%) and six (13.3%) of the broilers and pullets respectively showed clinical signs, while eight (26.7%) and two (4.4%) of these same respectively died.

Feeding of birds with the wrong or badly composed feed, coupled with the absence of appropriate prophylactic medication, e.g. anticoccidial, in addition to the use of a built-up deep litter could be said to have provoked the outbreak of the disease complex which responded favourably to Duocoxin® and Sevenseas® (cod liver oil) medication within a period of 2-3 weeks.

Introduction

Coccidioidosis in addition to being a very important disease of poultry as it causes a lot of death (1) and drop in production (2) has been demonstrated to impair not only the absorption of vitamin A (3,4), but also the conversion of carotene to vitamin A (5). The overall effect of all these is the reduction in the amount of vitamin A
storage by the liver (6,7) which often manifests in a clinical disease. Vitamin A is essential for the health and integrity of epithelial tissues. Its deficiency results in the keratinisation of tissues (8).

Although some workers have demonstrated the combined effect of experimental vitamin A deficiency and coccidiosis in chickens (5,6,9) a combined natural field outbreak of the two diseases, especially in a flock of broilers and pullets, does not appear to have been studied.

The aim of this paper, therefore, is to describe the clinicopathological manifestations and therapeutic management or control of a concurrent natural outbreak of the two diseases in a flock of broilers and pullets in a poultry farm.

Case History

Fifty pullet chicks and 120 broiler chicks were reared on deep litter in different pens from day-old and fed with chick mash and broiler starter mash respectively. The feed was changed to grower's mash at the eighth week of life in the former, and broiler finisher mash at the seventh week of life in the latter.

At the twelfth week the remaining broilers after sales, i.e. 30 birds, were put into the pen that housed the 45 remaining pullets and fed irregularly with grower's mash as the cost of broiler feed was prohibitive.

These birds were duly and properly immunised against important prevalent poultry diseases, viz: Newcastle disease, gumboro disease, fowlpox and fowl typhoid.

A general loss of weight, weakness, gluing together of the eyelids and the passage of bloody diarrhoea heralded the outbreak of the disease complex. Eight broilers and two pullets died while two birds from each group were culled after bleeding.

Clinical Observations

Broiler Flock

Loss of weight, severe emaciation, stunted growth, gluing of the eyelids by sticky purulent ocular discharges and at times the ulceration of the eyelids (Fig. 1) and weakness were the most outstanding clinical observations. Anorexia, slight watery nasal discharges, ruffling of the feathers and droopiness (Fig. 1) were the other accompanying signs. Nervous signs which took the form of ataxia were observed in two (8%) of the affected birds while diarrhoea which was mostly bloody was evident. Small roundish swellings were also observed very close to the inner angle of the nostril in some of the birds (Fig. 1). Emaciation and gluing of the lids which were the first signs to appear were observed after about 3-4 weeks of the

Figure 1: A. Purulent ocular discharge; B. Small roundish swelling; C. Ulcerated eye lid.
irregular feeding.

**Pullet Flock**

Loss of weight, emaciation, anorexia and weakness were evident but not as severe as in the broilers. Stunted growth was not pronounced while ataxia was observed in only one of the six birds that showed clinical signs.

Ocular discharges were mostly watery in nature with the gluing of the eyelids seen in only one bird. Nasal discharges and the small roundish swelling near the nostril were not evident. Diarrhoea which was mostly bloody was observed.

**Post-mortem Findings**

**Broiler Flock**

At necropsy, very thick dirty-white caseous deposits were found on the tongue and oral cavity of the severely affected birds (Fig. 2). These lesions were easily removed with no bleeding.

The pharynx and oesophagus were liberally covered with small nodules (Fig. 2) while the kidney which appeared swollen had a degree of whitish discolouration.

The intestines, especially the duodenum and mid-gut, were quite haemorrhagic and ballooned. These yielded numerous *Eimeria* oocysts on microscopic examination using the method described by (10). An ammonia-like odour was noticed coming from some of the necropsied carcasses.

**Pullet Flock**

Dead birds showed very faint pharyngo-oesophageal lesions as described for the broilers. The tongue and oral cavity lesions were also present but mild. There was a complete absence of the dirty-white caseous deposits in place of which a lot of ulcers were seen, especially on the palate.

The kidneys appeared normal while the ballooned and haemorrhagic intestines (duodenum and mid-gut) yielded a lot of *Eimeria* oocysts on microscopic examination. The ammonia-like odour was inconspicuous.

---

**Figure 2:** Caseous dirty-white deposits on the tongue and oral cavity. Pharynx and oesophagus showing nodular lesions:

T = tongue, E = oesophagus, t = trachea, C = caseous dirty-white deposits in the oral cavity.

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**Determination of Blood Uric Acid Levels**

About 2 ml of serum was collected after bleeding each of the four birds (two pullets and two broilers) that showed clinical signs. These were used for the determination of the blood uric acid levels using the method described by (11).

The mean blood uric acid levels were 21 and 5.9 mg/100 ml for the broilers and pullets respectively.
Discussion

The diagnosis of the combined vitamin A deficiency and coccidiosis in this report was based on the history, clinical signs, post-mortem, histopathologic and microscopic findings in addition to blood biochemical analysis and response to treatment which was evidenced by the remission of clinical signs following treatment.

The characteristic pharyngo-oesophageal pustule-like lesions of vitamin A deficiency (12), which were not observed in the dead birds in this outbreak, may be as a result of early death due to the combined effect of the two diseases. This observation is in line with those of (13), (14) and (6) who showed that chickens suffering from vitamin A deficiency stood a higher risk of dying of coccidiosis. Hence the observation of only nodular lesions in the oesophagus and pharynx (Fig. 2) even in those birds that died of the disease.

It is also an observation worthy of mention that in all the affected birds, gastrointestinal lesions due to vitamin A deficiency were severest in the tongue and oral cavity and decreased in severity towards the crop.

The rapid growth rate and weight gains in broilers which require more nutrients, e.g. vitamin A which (15) placed at 1,100 i.u./kg of feed as against 6,500 i.u./kg of feed for the growers, may explain why the broilers were more severely affected by the outbreak than the pullets.

Poor management, like feeding with low quality or the wrong feed, e.g. grower's mash to broilers, and the rearing of chickens on a built-up deep litter with no history of anti-coccidial medication which can be said to have led to the outbreak on this farm, is a practice that should be discouraged amongst farmers.

The mean blood uric acid level of 21 and 5.9mg/100 ml for the broilers and pullets, respectively, indicates a substantial increase when compared with the standard value of 2 mg/100 ml for non-laying birds given by (16).

This increase agrees with the findings of (17) who observed a significant rise in the blood uric acid level in chickens.
suffering from confirmed cases of avitaminosis A.

The apparently lower rise in the blood uric acid level observed in birds at the height of ill-health in this case as compared to those of (17) could be due to the combined effect of coccidiosis and avitaminosis A which weighed down seriously on the birds, killing some of them, even before each of the conditions could develop fully.

The disparity in the blood uric acid levels of the broilers and pullets show further that the broilers were more severely affected, and this may possibly throw more light on why more damage was observed both grossly and histopathologically in the broilers than the pullets. This observation agrees with that of (12) who observed a greater number and more severe lesions in birds suffering from clinical avitaminosis A than those just suffering from the effects of sub-optimal levels of vitamin A in their mash.

The principal clinical signs of emaciation, stunted growth, eye lesions, muscular incoordination, etc., observed in this case agrees with the observations of (18) in birds suffering from confirmed cases of vitamin A deficiency. The presence of the small roundish swellings near the nostril (Fig. 1), an indication of sinusitis, has also been recorded (12). The ammonia-like odour coming from some of the necropsied carcasses may be associated to high uric acid levels in the blood.

The complete disappearance of the signs of the disease especially those of avitaminosis A and the cessation of mortalities following treatment shows that treatment with cod liver oil (Sevenseas® and Duocoxin®) is effective. Cod liver oil, in addition, may have also hastened the remission of the signs of coccidiosis in line with the findings of (14) that chickens suffering from a natural outbreak of coccidiosis if placed on cod liver oil alongside whatever other treatment, stood a higher chance of surviving than those not receiving cod liver oil.

Cod liver oil (Sevenseas®), therefore, can serve as a ready substitute of stabilized vitamin A for the treatment of avitaminosis A (12), a drug that may be difficult to find in most local veterinary drug houses.

Further work is therefore required in determining what amount of Sevenseas® will serve as a prophylactic or therapeutic dose for the management of avitaminosis A or in combination with other diseases like coccidiosis, since the dose used in this outbreak was arbitrarily chosen. This, when done, will not only solve the attendant problem of insolubility in cold water, as the required dose could be given per os to chickens, but will in addition reduce drug wastage and cost of treatment.

Acknowledgement

I am grateful to Dr J.O.A. Okoye of the Veterinary Pathology Department, University of Nigeria, Nsukka, for making necessary corrections and suggestions on this paper.

References

Isolation of Bovine Herpesvirus I (Infectious Bovine Rhinotracheitis Virus) and Observations on Primary Recurrent and Contact Infections in Cattle in Kenya

J.S. WAFULA, E.Z. MUSHI and R.G. IRERI

Kenya Agricultural Research Institute, Veterinary Research Department, Muguga, P.O. Box 32, Kikuyu, Kenya

Isolation du Virus Herpétique I Bovin (Virus de la Rhinotrachéite Infectieuse Bovine) et Observations sur les Infections Primaire et Recurrentes et Contagion Chez les Bovins au Kenya

Résumé

Le virus herpétique I bovin a été isolé du tractus respiratoire d'un bouvillon cliniquement normal. L'inoculation expérimentale de huit veaux avec le virus par voies intranasale, intravaginale, intraoculaire ou intraveineuse a donné des syndromes cliniques différents. Une maladie respiratoire légère s'est développée chez six des huit bovins infectés, tandis que des symptômes vaginaux et oculaires graves n'ont affecté que ceux infectés respectivement par voies génitale et oculaire. Le virus a été récupéré chez sept des huit animaux, dans les sécrétions vaginales, oculaires et nasales 1-11 jours après l'infection.

Le traitement au dexaméthasone des veaux inoculés par voies intranasale et intravaginale, 4 mois plus tard, a provoqué une rechute du virus. Les changements cliniques et l'élimination du virus dans l'infection récurrente étaient limités aux organes qui ont fait l'objet de la première inoculation du virus. Les veaux gardés en contact avec ceux traités au dexaméthasone ont réagi avec une légère maladie respiratoire.

Summary

Bovine herpesvirus I was isolated from the respiratory tract of a clinically normal steer. Experimental inoculation of eight calves with the virus either by the intranasal, intravaginal, intraocular or intravenous routes resulted in varying clinical syndromes. A mild respiratory disease developed in six of the eight infected cattle while severe vaginal and ocular signs occurred only in those animals infected by the genital and ocular routes respectively. Virus was recovered from seven of the eight animals either from vaginal, ocular or nasal secretions 1-11 days after infection.

Dexamethasone treatment of intranasally and intravaginally inoculated calves 4 months later resulted in virus recrudescence. Clinical changes and virus shedding in the recurrent infection were confined to the organs of primary virus inoculation. Calves kept in contact with dexamethasone-treated cattle reacted with mild respiratory disease.

Introduction

Bovine herpesvirus I (BHV I) has been implicated in many clinical syndromes in addition to the typical infectious bovine rhinotracheitis (1,2), pustular vulvo-vaginitis (3), conjunctivitis (4), abortion (5) and meningoencephalitis (6). The virus is readily transmitted and has a worldwide distribution (2). In Africa, the disease has been reported in cattle in South Africa (7), Central Africa (8), East Africa (9,10), and also in West Africa (11). Virus isolations have been made from cattle in Tanzania (10), Nigeria (11), Zimbabwe (12) and Kenya (9), and serological surveys of cattle indicate widespread infection (12,13). Serum surveys have also shown occurrence of antibodies to BHV I in a number of wildlife species in East Africa (14,15) and in a number of countries in Southern Africa (16,17). (18) and (19) reported the isolation of BHV I from vaginal swabs and occurrence of the genital form of the disease in wildebeest following the administration of a corticosteroid. This paper reports the isolation of BHV I from
the respiratory tract of a clinically normal steer and the results of subsequent experimental infection of cattle.

Materials and Methods

Original Sampling
During routine potency and efficacy testing of rinderpest vaccine at Muguga, nasal swabs were taken from six clinically normal steers 3 days prior to challenge with virulent rinderpest virus. The swabs were treated for virus isolation as described below.

Cell Cultures
Primary confluent monolayer tube cultures of bovine kidney (BK) cells were prepared as described by (20), using Minimum Essential Medium* supplemented with 10% BHV I antibody-free ox serum.

Experimental Animals
Five steers and three heifers aged between 1 and 2 years were obtained and shown to be devoid of BHV I neutralising antibody at 3 weeks and again immediately before inoculation of virus. The cattle were placed in isolation quarters for at least 1 week before inoculation.

The animals were inoculated with second cell culture passage virus isolate containing $10^7.2$ TCID$_{50}$ of virus per ml. Two heifers received 5 ml of virus each given by intravaginal spray via an atomizer.

Two steers were each sprayed intranasally with 5 ml of virus, given via an atomizer. A third group of two steers was inoculated intravenously with 5 ml of virus per animal. One steer and one heifer received 2 ml of virus by intraocular instillation.

Dexamethasone Treatment
Four months after virus inoculation, the intranasally and intravaginally inoculated cattle were given a series of five daily intramuscular injections of 0.2 mg dexamethasone per kg bodyweight. Two heifers seronegative to BHV I were housed together with the intravaginally inoculated heifers while a heifer and a steer also devoid of antibodies to BHV I were housed with the intranasally inoculated steers. The animals were kept in contact from the first day of treatment.

Sampling and Virus Isolation
Orial, nasal, conjunctival and vaginal tract secretions were collected by placing a sterile cotton wool swab into the organ until it became wet. Swabs were collected into 3 ml of phosphate buffered saline containing 0.1% bovine albumin (BAPBS) and supplemented with 400 i.u./ml penicillin, 400mg/ml streptomycin and 100 units mycostatin. These specimens together with buffy coat cells were collected before inoculation and then daily for 14 days post inoculation (p.i.). The samples were either stored at -70°C for 1 week or inoculated immediately into confluent primary BK cultures. The cultures were observed for development of cytopathic effects (c.p.e.) for 12 days.

Virus-Serum Neutralisation
Sera collected daily up to day 21 p.i. were tested for BHV I neutralising antibody to the virus isolate. Aliquots of diluted sera were heat inactivated at 56°C for 30 min, then mixed with an equal volume of virus at a dilution calculated to contain $10^{2.0}$TCID$_{50}$ per 0.1 ml. The mixture was subsequently incubated at +4°C for 18-24 hours then inoculated into three BK tubes. Inoculated cultures were examined for 6 days for development of c.p.e.

Results

Isolation and Identification of BHV I
Within 24 hours of culture inoculation with nasal swab extracts from rinderpest vaccine testing animals, a cytopathic virus with the following features was isolated from one of the six steers. The agent induced cell rounding and small refractile syncytia. The c.p.e. advanced progressively to cover the entire cell sheet in about 4 days. Infected coverslips stained with haematoxylin and eosin revealed small syncytia containing three to six nuclei and intranuclear inclusion bodies of Cowdry Type A. The agent was completely neutralised with antisera raised in rabbits against the Oxford strain* of BHV I and was inactivated by ether and chloroform.

*Originally obtained from Dr J.H. Darbyshire, C.V.L. Weybridge, Surrey, UK.

†The DeVlbiss Co., Somerset, Pennsylvania, USA.
Clinical Responses

Clinical signs following intranasal inoculation of cattle with the virus isolate were generally mild. Hyperemia of the nasal mucosa and serous nasal discharges were seen from day 2 p.i. These discharges progressively became mucoid, then mucopurulent by day 4 p.i. Coughing and pin-point necrotic plaques on the ventral and lateral surfaces of the nasal epithelium were evident on day 5 p.i. and persisted for 6 days. Fever occurred on day 3 p.i. and persisted for 3 days reaching a peak of 39.7°C on day 4 p.i. One of the two steers developed a mild conjunctivitis with serous ocular discharge which lasted for 3 days. The two animals had completely recovered by day 12 p.i.

Intravenous inoculation of 5ml virus into two steers resulted in a mild febrile reaction in only one animal. Fever of 39.8°C was recorded in this animal only on day 7 p.i. Hyperemia of the nasal mucosa accompanied by slight seromucoid nasal discharges were observed in the same animal from day 4 to 9 of inoculation. Clinical signs were not observed in the other steer.

Clinical changes following intravaginal exposure of animals to the virus isolate occurred from day 2 of infection. Fever was observed from day 2 to 5 p.i. and reached a maximum of 39.9°C on day 3 p.i. White, raised pustules and hyperemia developed on the vaginal mucosa on day 2 of infection. The pustules progressively enlarged and coalesced to form large areas of ulceration by day 5 p.i. By day 3 of inoculation, the animals had begun to show discomfort, straining and stamping of feet. No vaginal discharges were observed throughout the course of this disease. A mild respiratory syndrome manifested by seromucoid nasal discharges, hyperemia of the nasal mucosa and development of discrete whitish nasal plaques occurred in one of the two heifers.

The animals given 2 ml of virus per eye developed a temperature reaction by day 2 of infection. The fever which persisted for 5 days with a peak of 40.4°C on day 3 p.i. was initially accompanied by hyperemia of the ocular membranes, photophobia and serous ocular discharge. Severe conjunctivitis, infected scleral vessels and mucopurulent ocular discharges later developed and persisted up to day 10 of infection. Small discrete, whitish plaques developed on the sclera on day 4 of inoculation but lasted for only 2 days. In addition to the ocular disease, a mild respiratory syndrome also developed in these animals. This was manifested by seromucoid nasal discharges, coughing and occurrence of necrotic plaques on the nasal mucosa. Total recovery was observed in these animals by day 11 p.i.

Clinical Responses After Dexamethasone Injection

Unlike in the initial infection there was no febrile response in intranasally inoculated steers following dexamethasone treatment. A mild respiratory reaction manifested by serous nasal discharges and pustules on the nasal mucosa developed on day 5 after the beginning of dexamethasone injection but had resolved by day 10 of injection. Clinical changes following dexamethasone injection of the intravaginally inoculated heifers were first seen on day 3 as hyperemia of the vaginal mucosa. Pinpoint pustules were observed on the vaginal epithelium from day 6 of dexamethasone injection and persisted for 4 days.

Clinical Response of In-contact Calves

Calves kept in contact with intranasally inoculated steers developed respiratory disease from day 8 of contact. The clinical signs in in-contact calves were similar to the initial clinical signs in the intranasally inoculated steers and lasted till day 14 of contact. No clinical disease was observed in the heifers kept in contact with intravaginally inoculated cattle.

Virus Isolation from Experimental Animals

(a) After Primary Infection. BHV I was recovered from nasal secretions of intranasally inoculated animals for 7 days starting from day 2 p.i. Virus was recovered at a maximum titre of 10^6.0 TCID 50 per swab from one animal and 10^7.8 TCID 50 per swab from another on day 5 p.i. (Fig. 1). Virus was recovered from nasal secretions of one of the intravenously inoculated animals from day 8 to 11 of inoculation. Maximum virus titre of 10^4.2 TCID 50 per swab was observed on day 10 (Fig. 2).
demonstrated in vaginal secretions of the two female animals for 7 days. Peak titres of $10^7.5$ and $10^7.2$ TCID$_{50}$ per swab were attained on day 4 p.i. Low titres of virus were detected in nasal secretions from one of the two animals on days 6 and 7 p.i. The virus was also recovered from conjunctival secretions of the same animal only on day 6 p.i.

The virus was detected both in ocular and nasal secretions from animals infected by the conjunctival route. Virus was demonstrated in ocular secretions from day 1 to 9 p.i. and reached maximum titres of $10^8.8$ and $10^7.8$ TCID$_{50}$ per swab on day 5 p.i. Virus was first recovered from the respiratory tract of these animals on day 3 p.i. and reached a peak of $10^5.5$ and $10^3.5$ TCID$_{50}$ on day 6 p.i. No further virus recovery could be shown in nasal secretions after day 8 of infection (Fig. 4). Neither viraemia nor virus in oral secretions were demonstrated in this study.

(b) After Dexamethasone Treatment. BHV I was isolated only from nasal secretions of intranasally inoculated steers from day 5-15 after the beginning of dexamethasone injection. Virus was recovered from vaginal secretions of the two heifers inoculated intravaginally and subsequently treated with dexamethasone; no virus was however, isolated from the nasal cavities of these animals.

Virus shedding in these animals began on day 5 and continued until day 13 after
the beginning of dexamethasone injection. Maximum virus titres of $10^{6.9}$ TCID$_{50}$ per swab were attained on day 11 following DM injection.

BHV I was recovered from nasal secretions of all the four in-contact calves from day 9 of contact. Virus was not isolated from vaginal samples from calves in contact with intravaginally inoculated heifers.

**Antibody Response**

Neutralising antibodies were not detected during the first 13 days following initial infection. Low levels of neutralising antibodies were first detected on day 14 in intravenously and one intravaginally inoculated animals. All animals had seroconverted by day 21 p.i. (Table 1).

<table>
<thead>
<tr>
<th>Route of inoculation</th>
<th>Animal No.</th>
<th>Days (p.i.)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>7</td>
</tr>
<tr>
<td>Intranasal</td>
<td>177</td>
<td>0.0</td>
</tr>
<tr>
<td></td>
<td>145</td>
<td>0.0</td>
</tr>
<tr>
<td>Intravenous</td>
<td>170</td>
<td>0.0</td>
</tr>
<tr>
<td></td>
<td>131</td>
<td>0.0</td>
</tr>
<tr>
<td>Intravaginal</td>
<td>371</td>
<td>0.0</td>
</tr>
<tr>
<td></td>
<td>327</td>
<td>0.0</td>
</tr>
<tr>
<td>Intraocular</td>
<td>175</td>
<td>0.0</td>
</tr>
<tr>
<td></td>
<td>333</td>
<td>0.0</td>
</tr>
</tbody>
</table>

*Log$_{10}$ VN$_{50}$

**Discussion**

BHV I was isolated from the respiratory tract of a healthy steer which up to the time of healthy recovery had been under close confinement for 3 weeks. It is likely, therefore, that the stress of confinement may have triggered the reactivation of latent BHV I virus in this case. Although clinical signs of disease were not manifested in this steer, experimental infection of eight cattle with the virus isolate resulted in clinical syndromes similar to those previously described for BHV I in cattle (3,21).

Intranasal inoculation of the virus into two steers resulted in a febrile reaction and respiratory disease resembling that described by others (21). Although high titres of virus were detected in nasal secretions from these animals, observations on clinical signs showed that the virus did not induce a severe respiratory disease.

The two heifers exposed to the virus by the genital route both reacted with severe vaginal lesions and development of upper respiratory tract lesions in one of the heifers. High titres of virus were recovered from vaginal secretions of both animals while low quantities of virus were detected in nasal and ocular secretions only from the heifer with respiratory disease. The occurrence of low quantities of virus in the ocular secretions of this animal in the absence of signs of the eye disease was in correlation with the short duration of virus shedding through the eyes.

The highest amount of virus in this study was recovered from ocular secretions of animals inoculated by the conjunctival route. In correlation, the eye disease in these animals constituted the severest disease syndrome observed in this investigation. In addition, these animals also developed mild respiratory signs similar to those observed in intranasally inoculated calves.

In their work on the clinical course of the disease and distribution of virus in various organs, (22) observed a generalised infection following intranasal and intravaginal inoculation of cattle with BHV I. Similar findings were observed in this study particularly following initial intravaginal and intraocular infection. Although the virus did not produce severe respiratory syndrome the development of respiratory disease in six of the eight inoculated animals indicated that this isolate had a predilection for the respiratory system.

Several workers have reported the occurrence of latent BHV I infections in cattle and wildebeest (23,22,19). Such infections are capable of reactivation under conditions of stress such as confinement, travelling long distances or following treatment with synthetic corticosteroids; this is often accompanied by virus shedding. Dexamethasone-induced virus recrudescence was demonstrated in this investigation in all the four treated animals regardless of the route of initial infection. Recurrent infections in the respiratory and genital tract were of a
shorter duration although clinically similar to those observed during the initial infection. Some workers (23,22,24) have observed generalised clinical signs and virus replication following dexamethasone injection of intranasally and intravaginally inoculated calves. In the present investigations, however, the clinical lesions and virus shedding after dexamethasone injection were confined to the initial sites of virus inoculation, and these correlated with initial sites of extensive virus replication. These findings are similar to those of (25) who observed virus shedding only from sites of initial virus inoculation.

(24) could not detect virus in secretions of calves kept in contact with BHV I infected, dexamethasone treated cattle although they could detect virus in the trigeminal nerves only. Similarly, (19) could not demonstrate transmission of infection by contact from dexamethasone injected wildebeest to susceptible cattle and wildebeest. In the present study, calves kept in contact with dexamethasone injected BHV I infected cattle developed only the respiratory form of BHV I and shed virus through the respiratory route. These findings indicate the ease with which BHV I can be transmitted from latently infected cattle following reactivation.

The failure to transmit infection from the intravaginally infected dexamethasone treated cattle to in-contact heifers might suggest that the genital form of BHV I (infectious pustular vulvovaginitis) is probably a venereal disease.

(9) and (26) described clinical cases of genital BHV I infection in Kenyan cattle and isolated the virus from the genital tract. The epidemiological significance of the virus in respiratory and ocular infections of cattle in this country has not been explored. The present isolate probably represents a recrudescence which in respiratory infections in the field could be mild or subclinical and therefore not easily recognisable by the average farmer. However, the primary infection, especially the genital and ocular forms, might be severe and easily noticeable (27,28).

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References

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Isolation of Salmonella from Sick Horses in Sudan

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Isolément de Salmonella des Chevaux Malades au Soudan

Résumé

Au cours d’une épidémie de diarrhée qui a affecté des chevaux de course dans la banlieue de Khartoum, on a trouvé que 13 des 59 chevaux examinés étaient infectés de Salmonellae appartenant à quatre sérotypes de Salmonella, à savoir; S. kentucky, S. eimsbuettel, S. eastbourne et S. locklease var 14. Chacun de ces cas de salmonellose était provoqué par un seul sérotype. Des essais pour traiter les cas chroniques d’infection par Salmonella se sont avérés sans succès et la preuve d’une résistance au médicament a été obtenue grâce à la mise au point de trois types de réactions au médicament, produites par les sérotypes isolés.

Summary

During an epidemic of diarrhoea in race horses in the suburbs of Khartoum, 13 out of 59 horses examined were found infected with Salmonellae that belonged to four Salmonella serotypes which were S. kentucky, S. eimsbuettel, S. eastbourne and S. locklease var 14. Each of these cases of salmonellosis was caused by a single serotype. Attempts made to treat the chronic cases of Salmonella infection were unsuccessful and evidence of drug resistance was obtained from the development of three patterns of drug reactions produced by the serotypes isolated.

Introduction

Equine salmonellosis has a world wide distribution (1) and occurs in several forms including profuse diarrhoea associated with acute colitis and typhilitis in foals and adult horses (2,3), peracute septicaemic disease in foals (4,5), mild enteric disturbances accompanied by fever, anorexia and depression (6) and asymptomatic carrier states (7,3).

In the Sudan sporadic cases of fatal diarrhoea have been observed in mixed-breed horses since 1966 and the situation thereafter recurred with casualties ranging between 10 and 12 annually (personal observation). Unfortunately, the possibility of Salmonella infection was not investigated at that time and animals were considered to have died of diarrhoea of unknown cause.

The recorded history of Salmonella infection in farm animals and birds in the Sudan dates back to 1943 when S. gallinarum was isolated from two outbreaks in chickens (8). A few scattered reports have since been published indicating the existence of disease in other animal species. Following the isolation of S. dublin from cases of poisoning in humans (9), the same serotype was isolated from three diseased calves (10) and 11 Salmonella serotypes were also isolated from apparently healthy cattle (11). The main clinical signs of bovine salmonellosis were dullness, anorexia, lacrimation from both eyes, pyrexia (104-107°F), accelerated pulse and respiration, congested mucous membranes and diarrhoea (10).

In sheep, 27 Salmonella serotypes were isolated from ovine materials collected from the slaughter house (12).

Whereas only eight serotypes were isolated from goats (11), of 422 dogs examined, 104 (23.5%) were infected with 46 serotypes, and of 19 cats examined, two were infected with salmonellae (12).

Generally, although animal salmonellosis had a wide distribution in African countries (12, 11, 13, 14), little work has so far been done to demonstrate the various aspects of the disease problems.
In Sierra Leone three Salmonella serotypes were isolated from five (0.8%) of 605 apparently healthy slaughtered N'Dama cattle whereas two serotypes were responsible for two outbreaks of salmonellosis (15). An outbreak of the disease with diarrhoea, dysentery and abortion involving eight N'Dama cattle was also reported (16). In Zaire, S. nairobi was isolated from an Indo-Brazil zibo which died of an acute enteritis (17). In Senegal, 35 serotypes were recovered from 1,042 mesenteric lymph nodes of slaughtered cattle, while 505 faecal cultures obtained from healthy horses yielded 28 serotypes (18). Fifty-two serotypes were isolated from sheep and goats in Dakar; they constituted an infection rate of 4.7% and 3.6% respectively (19). In Nigeria, surveys carried out by (20) revealed that Salmonella infection rate among dogs was 8%, and resulted in the isolation of no Salmonella organisms from 375 goats examined, and six serotypes from dogs.

Furthermore, (21) isolated S. poona from seven diarrhoeic kids.

The purpose of this article is to report the occurrence of equine salmonellosis for the first time in the Sudan.

**Materials and Methods**

**Culturing Methods**

Faeces were directly collected from the rectum and immediately sent to the laboratory where 1 g from each sample was suspended in 10 ml normal saline (0.85% NaCl), blended with Stomacher 80* for 1-2 min and left to sediment. A loopful and 1 ml of the supernatant were directly plated out onto desoxycholate citrate agar, DCA (Oxoid, CM 227) and transferred into 10 ml selenite broth (Oxoid, CM 395 L 121), respectively. Enriched cultures were incubated at 43°C; subcultures were made onto DCA after 24 and 48 hours incubation and further incubated overnight at 37°C.

Specimens obtained from dead horses including portions of liver, spleen, intestines and caecum, as well as intestinal and caecal scrapings, were similarly prepared and cultured.

**Identification of Salmonella Organisms**

For non-lactose fermenting Salmonella suspect colonies were picked up from each plate and cultured on MacConkey agar (Oxoid, CM 7) and urea slopes (Oxoid, CM 53 + SR 20) and stab-inoculated into triple sugar iron agar, TSI (Oxoid, CM 277). Isolates that gave characteristic reactions of Salmonella were subject to further bacteriological examination (22). Their antigenic structures were determined serologically as indicated by the agglutination reactions with Salmonella agglutination antisera* and confirmed by Dr S. Aleksic, Hygienisches Institut, Hamburg, West Germany.

**In Vitro Drug Sensitivity Tests**

The technique used was as that described by (23), by which a single colony was picked up from an overnight culture on blood agar base No. 2 (Oxoid, CM 271), inoculated into 3 ml nutrient broth No. 2 (Oxoid, CM 67) and incubated at 37°C for 4 hours. Two drops (0.04 ml) from each broth culture were mixed with 3 ml normal saline. The diluted culture was evenly spread onto a diagnostic sensitivity test agar plate (Oxoid, CM 261) and allowed to dry before it was overlaid by the sensitivity disc†. Results were read after overnight incubation at 37°C and bacteria were considered resistant to the drug in question if the inhibition zone was not greater than 2 mm in diameter, measured from the disc margin. Sensitivity discs contained bacitracin (8 units), penicillin (4 units), chloramphenicol (50 μg), neomycin (30 μg), polymixin B (250 units) and streptomycin (25 μg).

**Treatment Trials**

Wherever appropriate, the doses of drugs used were calculated per weight units indicated on basis of live bodyweight. Treatment attempted involved the use via stomach tube of 2-4 litres of liquid paraffin followed by 60 g sodium bicarbonate and either 2-6 oz of kaoline or 22% tannic acid solution, or 70-100 g of carbopolvit (Bayer).

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†Mastring - S. Mast Laboratories Ltd, Liverpool, England
treatment has also included the use of sulphathiazole (4 grains/kg for 4-5 days), tribrisben bolus (Wellcome: 30 mg/kg for 5 days), streptomycin (20 g initially followed by 5 g four times daily for 4 days), liquamycin (Pfizer: 2-5 g for a week), ampicillin powder (4-10 mg/kg daily), neomycin sulphate (Upjohn, Bisol M; one bolus per 45 kg for 3 days), furazolidin (TAD, 7 g for an adult horse), and Lactobacillus acidophilus (12 and 120 g twice daily for a week for foals and adult horses respectively).

Intestinal mobility was reduced with either chlorodyne (0.5-1 fl oz), or tincture of opium (1 fl oz two to three times daily for 5 days), or chloral hydrate (30 g). Faeces of healthy horses were suspended in water and administered via tube to restore normal intestinal flora (24).

Control of dehydration was achieved by the intravenous injections of 3-7 litres of 5% dextrose (Vitor) or 3-7 litres of 0.9% sodium chloride with 1-2 litres Darrow’s solution.

Parenteral administration of antimicrobial drugs for treatment of sick animals was also attempted. These drugs were penicillin (10,000 units/kg) mixed with 10 mg/kg of dihydrostreptomycin (Specia, France), neomycin sulphate (Neobiotic, 1-2 mg/1b every 12 hours), ampicillin (Bristol: 2-7 mg/kg), trivertin (Wellcome: 1 ml / 10 kg), borgal 24% (Hoechst: 1 ml/10 kg) and chloramphenicol (chloromycetin, Parke Davis; 4-10 mg/kg for large animals and 2-4 mg/kg for foals).

Vitamin B complex (Aquab; Upjohn, 8-15 ml) and cortizones (Predef 2x; Upjohn, 5-20 mg for adult horses) were frequently used.

Results

Bacteriological Examination

Table 1 shows that Salmonella organisms were isolated from 13 of 59 animals examined. Four Salmonella serotypes, S. eastbourne, S. eimsbuetell, S. kentucky and S. locklease var 14, were isolated from the faeces and tissue specimens obtained from live and dead animals. S. kentucky was also isolated from a faecal sample collected from a diseased calf that was held in-patient in the veterinary clinic attached to the stud (property A; Table 1). Freedom of stud horses from Salmonella infection after the last incidence of the disease was ascertained by failure to isolate Salmonella organisms from four successive faecal samples collected from each animal within a fortnight.

Drug Sensitivity Reactions

All 14 Salmonella cultures examined were sensitive to chloramphenicol and polymixin B and resistant to bacitracin and penicillin. Seven cultures which included those of S. eastbourne, S. eimsbuetell, and S. locklease var 14, and three cultures of S. kentucky were also sensitive to neomycin and streptomycin. On the contrary, six cultures of S. kentucky that included the bovine strain were resistant to the latter drugs. Only one culture of S. kentucky was sensitive to streptomycin and resistant to neomycin.

Clinical Signs

During 1980, the incidence of enteric disturbances in race horses in Khartoum area had acquired the nature of an epidemic, involving varying numbers

<table>
<thead>
<tr>
<th>Property</th>
<th>No. at risk</th>
<th>No. with diarrhoea</th>
<th>No. examined</th>
<th>No. positive</th>
<th>Salmonella serotypes isolated*</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>14</td>
<td>2</td>
<td>14</td>
<td>2</td>
<td>S. kentucky (1) and S. locklease var 14 (1)</td>
</tr>
<tr>
<td>B</td>
<td>50</td>
<td>6</td>
<td>23</td>
<td>3</td>
<td>S. kentucky (3)</td>
</tr>
<tr>
<td>C</td>
<td>40</td>
<td>20</td>
<td>8</td>
<td>3</td>
<td>S. kentucky (2) and S. eimsbuetell (1)</td>
</tr>
<tr>
<td>Others</td>
<td>NK</td>
<td>14</td>
<td>14</td>
<td>5</td>
<td>S. kentucky (3), S. eimsbuetell (1) and S. eastbourne (1)</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>42</td>
<td>59</td>
<td>13</td>
<td>S. kentucky (9), S. eimsbuetell (2), S. eastbourne (1) and S. locklease var 14 (1)</td>
</tr>
</tbody>
</table>

NK - Not known.

A - Government stud; B and C - private stud farms. Others - animals belonged to twelve different possessions.

*Numbers in brackets indicate the number of positive cases in relation to the respective serotype.
<table>
<thead>
<tr>
<th>Case No.</th>
<th>Description</th>
<th>Age</th>
<th>Infection</th>
<th>Strain isolated</th>
<th>Antimicrobial drugs used</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Three-quarter thoroughbred filly</td>
<td>1</td>
<td>Chronic</td>
<td>S. eimsbuetelli</td>
<td>SU, TE, C, N, FR, La</td>
<td>Improved</td>
</tr>
<tr>
<td>2</td>
<td>Half thoroughbred pregnant broodmare</td>
<td>6</td>
<td>Chronic</td>
<td>S. kentucky</td>
<td>Su, S, TE, C, N, FR, La</td>
<td>Aborted 10 months foetus and died</td>
</tr>
<tr>
<td>3</td>
<td>Half thoroughbred colt</td>
<td>4</td>
<td>Acute</td>
<td>S. eimsbuetelli</td>
<td>—</td>
<td>Died</td>
</tr>
<tr>
<td>4</td>
<td>Native mare</td>
<td>10</td>
<td>Acute</td>
<td>S. eastbourne</td>
<td>PS</td>
<td>Improved</td>
</tr>
<tr>
<td>5</td>
<td>Half thoroughbred filly</td>
<td>4</td>
<td>Chronic</td>
<td>S. kentucky</td>
<td>PS, TR, N</td>
<td>Died</td>
</tr>
<tr>
<td>6</td>
<td>Three-quarter thoroughbred mare</td>
<td>6</td>
<td>Acute</td>
<td>S. kentucky</td>
<td>PS, N</td>
<td>Treated and mated, it conceived and delivered a normal foal</td>
</tr>
<tr>
<td>7</td>
<td>Half thoroughbred broodmare</td>
<td>10</td>
<td>Chronic</td>
<td>S. kentucky</td>
<td>SU, TR, C, N, FR</td>
<td>Died</td>
</tr>
<tr>
<td>8</td>
<td>British thoroughbred stallion</td>
<td>4</td>
<td>Acute</td>
<td>S. kentucky</td>
<td>N, FR</td>
<td>Died</td>
</tr>
<tr>
<td>9</td>
<td>British thoroughbred stallion</td>
<td>8</td>
<td>Acute</td>
<td>S. locklease</td>
<td>C, N, FR</td>
<td>Treated</td>
</tr>
<tr>
<td>10</td>
<td>Half thoroughbred filly</td>
<td>2</td>
<td>Chronic</td>
<td>S. kentucky</td>
<td>SU, C, N, FR</td>
<td>No response to treatment</td>
</tr>
<tr>
<td>11</td>
<td>Three-quarter thoroughbred filly</td>
<td>3</td>
<td>Chronic</td>
<td>S. kentucky</td>
<td>PS, TR, C, N, FR, La</td>
<td>Died</td>
</tr>
<tr>
<td>12</td>
<td>British thoroughbred broodmare</td>
<td>3</td>
<td>Chronic</td>
<td>S. kentucky</td>
<td>PS, TR, C, N, La</td>
<td>No response to treatment</td>
</tr>
<tr>
<td>13</td>
<td>Half thoroughbred pregnant broodmare</td>
<td>7</td>
<td>Chronic</td>
<td>S. kentucky</td>
<td>PS, C, N, La</td>
<td>Aborted and improved</td>
</tr>
</tbody>
</table>

PS — penicillin and streptomycin; S — streptomycin; SU — sulphathiazole; TE — terramycin (Liquamycin); C — chloramphenicol; N — neomycin; FR — furazolidone; TR — triple sulph; La — Lactobacillus acidophilus.

Post Mortem Examination
Lesions observed at necropsy were pulmonary oedema, subendocardial echymotic haemorrhages, severe congestion of the liver, kidneys and lungs, catarrhal enteritis, haemorrhagic typhilitis, colitis and severe adrenal

of animals that belonged to 15 different farms. The main clinical signs displayed were depression, pyrexia (103-104°F), profuse diarrhoea, congested mucous membranes, increased intestinal mobility and rapid thready pulse. Colic and laminitis were infrequently observed.
cortical haemorrhage (cases 3 and 8, Table 2). In case No. 5 (Table 2), both ventricles of the heart were studded with small greyish areas (2-4 mm in diameter), lungs were congested and oedematous, the liver was friable and moderately enlarged and the kidneys were slightly enlarged and had a tense capsule which when peeled off revealed an uneven surface.

The main histopathological findings were hepatic hydropic degeneration, focal myocardial infarction and degeneration and interstitial nephritis and proliferative glomerulonephritis.

**Discussion**

It is recognised that occurrence of salmonellosis in animals is closely related to the husbandry methods and there is evidence that intensification of livestock favours the incidence of Salmonella infection (25). It is also suggested that high stocking densities predispose horses to salmonellosis and with a few exceptions, equine salmonellosis would only take place when precipitated by one or more of some other agents or causes including stress (26, 1). With one exception (property A; Table 1), all horses reported here were kept in heavily contaminated environments due to inadequate measures of disinfection, lack of hygienic disposal of contaminated materials and the ignorance of attendants and animal owners of the dangers of spreading infection. This situation may have been responsible for the recurrence of the disease because it provided a permanent source of infection.

This report records for the first time the isolation of Salmonellae from horses in the Sudan. *S. kentucky* had not been isolated from animals or birds in this country before and it appeared to have a wider distribution in the horse population than the other serotypes isolated. Although the source of infection of the bovine host with *S. kentucky* could not be accurately determined, its possible association with the disease which was caused by the serotype as in case No. 8 (Table 2) was suggested. The fact that both cultures had the same pattern of drug sensitivity reactions, diarrhoeic involvement in the calf followed infection in the horse, and the management and husbandry conditions of the stud could not preclude the spread of infection to the veterinary clinic attached. Of the remaining serotypes reported here, *S. eimsbuttei* was isolated from a stray dog that was killed during an anti-rabies campaign in Khartoum province (12), while *S. eastbourne* was also isolated from eight dogs (12) and one sparrow hawk (13); *S. lockelease var 14* was a new serological variant (27).

Treatment of horses with chronic salmonellosis was also unsuccessful and it appeared to have only a transient effect. This observation may be partially explained by the existence of contaminated environments and the questionable efficacy of antibacterial drugs and their tendency to develop drug resistant enteric bacteria (28, 7) particularly when they are used for the elimination of Salmonella organisms from the intestines of infected horses.

It may be suggested that chronic salmonellosis follows a severe acute case of the disease and is produced as a result of caecal and colonic changes and their subsequent intensive tissue reaction. One horse (case No. 8, Table 2) showed acute signs of salmonellosis that had some resemblance to the peracute septicaemic infection described in foals (4, 5, 1).

Although fluid therapy directed towards the correction of electrolyte imbalance can be very effective in the treatment of equine salmonellosis (2) and is recommended for treatment of diarrhoeic disturbances (29,28,30) its beneficial effect could not be separately demonstrated because it was always used in combination with antibacterial drugs in all the animals included here.

The incidences of abortion were neither immediately reported to us nor were appropriate samples collected and sent to the laboratory and so their association with Salmonella infection could not be established. However, *S. kentucky* was isolated from the vaginal excretions of one mare during oestrus 10 days following the incidence of abortion.

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References


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Use of SBX Dye in Plasma and Blood Volume Measurements in the Nigerian Indigenous Pig from Birth to Weaning

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Utilisation du Colorant SBX pour Mesurer le Volume du Plasma et du Sang Chez les Porcelets Indigènes du Nigéria de la Naissance au Sevrage

Résumé

Le volume du plasma et du sang chez les porcelets autochtones du Nigéria, âgés d'un jour à 4 semaines, a été déterminé grâce au colorant pontamine bleu ciel (SBX). Afin de calculer ces paramètres, l'hématocrite (PCV) et l'hémoglobine (Hb) ont été obtenus. Les poids vitaux variaient entre 0,75 et 6 kg; le PCV entre 22 et 40%; Hb entre 7 et 13 g; le volume du plasma entre 325 et 2,545 ml et le volume du sang entre 950 et 3,000 ml. Il y avait des différences selon le sexe dans les chiffres obtenus, les femelles ayant des volumes de plasma (P > 0,05) et de sang inférieurs à ceux des mâles. Les volumes du plasma et du sang augmentaient (P > 0,05) avec l'âge.

Summary

The plasma volume and blood volume in Nigerian indigenous piglets from day-old to 4 weeks of life were determined using pontamine sky blue (SBX). In order to calculate these values the PCV and Hb values were obtained. The live bodyweights ranged between 0.75 and 6.0 kg; PCV 22-40%; Hb 7-13g; plasma volume 325-2,545ml and blood volume ranged between 950 and 3,000ml. There were sex differences in the values obtained, the female having lower (P > 0.05) plasma and blood volumes than the male. Plasma and blood volumes increased (P > 0.05) with age.

Introduction

Although there are several reports on the plasma and blood volume measurements in conventional pigs (1,2,3) there are none on Nigerian indigenous pigs. Packed cell volume (PCV), haemoglobin (Hb) content, erythrocyte counts and other haematological values have been known to be altered with blood volume changes (4), thereby making the diagnosis of haemoconcentration and haemodilution difficult. (5) have reported variations in some haematological values between the Large White and the Nigerian indigenous pig. This investigation was carried out to determine the plasma and blood volume measurements in this breed of pig.

Materials and Method

Twenty-one male and female piglets used in this study were from four apparently healthy sows that were regularly dewormed and kept intensively on a concrete floor at the University Teaching and Research Farm. Their ages ranged from day-old to 4 weeks. Each animal was weighed weekly and plasma and blood volume determined. 5ml of blood was taken from each animal into a bottle containing di-sodium salt of ethylenediamine tetraacetic acid (EDTA) as anticoagulant. From this sample, the PCV was determined using the microhaematocrit method. The PCV was multiplied by 0.97 to correct for the amount of plasma trapped in the packed cell fraction (2). The Hb content was estimated by cyanmethaemoglobin method. The remaining blood sample was centrifuged at 3,000 rpm for 20 min to obtain the plasma.

A 10% pontamine sky blue dye (SBX) solution was injected into the anterior vena cava of each piglet at the rate of 10mg/kg body weight. After 10 min, by which time a complete mixing of the dye would have occurred, another 5ml blood sample was withdrawn from the vessel
opposite that used for injecting the dye for plasma extraction. The dilution of the dye in each plasma was determined by measuring the optical density on a Cecil Colorimeter at 540μm using the plasma free of dye as blank. A standard dye solution of 40 μg/ml was made and its optical density noted on the same instrument, using distilled water as blank. The plasma volume and total blood volume were calculated as described by (5).

Results and Discussion

The results are shown in Table 1 and Figure 1. The mean values for Hb and PCV were lowest at the third week of life but there was no significant (P > 0.10) difference when compared with the values for the 1 week and 4 week old piglets. Similarly, the values in the female were lower than those for the males, however, this difference was not significant (P > 0.10).

![Figure 1: Plasma volume and blood volume with increase in age.](image)

<table>
<thead>
<tr>
<th>Age</th>
<th>Sex</th>
<th>Weight (kg)</th>
<th>Hb (g%)</th>
<th>PCV (%)</th>
<th>Plasma volume (kg/bodywt)</th>
<th>Blood volume (kg/bodywt)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day-old</td>
<td>M (5)</td>
<td>0.94 ± 1.0</td>
<td>12.6 ± 1.2</td>
<td>35 ± 2.5</td>
<td>21.1 ± 3.5</td>
<td>23.5 ± 2.5</td>
</tr>
<tr>
<td></td>
<td>F (6)</td>
<td>0.80 ± 0.5</td>
<td>8.7 ± 2.1</td>
<td>28 ± 3.6</td>
<td>32.7 ± 5.2</td>
<td>34.4 ± 3.2</td>
</tr>
<tr>
<td>1 week</td>
<td>M (4)</td>
<td>1.8 ± 0.5</td>
<td>11.8 ± 1.2</td>
<td>35 ± 2.5</td>
<td>60.7 ± 3.4</td>
<td>71.4 ± 6.5</td>
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<tr>
<td></td>
<td>F (6)</td>
<td>1.6 ± 0.5</td>
<td>9.9 ± 2.0</td>
<td>30 ± 3.0</td>
<td>51.5 ± 5.1</td>
<td>60.6 ± 5.1</td>
</tr>
<tr>
<td>2 weeks</td>
<td>M (5)</td>
<td>2.3 ± 1.5</td>
<td>10.9 ± 2.3</td>
<td>35.5 ± 6.0</td>
<td>72.5 ± 4.2</td>
<td>87.5 ± 3.5</td>
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<td>F (5)</td>
<td>2.5 ± 0.9</td>
<td>9.8 ± 2.5</td>
<td>32.0 ± 4.5</td>
<td>64.3 ± 6.0</td>
<td>75.0 ± 3.0</td>
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<tr>
<td>3 weeks</td>
<td>M (6)</td>
<td>3.2 ± 0.7</td>
<td>9.8 ± 1.2</td>
<td>27.5 ± 2.5</td>
<td>73.0 ± 3.0</td>
<td>88.6 ± 2.5</td>
</tr>
<tr>
<td></td>
<td>F (4)</td>
<td>2.9 ± 0.6</td>
<td>7.9 ± 1.5</td>
<td>25.0 ± 3.0</td>
<td>66.4 ± 2.5</td>
<td>80.1 ± 3.0</td>
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<td>4 weeks</td>
<td>M (4)</td>
<td>5.3 ± 2.1</td>
<td>10.1 ± 0.5</td>
<td>37.0 ± 4.2</td>
<td>82.5 ± 3.3</td>
<td>94.4 ± 4.3</td>
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<td>F (4)</td>
<td>4.4 ± 0.5</td>
<td>9.9 ± 0.4</td>
<td>35.0 ± 3.5</td>
<td>77.5 ± 7.6</td>
<td>91.5 ± 5.0</td>
</tr>
</tbody>
</table>

(No. of animals in parenthesis)

There was an increase in plasma and blood volume values with increasing age and weight, although there was no difference between the values in the 2 and 3 week old piglets. This was manifested in the lowered values for Hb and PCV at that age. The female piglets had lower values (P > 0.05) than males of same age.

Previous workers (3) reported decreasing total blood volume per kg bodyweight in the iron dextran treated pigs from 90.0 ± 6.1 ml/kg at birth to 81.2 ± 10.5 at 6 weeks of age. However, the values for their control pigs fluctuated above and below the non-dextran treated pig values. The Nigerian indigenous pigs used in this study were not iron dextran treated and probably this might have contributed to the increase in plasma volume and blood volume with age. (4) suggested that physiologic anaemia
could cause a significant increase in plasma volume. Similarly, it was shown that the reduction in plasma volume was an indirect result of the iron treatment that caused an increase in total red cell mass (4).

Acknowledgement
The authors are grateful to Messrs Iruoje and Ogunpolo for their technical assistance.

References

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The Erythrocyte Osmotic Test of Nigerian Indigenous Piglets

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Test de Résistance Globulaire Chez les Porcelets Indigènes au Nigéria

Résultats

Une recherche a été effectuée sur le test de résistance globulaire chez les porcelets indigènes du Nigéria, âgés 1 jour à 6 semaines. Certains paramètres hématologiques ont été aussi évalués afin de déterminer s’ils influaient sur la propriété osmotique de la membrane des érythrocytes.

L’hématocrite (PCV), l’hémoglobine (Hb), le nombre d’érythrocytes (Rbc) et le sexe n’avaient pas d’influence directe sur la résistance osmotique des érythrocytes qui étaient toutefois influencées par le volume corpusculaire moyen (VCM). Le VCM était plus élevé pendant la première semaine de vie chez cette race de porcelets et une plus grande résistance osmotique a été observée au cours de la même période. Cela prouve que plus petit est le globule rouge, plus faible est sa résistance à l’hémolyse.

Les fragiligrammes montraient également des érythrocytes de type foetal et de type adulte, les premiers diminuent et les derniers augmentent proportionnellement avec l’âge.

Summary

Erythrocyte fragility tests of Nigerian indigenous piglets between day-old and 6 weeks of age were investigated. Some haematological parameters were also estimated to determine if they influence the osmotic property of the erythrocyte membrane.

Packed cell volume (PCV), haemoglobin (Hb), erythrocyte count (Rbc) and sex had no direct influence on the osmotic fragility of erythrocytes which was influenced, however, by the mean corpuscular volume (MCV). MCV value was largest in the first 1 week of life in this breed of piglets and higher osmotic resistance was obtained during this period. This showed that the smaller the size of a red cell, the lower its resistance to haemolysis.

The fragiligrams also showed the erythrocyte populations of foetal-type and adult-type, with the former diminishing with age and latter increasing regularly with age.

Introduction

The extent to which the osmotic pressure of plasma can be lowered without causing haemolysis of the corpuscles varies considerably among different species (1). Data on the erythrocyte osmotic fragility in young domestic animals are limited to those domestic breeds kept in the temperate zone. The erythrocyte fragility test has some clinical application; it is of importance in correction of solutions for intravenous injection, and it has also been shown that the resistance of the erythrocyte varies in some diseases (2).

The Nigerian indigenous pig is being actively studied in order to establish its physiological parameters and productivity. There is therefore the need to establish the osmotic fragility in piglets of this relatively unknown breed of pig which will serve as a reference point in clinical investigations of haematological disorders.

Materials and Methods

The 35 piglets of both sexes used in this study were from clinically normal Nigerian indigenous sows kept intensively at the University of Ibadan Teaching and Research Farm. Their ages varied from day-old to 6 weeks.

Blood was drawn from the anterior vena cava of each piglet at day-old and at weekly intervals up to the sixth week of age using ethylene diaminetetra-acetic acid (EDTA) as anticoagulant. Haemoglobin (Hb) and haematocrit (CV)
values were estimated in each sample by the cyanmethaemoglobin and microhaematocrit methods respectively. Erythrocyte population in each sample was counted using the haemocytometer.

Five ml of various dilutions of 10% phosphate buffered sodium chloride (NaCl) in distilled water were placed in 15 centrifuge tubes. This dilution gave NaCl concentrations of 0.85% in the first tube and 0.1% in the fifteenth tube. 0.02 ml of blood was added to each tube. The contents were mixed and allowed to stand at room temperature (28-29°C) for 30 min. The tubes were then centrifuged at 2,000 rpm for 10 min and about 3.5ml of supernatant was later transferred to another set of 15 tubes. The optical density of each solution was read on a Cecil Colorimeter at 540 μM wavelength against water as blank. The percentage of haemolysis was calculated by assuming haemolysis in the distilled water as 100%. The fragilogram was obtained by the method of gradual haemolysis (3). Cumulative fragilogram was obtained by plotting percent haemolysis against saline concentrations. All data were subjected to student t statistical analysis.

Results

The mean values of Rbc, MCV and the salt concentrations at which minimum (less than 5%) and maximum haemolysis (above 95%) occurred with the erythrocytes are presented in Table 1. The mean PCV and Hb values were lowest at the third week of life. The mean PCV value at 3 weeks was significantly lower (P < 0.05) than the values at 2 and 4 weeks old respectively. There was no significant difference between the mean Hb values at 3 weeks and those at either 2 or 4 weeks of age. There was a significant

![Figure 1: Cumulative (C) and derivative (D) fragilogram of the day-old indigenous piglets.](image)

(P < 0.05) 60% reduction, in the mean erythrocyte count in the third week relative to the values at birth. The difference was also significant when compared with the values for 2 week old (P < 0.05) and 4 week old (P < 0.05) piglets.

<table>
<thead>
<tr>
<th>Age</th>
<th>Mean PCV (%) ± S D</th>
<th>Mean Hb (%) ± S D</th>
<th>Mean Rbc (x 10^6) ± S D</th>
<th>Mean MCV (μL) ± S D</th>
<th>Min. resistance</th>
<th>Max. resistance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day old (4)</td>
<td>35.5 ± 4.9</td>
<td>9.5 ± 1.6</td>
<td>5.3 ± 1.5</td>
<td>66.4 ± 0.6</td>
<td>0.65</td>
<td>0.45</td>
</tr>
<tr>
<td>1 week (4)</td>
<td>33.5 ± 7.8</td>
<td>8.4 ± 0.9</td>
<td>4.2 ± 0.9</td>
<td>80.5 ± 2.8</td>
<td>0.65</td>
<td>0.30</td>
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<tr>
<td>2 weeks (6)</td>
<td>25.5 ± 6.0</td>
<td>7.4 ± 3.2</td>
<td>4.0 ± 0.8</td>
<td>62.8 ± 2.5</td>
<td>0.80</td>
<td>0.50</td>
</tr>
<tr>
<td>3 weeks (6)</td>
<td>20.7 ± 3.3</td>
<td>6.4 ± 0.9</td>
<td>3.7 ± 0.5</td>
<td>56.1 ± 2.5</td>
<td>0.80</td>
<td>0.50</td>
</tr>
<tr>
<td>4 weeks (5)</td>
<td>25.3 ± 3.2</td>
<td>8.1 ± 1.1</td>
<td>5.0 ± 0.2</td>
<td>51.0 ± 4.9</td>
<td>0.80</td>
<td>0.45</td>
</tr>
<tr>
<td>5 weeks (5)</td>
<td>30.3 ± 2.1</td>
<td>9.6 ± 1.0</td>
<td>6.0 ± 0.5</td>
<td>51.0 ± 8.3</td>
<td>0.80</td>
<td>0.50</td>
</tr>
<tr>
<td>6 weeks (5)</td>
<td>38.3 ± 2.9</td>
<td>11.6 ± 0.6</td>
<td>7.1 ± 0.7</td>
<td>53.9 ± 6.2</td>
<td>0.80</td>
<td>0.50</td>
</tr>
</tbody>
</table>

(No. in parenthesis indicate no. of piglets used)
Mean corpuscular volume (MCV) decreased significantly (P < 0.05) from the first week (80.5 ± 2.8 μm³) to the third week (56.1 ± 2.5 μm³). The erythrocytes were largest during the first week of life measuring between 66 and 82 μm³.

Higher osmotic resistance was obtained in the very young piglets (minimum resistance 0.65% saline and maximum resistance at 0.30-0.45% saline), showed the cumulative (C) and derivative (D) fragiligrams in the indigenous piglets at day-old and at 1, 2

Figure 2: Cumulative (C) and derivative (D) fragiligram of the 1 week old indigenous piglets.

Figure 3: Cumulative (C) and derivative (D) fragiligram of the 3 week old indigenous piglets.

and 5 weeks of age. The cumulative curves obtained were all sigmoid with differences in the steepness. The derivative curves showed two peaks at the different ages and this was not well indicated in the day-old piglets.

Discussion

The decline in the haematological parameters occurring at the third week was similar to the reported decline in these parameters in conventional pigs at the tenth day, which occurred as a result of the rapid growth and lack of significant body reserve of iron (4). Lack of supplementary iron may account for the more pronounced decline in Large White piglets.

Initial haemolysis occurred within the range of salt concentration reported for piglets in the temperate zone, but complete haemolysis occurred faster in the exotic piglet erythrocytes (1). The erythrocytes of the very young piglets less than 1 week old had higher MCV values and were more resistant. The older piglets had low MCV values and higher osmotic fragility when compared with younger piglets. The MCV values obtained here were in conformity with values earlier reported in conventional pigs (5).

The derivative fragiligram (Figs 2, 3 and 4) showed two clear peaks indicating two erythrocyte populations. The more
resistant erythrocytes found in the very young were foetal-type and the osmotically more fragile population were adult-type erythrocytes. The ratio of this mixed population and the age at which the erythrocyte fragility curve developed into the adult type had been found to vary widely in different species (2). In Figures 2-4, the first peak corresponded to the adult-type population of erythrocyte which increased in the circulation and the second peak corresponded to a more resistant population diminishing with age.

From the results obtained in this study it would appear that the osmotic fragility of erythrocytes of indigenous piglets was higher than that reported in conventional piglets. Also erythrocyte osmotic fragility was not influenced by erythrocyte count, Hb and PCV values, but MCV decrease early in life can be associated with an increase in osmotic fragility of the erythrocytes.

References


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Cervical Mucus Patterns of Two Tropical Breeds of Cattle: Muturu — *Bos brachyceros* and Ndama — *Bos taurus*

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Aspects du Mucus Cervical de Deux Races Tropicale de Bovins: Muturu — *Bos brachyceros* et Ndama — *Bos taurus*

Résumé

Le changements d’aspect du mucus cervical qui accompagnent l’oestrus et d’autres phases du cycle oestral ont été étudiés chez des bovins tropicaux indigènes non-gravides (des génisses Muturu et Ndama). La durée du cycle oestral était respectivement de 19.5 ± 0.8 et 18.84 ± 0.44 jours pour les génisses Muturu et Ndama utilisées. Des quantités variables de mucus cervical ont été prélevées des vaches au cours des différentes phases du cycle oestral. Quatre formes de cristallisation ont été observées, à savoir: A, B, C et D. L’oestrus se produisait et le mucus cervical était plus abondant quand on a enregistré une arborisation de type A. Les aspects du mucus cervical chez les deux races ont été comparés et rapprochés aux études antérieures sur le même sujet.

Summary

The changes in the pattern of the cervical mucus that accompany oestrous and other phases of the oestrous cycle were studied in non-pregnant indigenous tropical cattle — Muturu and Ndama heifers. Oestrous cycle length was 19.5 ± 0.8 and 18.84 ± 0.44 days for the Muturu and Ndama heifers used respectively. Variable quantities of cervical mucus were collected from the cows during the various phases of the oestrous cycle. Four forms of crystallisation patterns were observed, viz. A, B, C and D. Heat occurred and cervical mucus was most copious when A arborisation pattern was recorded. Cervical mucus patterns in the two breeds were compared and also related to previous reports in the literature.

Introduction

Cervical mucus of the reproductive tract has been studied essentially to determine changes in the mucus at various phases of the oestrous cycle in cows (breed unspecified) (1); in White Fulani crossbreds (2); to determine the time of ovulation in women (3); to determine ovarian function in cows (4); as an aid in the detection of heat in cows (5,6), and in assessing the endocrine status of non-pregnant sows (7). There is, however, no report in the literature on cervical mucus of trypanotolerant beef breeds of tropical cattle which though largely unimproved genetically are used for beef production and traditional ceremonies in the generally traditional societies of the forest belt of West Africa. The present study is aimed at comparative evaluation of cervical mucus patterns of two indigenous tropical breeds of cattle Ndama (*Bos taurus* cattle) and Muturu (*Bos brachyceros* cattle) and their relationship to oestrous cycle in these cattle breeds.

Materials and Methods

Animals

A total of 12 heifer cows seven Ndamas and five Muturus, 2½ to 3 years of age were used for the study. The cows were reared intensively, grazed in enclosed paddocks and supplemented with Brewers' Spent grain and palm kernel meal and silage. The study lasted for 3 months in the dry season; ambient temperature, 26.0°C to 32.5°C and 50-60% relative humidity.

Experimental Procedure

All the cows were observed daily for external signs of heat before the collection of cervical mucus from the cervical os. In the process, the heifers were led singly from the crowding area into the crush, the external genitalia examined for swelling and redness, washed with soap and water (a separate towel for each animal) and rinsed in clean water. With the aid of an assistant holding
the tail to one side, a sterile bivalve vaginal speculum lubricated with sterile K-Y jelly (Johnson and Johnson, USA) was carefully inserted into the vagina and opened. Using a microtorch light, the rounded end of a plastic catheter, 45 cm long by 4 mm inner diameter was guided 1-2 cm into the os of the cervix. Cervical mucus was drawn into the tube by aspiration with the aid of an affixed hand pump. The flow characteristics (quantity and viscosity) of the mucus were recorded. A separate catheter was used for each animal. Smears were thereafter made on labelled clean glass slides and left to dry at room temperature. The slides were first examined under reduced illumination with low power (x 100) within 8 hours of collection and later stored.

Each smear was assessed for arborisation using (1) method of classification of cervical smear patterns as belonging to one of four classes: A, B, C and D. A representative sample of each class was later selected for photography.

Analysis of Data

Although smear collection started simultaneously for all the cows, the first day of heat for each cycle for each cow was taken as day 0 and the daily records arranged left and right of the reference day for close study. Tables showing the pattern of occurrence of the cervical mucus, and also histograms showing the relative frequency of occurrence of each arborisation pattern on each day of the oestrous cycle were drawn and compared.

Results

Length of Oestrous Cycle

The length of oestrous cycle in the Ndama heifers used for the study was 18.84 ± 0.44 days with a range of 14-23 days. In the case of Muturus, it was 19.5 ± 0.81 days, range 15-22 days. The histogram showing the relative frequency of occurrence and the pattern of distribution of the oestrous cycles in the two breeds is presented in Figure 1. The modal value of the oestrous cycle of the Ndamas was 19 days while that of the Muturus was 20 days. Respectively, 80% and 84% of all the oestrous cycles of the Ndamas and Muturus studied were in the range of 17 and 22 days.

Figure 1: Frequency distribution of the length of oestrous cycle in Ndama and Muturu cattle.
Figure 2: (i)-(viii) Photomicrographs showing crystallisation patterns A to D in the two cattle breeds — Ndama and Muturu, x 100.
Flow Characteristics of Cervical Mucus

In both breeds, on the average 2-3 days before the onset of behavioural heat and 2-4 days after, mucus was very easily obtained, light, thin and copious with low viscosity. With the hand pump, half the length of the catheter could be filled with mucus. About 4-8 days after heat, mucus obtained was less clear, more turbid and elastic and one-quarter of the length of the catheter could still be filled by aspiration. About 9-13 days after heat on the average, the mucus obtained became more viscous and required up to 1-2 min to draw some quantity of mucus up the catheter. About 14-17 days after heat, the mucus was highly viscous, scanty with thick consistency and the mucus could hardly be said to flow. Most of the heifers fitted into the above broad classes although in one or two cycles in one or two heifers, there were deviations.

Arborisation Patterns

The arborisation patterns looked similar in both breeds although with minor variations in intensity. The four basic types of arborisation as reported by (1) were observed: type A pattern represented a well-formed ferning pattern that could be seen in over 90% of the smears. The stem of the ‘ferns’ were thin, long and wavy with clearly developed variations and subvenations (Fig. 2: (i) Ndama and (ii) Muturu). These type smears were obtained around the time of heat. Type B arborisation patterns had positive ferning also, but the stems were thicker and the ferning was not as intense as in A (Fig. 2: (iii) Ndama and (iv) Muturu). Type C ferning was very weakly positive with scattered irregularly occurring ferning over the smear (Fig. 2: (v) Ndama and (vi) Muturu). Type D smears had no crystallisation or ferning at all (Fig. 2: (vii) Ndama and (viii) Muturu). These later smears occurred when it was extremely difficult to collect mucus from the cervix. The sequence of occurrence of these ferning types in the Ndamas was A, B, C, D although in some cases it was A, B, D, C; in the Muturus it was the same except in one cycle in one of the heifers when it was C, A, B, C and in another, it was A, B, D, C.

When the ferning patterns were classified simply as either positive or negative, the percentage score in the two breeds relative to the day of heat is shown in Table 1. Positive ferning was observed in all smears at heat and declined thereafter before and after heat.

| Score (%) | Ndama |  | Day of cycle | |  | Muturu |  | Score (%) | No. showing ferning smear | No. of smears | Day of cycle relative to day of heat | | No. showing ferning pattern | Score |%
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Cervical Mucus Spread

The spread of type A arborisation pattern was over a range of 1-7 days with a mode of 4 days duration for the Ndamas; in the Muturus it covered a range of 1-10 days, with 1-6 days being the most frequent. Type B had a range of 1-7 days in both the Ndamas and Muturus with 1-4 days being commonest. Type C pattern had a range of 1-5 days, with 1-2 days forming over 90% of all the cases of C recorded. Type D patterns had a spread of 1-4 days although in most cases, in both breeds, they occurred as intermittent 1-day smears that broke a continuous spread of C pattern. The histogram showing the pattern of distribution of the ferning patterns is shown in Figure 3. The type A pattern in Ndamas and Muturus was concentrated.
from -3 days to heat to 2 days after, although in the case of Muturus, pattern A smear was found in other days in some other cycles but to a lesser degree. The peak of occurrence of type A pattern in both breeds, being concentrated mainly from the second day after heat to the eighth day in Ndamas but to the ninth day in Muturus. Type C patterns were next in sequence, the percentage of occurrence showed a gradual rise from day 7 after heat (Fig. 3) reaching the peak at mid-cycle when both cycle patterns — C and D, appeared to occur in greater percentages in both breeds. The transition from types C and D to type A appeared to be abrupt and did not show a gradual transition leading up to A.

### Discussion

The length of oestrous cycle in Ndamas although generally slightly lower than that of Muturus studied, was not significantly different and was still within the normal range of oestrous cycles reported for beef breeds of cattle (5,2). However the slight differences confirm the importance of breed variation in the all important physiological event.

All the cows yielded cervical mucus whose intensity varied with the stage of oestrous cycle and whose pattern of occurrence was consistent with patterns of cervical mucus in other breeds of cattle (1), White Fulani cows (2), and beef cows (5). Although ferning was greatest on the

<table>
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<th>Ndama</th>
<th>Muturu</th>
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<td>Type D</td>
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<td>Type C</td>
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<td>Type B</td>
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<tr>
<td>Type A</td>
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**Figure 3**: Frequency distribution of the crystallisation pattern in Ndama and Muturu cattle
day of oestrus as shown in Figure 3, Ndama cow no. 1012 showed type B pattern on the day of standing heat in one of the cycles. Besides, intensively positive ferning (type A pattern) started some 3-4 days or more before the onset of heat and did not terminate in many cycles until upwards of the equivalent duration after heat. Judging from the spread of the various ferning types (Fig. 1), although the very distinct ferning types associated with oestrogen activity (type A) were easily discernible and the total lack of ferning (type D) also, it may be difficult from one or two samples to predict the exact physiological state of the cows especially in relation to the day of the oestrous cycle. If, on the other hand, cervical mucus smears are made over an extended period, it may be possible to estimate the nature of activities of the ovaries especially as ferning has been highly correlated with ovarian activity (1,8,7).

Acknowledgement

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References


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Isolation and Identification of Goat Pox Virus in Nigeria

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Introduction

The population of goats in Nigeria is estimated to be about 24 million (1). (2) reviewed the microbial diseases of goats in Nigeria. Earlier, (3) had mentioned the occurrence of goat pox in northern Nigeria where the Hausa population called it 'Agana'. Mortality was said to be about 50% of infected goats and slightly higher in the young stock. The lesions produced in the skin were of great economic importance because such skin would fetch little money or had no market value.

Other pox virus infections of sheep and goats have been described (4,5,6,7). But there appears to be no confirmatory evidence on the occurrence of goat pox in Nigeria (9).

The clinical signs, virus isolation and identification of goat pox in Nigeria are reported herein.

Materials and Method

History of the Disease Outbreak

In January 1981, outbreaks of skin disease occurred in three farms in Ibadan, Oyo State. The farms contained a mixed flock of about 70 sheep and goats of both young and adult animals. The clinical signs observed in the affected flock were skin nodules of various sizes, 0.5-2 cm in diameter and respiratory distress. The lesions were prominent in the face, thorax and the abdomen. Morbidity was 75% while mortality was less than 5%. Skin samples identified as ILCA, 487/80 and Fashola were taken from three of the affected animals for virus isolation.

In April 1981, another outbreak of goat pox occurred amongst Sokoto Red goats and Anglo-Nubian goats at a Government Goat Improvement and Breeding Farm in Rimi, a village in Kaduna State. The flock consisted of 120 adult goats and 80 kids. The morbidity of the disease was 100% while mortality was 10 and 30% in adults and kids respectively. Skin nodules of 0.5-1.5 cm in diameter were observed all over the body; oral, nasal and ocular erosions, unsteady gait and laboured breathing were other manifestations of the disease.

The third outbreak of the disease occurred in January 1982 amongst a mixed flock of sheep and goats in Baware village in Sokoto State. Twenty five percent of the total flock of 200 was affected. Mortality was 10%. Ten pregnant goats...
aborted, possibly as a result of the disease. The clinical signs were similar to the previous cases. The disease affected only the goats.

**Virus Isolation**
Ten per cent suspension (w/v) of the skin nodule biopsies from the naturally and experimentally infected goats were inoculated onto confluent foetal kidney (FGK) cell cultures. Cultures were incubated at 37°C and observed for cytopathic effect (CPE). If no CPE was evident after 7 days, the monolayers were subcultured.

**Transmission Trials**
Three goat kids Nos. 7107, 9995 and 9805 were scarified in the neck with 2 ml of cell culture virus, GPV (ILCA) having a titre of log10 4.8 TCID50/ml.
Three other goat kids Nos. 9010, 1961 and 980 were inoculated intravenously with 2 ml of cell culture virus, GPV 4890 having a titre of log10 4.4 TCID50/ml. Daily temperatures were recorded for 10 days post inoculation.

**Agar Gel Diffusion Precipitation Test**
A known positive sheep pox antigen was tested against known positive and negative sera and several field and experimental samples of goat pox sera in a 1% agar (Oxoid) gel. The petri dishes were kept in a humidified atmosphere for 48 hours at room temperature and then examined against transmitted light for precipitation lines.

**Virus Neutralisation Tests**
One Kenyan goat pox antiserum G.202 and another Kenyan goat-sheep pox hyperimmune serum, KSG/globulin and two Nigerian convalescent immune goat pox antisera, G.99 and G.4607 were each diluted two-fold in phosphate buffered saline and mixed with equal volumes each of 100 TCID50/ml of goat pox virus, GPV 4890 and GPV (ILCA). The mixture was inoculated onto confluent FGK cell cultures and again incubated at 37°C. The cultures were examined daily for 1 week.

**Histology**
Skin, lung and lymph node samples taken from G.4890 and G.9010 and preserved in 10% formol-saline were processed for histology and stained with H&E.

**Results**
The FGK cultures inoculated with skin sample G.4890. G. ILCA, G.9010 showed CPE typical of poxvirus infection 4-6 days post inoculation. Some of the cells were refractile, others formed spindles and later became pyknotic and rounded. Many cells showing CPE contained eosinophilic inclusion bodies in their cytoplasm. No virus was isolated from G.487/80, G.4696 and G. Fashola after three blind passages.
All the goats inoculated with G.ILCA, G.7107, 9995 virus isolates showed papules about 0.5 cm in diameter at the sites of inoculation. In addition, G.7107 had vesicles. All the three goats developed severe diarrhoea, coughing and nasal discharge and died 10-14 days post inoculation. The post mortem showed haemorrhagic enteritis possibly caused by strongyle and coccidia parasites which were present. Generalised skin nodules were not observed.

The goats inoculated with the G.4890 virus isolate had thermal reactions 1 week post-infection. All three goats showed nodules at the site of intravenous inoculation. The clinical signs were ocular and nasal discharge and closure of the eye-lids. Generalised skin reactions were not observed. All the three goats were killed in extremis on day 13 post infection. Nodules, 0.5 to 1 cm in diameter were observed in the lungs.

The lungs were haemorrhagic and emphysematous. There was oedema of the alveoli. Hyperplasia of the bronchiolar epithelium, moderate cellular infiltration of neutrophils and monocytes were also observed.

The serum samples of three goats inoculated intravenously with the cell culture virus and 22 other sera taken from the natural disease were all positive in the agar gel test. The Nigerian antisera neutralised the two goat pox virus isolates while one Kenyan antiserum against goat pox failed to neutralise any of the Nigerian viruses (Table 1).
Table 1: Serum Neutralisation (SN) Titres of Goat Pox Virus Isolates

<table>
<thead>
<tr>
<th>Virus</th>
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<th>G.99</th>
<th>G.4607</th>
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<td>1:320</td>
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<td>1:320</td>
<td>1:320</td>
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<tr>
<td>GPV (ILCA)</td>
<td>1:160</td>
<td>1:10</td>
<td>1:320</td>
<td>1:640</td>
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</table>

Discussion

It is believed that goat pox is present in Nigeria but confirmatory evidence of the disease by virus isolation was not shown until this study. The disease is probably endemic in this country.

The cytopathic effect of GPV 4890 isolate had semblance to those of sheep pox and lumpy skin disease viruses that have been isolated in this laboratory (8,6). The GPV (ILCA) isolate differed from that of GPV 4890 in both cell culture characteristics and transmission experiments. The cytopathic effect with the former isolate tended to be localised rather than the generalised one produced by the latter.

The Nigerian convalescent goat pox antisera and the Kenyan goat/sheep pox globulin neutralised the two isolates of the virus, indicating some relationship between the Nigerian and Kenyan viruses. However, the failure of one Kenyan goat pox antiserum to do so could be due to strain differences between the Nigerian and Kenyan strains of the virus.

The incidence of the disease and the economic loss sustained are not known. The isolation of the virus in cell culture has proved of advantage since one of the isolates is being attenuated in foetal lamb kidney cell culture with a view to producing a homologous vaccine to control the disease.

Acknowledgements

The authors are grateful to the Director, National Veterinary Research Institute, Vom for permission to publish this paper and to the Director, Veterinary Research Laboratory, P.O. Kabete, Kenya, for supplying the antisera.

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Influence of Ration Supplementation with Zinc on Newcastle Disease Vaccination in Cockerels

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Influence d’un Supplement de Zinc dans la Ration sur la Vaccination Contre la Maladie de Newcastle Chez des Jeunes Coqs

Résumé
Des jeunes coqs Harco (m) âgés d’un jour ont été répartis en trois groupes A, B et C qui ont été alors soumis à des rations enrichies de zinc, à raison de 50,200 et 400 mg Zn/kg de nourriture pour les trois groupes respectifs. Tous les groupes ont subi la vaccination contre la maladie de Newcastle (ND) effectuée régulièrement au Nigéria et la réaction immunologique subséquente a été mesurée grace aux valeurs de l’inhibition/hémagglutination (IH).
Les jeunes coqs nourris de rations plus riches en zinc donnaient des valeurs IH plus élevées. Le rapport entre le taux de zinc et les valeurs IH semble être relatif puisque les chiffres obtenus pour le groupe C étaient inférieurs à ceux du groupe B. Selon les résultats, les poulets vaccinés sont mieux protégés contre la maladie de Newcastle si leurs régimes sont enrichis de zinc.

Summary
Harco® cockerels at day-old were divided into three groups: A, B and C which were then placed on rations supplemented with zinc to the levels of 50, 200 and 400 mg Zn/kg of feed for the three groups respectively. All groups were subjected to the routine Newcastle Disease (ND) vaccination programme in Nigeria, and the post-vaccination immunological response measured using the haemagglutination-inhibition (HI) values.

Cockerels on rations with higher levels of zinc gave higher HI values. The positive relationship between zinc level and HI values seems to be limited as lower values were obtained from group C as compared with group B. The results suggest that vaccinated chickens can be better protected from Newcastle disease when their diets have increased zinc values.

Introduction
Newcastle disease was first diagnosed in Nigeria in 1952 (1), and ever since there have been attempts to control the disease through a vaccination programme based on initial intraocular vaccination using the B1-strain of the virus within the first 2 weeks of life of chickens followed with a booster by the sixth week of life using the Komarov strain intramuscularly. Popular as this vaccination programme is within the country, Newcastle disease continues to be the most serious disease problem facing the poultry industry in Nigeria. Most of the outbreaks have been reported in supposedly vaccinated flocks. However, serological studies have revealed that chickens do not maintain antibody titres for long periods following vaccination (in print). While this may be understandable for the initial vaccination with the B1-strain of the virus which is a lentogenic strain (2), the response from the booster with the Komarov is believed to be far short of expectation. Hence various attempts are presently being made to help chickens maintain protective levels of antibody titres for a considerable period, post-vaccination. Recently, the role of zinc in the maintenance of immunological responses has started to attract attention as reported from previous studies with pigs (3) and rats (4).

It is, therefore, here, intended to see how this quality of zinc can be exploited to achieve effective Newcastle disease vaccination in chicken.

Materials and Methods
This experiment was carried out using 160 day-old cockerels. Ten chicks of this number were bled to death at day-old for
serum collection while those remaining were divided into three groups: A, B and C, each containing 50 chicks. Group A birds were fed a commercial ration containing final zinc level of 50 p.p.m. feed, as was determined by atomic absorption spectroscopy similar to the procedure described by (5). Rations for groups B and C were supplemented to final zinc levels of 200 p.p.m. and 400 p.p.m. feed respectively.

All chicks in the three groups were vaccinated at the age of 4 days against Newcastle disease (ND) with the B-strain vaccine intraocularly. This was followed with the booster at the age of 6 weeks using the Komarov strain vaccine intramuscularly. At the age of 2 weeks, 10 chicks were randomly selected from group A and bled for serological analysis. Groups B and C were similarly treated. Serological analysis was repeated at 2 week intervals thereafter from randomly selected members of the groups until the age of 20 weeks when the experiment was terminated.

Serological response was evaluated by the measurement of the haemagglutination-inhibition (HI) antibody. The HI test employed was a modification of that described in Isolation and Identification of Avian Pathogens using the beta-procedure, two-fold dilution of serum and four units of haemagglutinin. Sera were assayed for the individual birds in the different groups and the geometrical mean haemagglutination-inhibition (GM-HI) titres for the different groups calculated.

### Results

All the cockerels in the three groups were in good body condition throughout the period of the experiments. There were no clinical signs of ill-health in any groups. There was no significant difference in the weight of birds from the different groups throughout the course of study.

The HI titre range for the different groups is as presented in Table 1. Values are for randomly selected members of each group at the corresponding ages.

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in Table 2 and expressed as log base 2 (log2).

The comparative antibody fluctuation for the three groups is as shown in Figure 1. The HI titre for chicks bled at day-old was found to range from 2 to 4 and this we consider to be suggestive of maternal immunity which is considered to be very low, hence the vaccination at the age of 4 days.

The initial intraocular vaccination increased the HI values to a peak at the fourth week of life in the three groups: 64(A), 128(B) and 128(C). These values do not appear to have been highly influenced by the zinc supplement in the rations, hence no significant difference in values. The booster vaccination at the sixth week elevated the HI-titres significantly and variously in the three groups with the peak of values achieved by the eighth week of life of the cockerels.
Zinc Supplementation and Newcastle Disease Vaccination in Cockerels

Figure 1: The haemagglutination — inhibition (HI) antibody response (geometric mean) in chickens vaccinated for Newcastle disease from zinc supplemented (B and C) and non-supplemented (A) flocks.

512(A), 2048(B) and 1024(C). Thereafter, there was a progressive decline in values until the age of 20 weeks when the experiment was terminated and values stood at 64(A), 256(B) and 256(C).

Discussion
Zinc is a relatively non-toxic, cheap but nutritionally essential trace element.

While clinical zinc deficiency in the past had been associated with hypogonadism, growth retardation, delayed wound healing, etc., recent studies have been directed to the role of zinc in immunocompetence. Detailed reports are available for pigs (3) and rats (6). Studies in poultry have not been carried out to our knowledge.

Marginal deficiency of zinc in poultry is likely to be common since the bulk of poultry ration is composed of cereals, which apart from being low in zinc is known to be high in fibre which has a binding effect on zinc in the gastro-intestinal tract, thereby reducing the bioavailability of zinc.

Fifty mg zinc per kg ration fed to birds in group A is about the recommended optimum (7). The 200 mg/kg ration for group B is about the maximum zinc level of rations permitted in U.K. diets, while the 400 mg/kg ration for group C is intended to create a supposed 'toxic level'.

From the results, the second vaccination at the sixth week substantially increased the HI value of the chicks; hence by the eighth week of age, the GM-HI values for the three groups were: 512(A), 2048(B) and 1048(C). The significant difference between values for groups A and B can only be attributed to the effect of the zinc supplement. The explanation for this difference in values may not be easy. However, zinc is known as an important component of metalloenzymes like thymidine kinase (8) and DNA polymerase (9). Both enzymes are important in protein synthesis. Zinc deficiency has also been known to cause decreased immunoglobulin production differentially (10) and this is probably due to the adverse effect of zinc on replicating T-helper cells.

It can thus be concluded that while clinical zinc deficiency in poultry may not be common with the level of zinc in most poultry rations, the fact that significantly higher HI values were obtained from birds on zinc supplemented ration indicates a positive effect. Since results of HI test are usually in agreement with challenge tests (11), it only shows that such birds are better protected against clinical disease. However, since HI values obtained from group B happen to be higher than those for group C (with a higher ration zinc level) it means that further studies should be carried out to standardise this relationship and draw an upper limit for best result; more so, since it has been reported that very low or very high concentrations of zinc may impair the host's resistance (12).

Acknowledgement
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References


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Concentrate to Roughage Ratio Requirement by Maradi Goats

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Department of Animal Science, Faculty of Agriculture, Ahmadu Bello University, Zaria, Nigeria

Besoin de Ration de Concentré et de Fourrage Grossier pour les Chèvres Maradi

Résumé
Dans une étude de la croissance qui a duré 60 jours, 15 boucs Maradi pesant en moyenne 20 kg et âgés d'environ 3 ans étaient nourris de différentes quantités de foin Gamba (Andropogon gayanus) et de concentré. Les boucs ont été répartis en trois groupes et soumis à trois régimes qui consistaient en concentré et en fourrage grossier dans les proportions suivantes: 60/40, 40/60 et 20/80. Bien que l'on n'ait pas observé de différences significatives quant à l'effet des traitements, les boucs ayant bénéficié d'un régime de 60/40 de concentré/fourrage grossier avaient des gains de poids relativement plus grands.

Summary
In a growth study lasting 60 days, 15 male Maradi goats weighing on average 20kg and about 3 years old were fed varying levels of Gamba hay (Andropogon gayanus) and concentrate. The goats were divided into three treatment groups. The treatments were concentrate to roughage ratio 60:40; 40:60 and 20:80. Although no significant differences in performance between treatments were shown, goats on the 60:40 concentrate to roughage ratio had relatively higher weight gains.

Introduction
Maradi goats commonly known as Red Sokoto are one of the defined breeds of goats in Africa. This breed has a uniform dark red coat and it is found mainly in the northern part of Nigeria. It is very famous for its Morocco leather in the world market. It stands about 65cm at the withers with mature weight varying between 20 and 30kg.

Goat production has not received much attention as compared to cattle and sheep. There is a need to investigate the nutritional requirements of Maradi goats to enhance the production of good quality leathers. The concentrate to roughage ratio requirements of Maradi goats was studied to provide necessary nutritional data relevant to improvement in the production of this breed of goat. (1) reported that from a practical feeding standpoint, increasing the ratio of concentrate to roughage has its greatest significance as a means by which the total digestible nutrients or energy intake of a growing animal may be increased. (2) reported that replacing a part of the concentrate ration with an equal weight of roughage would result in a reduced energy content of the ration and the rate at which such animals grow.

(3) In their investigation on the effect of concentrate to roughage ratios on the performance of steers, reported that diminishing returns operate with higher proportions of concentrate in the diet; in terms of rate of gain, carcass quality and carcass yield. Using bullocks on a diet with concentrate to roughage ratios 40:60, 60:40, 80:20, 97:3, daily gains, carcass quality and yield slaughter percentage were found to be nearly equal for 60:40, 80:20, 97:3 ratios but higher on the 40:60 level.

Materials and Methods
The goats used for the study were male goats and weighed on average 22 kg. They were pen-fed and watered ad lib with buckets. The goats were fed Gamba hay (Table 2) and a concentrate mixture containing 12% crude protein (Tables 1,
Table 1: Concentrate Composition

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Composition %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maize</td>
<td>90.6</td>
</tr>
<tr>
<td>Cotton seed cake</td>
<td>5.7</td>
</tr>
<tr>
<td>Salt</td>
<td>2.0</td>
</tr>
<tr>
<td>Bone meal</td>
<td>1.5</td>
</tr>
<tr>
<td>Vitamin-mineral premix*</td>
<td>0.2</td>
</tr>
<tr>
<td>Total</td>
<td>100.0</td>
</tr>
</tbody>
</table>

*Vitamin-mineral premix used is ZOODRY VM 702 and contains per kg: vit. A 10,000,000 i.u.; vit. D₃ 11,000,000 i.u.; vit. E 10,000mg; magnesium (Mg) 100,000mg; manganese (Mn) 50,000mg; iron (Fe) 80,000mg; zinc (Zn) 45,000mg; copper (Cu) 6,000mg; iodine (I) 2,500mg; cobalt (Co) 800mg.

2). A completely randomised design was used in the trial and the animals were randomly allocated to the three treatments. The Gamba hay to concentrate ratios were 60:40, 40:60 and 20:80. The hay and concentrate fed was increased in quantity according to the consumption by the animals keeping the ratio constant. The amount fed and refused were weighed daily to determine daily intake per head. Goats were weighed once a fortnight.

Table 2: Composition of Concentrate and Hay (DM basis)

<table>
<thead>
<tr>
<th></th>
<th>Hay (g)</th>
<th>Concentrate (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude protein (CP)</td>
<td>4.5</td>
<td>13.8</td>
</tr>
<tr>
<td>Ash</td>
<td>9.8</td>
<td>4.4</td>
</tr>
<tr>
<td>Ether extract (EE)</td>
<td>2.5</td>
<td>5.0</td>
</tr>
<tr>
<td>Crude fibre (CF)</td>
<td>42.6</td>
<td>9.1</td>
</tr>
<tr>
<td>Nitrogen free extract</td>
<td>40.6</td>
<td>67.7</td>
</tr>
</tbody>
</table>

Table 3: Performance of Animals

<table>
<thead>
<tr>
<th>Treatments</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>SE*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Concentrate roughage</td>
<td>60:40</td>
<td>40:60</td>
<td>30:80</td>
<td></td>
</tr>
<tr>
<td>No. of animals</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td></td>
</tr>
<tr>
<td>Initial weight (kg)</td>
<td>22.4</td>
<td>22.4</td>
<td>22.7</td>
<td></td>
</tr>
<tr>
<td>Final weight (kg)</td>
<td>24.5</td>
<td>23.7</td>
<td>22.9</td>
<td></td>
</tr>
<tr>
<td>Daily gain (g)</td>
<td>36.2</td>
<td>13.8</td>
<td>3.4</td>
<td>11.95</td>
</tr>
<tr>
<td>Hay intake (g) (DM)</td>
<td>200.4</td>
<td>360.4</td>
<td>353.0</td>
<td></td>
</tr>
<tr>
<td>Concentrate intake (g) (DM)</td>
<td>412.6</td>
<td>275.1</td>
<td>130.0</td>
<td></td>
</tr>
<tr>
<td>Total dry matter intake (g)</td>
<td>613.0</td>
<td>635.6</td>
<td>483.0</td>
<td></td>
</tr>
<tr>
<td>Feed conversion (feed/gain)</td>
<td>16.9</td>
<td>46.1</td>
<td>14.2</td>
<td></td>
</tr>
</tbody>
</table>

*Standard error
Means in the same row bearing different superscripts differ (P < 0.05).

Concentrate Consumption
A significant difference (P<0.05) exists between animals in treatment 3 and 1 (Table 3). The animals on treatment 1 had an average concentrate intake of 412.6g per day while those on treatment 3 consumed 130.0g daily.

Mean Daily Weight Gain
There was no significant (P>0.05) difference in mean daily weight gain among the three treatments (Table 3). The animals on treatment 1 performed relatively better with an average daily gain (ADG) of 36.2g while the ADG on treatments 2 and 3 was 13.8g and 3.4g respectively.

Discussion
Animals on treatment 1 (60:40 ratio) consumed the least amount of hay because they were fed the highest level of concentrate. This observation is in line with the findings of (4) who reported that giving of concentrate feed resulted in a drop in the voluntary intake of fodder: (5) also concluded from their trials with mountain ewes that hay intake declined with increasing concentrate level.

The average daily gain (ADG) was lowest for treatment 3 (20:80 ratio). This may be due to the lower energy level of
the ration, because of the high level of roughage. (2) reported that replacing a part of concentrate ration with an equal weight of roughage resulted in reduced energy content of the ration, which affected the rate at which such animals grew. The energy supplied to the animals did not meet the maintenance requirements for body functions, not to mention production. Therefore these animals must use body reserves to meet the basal metabolic maintenance requirements. This is common during the long dry season of about 8 months a year in northern Nigeria. The goats do not get adequate feed and consequently lose weight.

Feed efficiency was best on treatment 1 and poorest on treatment 3. The results indicate that treatment 1 (60:40 concentrate to roughage ratio) gave a superior or the best feed conversion efficiency ratio. This finding is in line with the results of (6) who reported that lambs fed 60:40 concentrate to roughage ratio had a more efficient feed conversion than lambs fed the high roughage ration. (7) reported the superior performance of growing Dutch Friesian bulls on 60:40 concentrate to roughage ratio.

References


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Incidence of Dicrocoeliasis in Cattle Slaughtered in Lagos Metropolis, Nigeria

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Incidence de la Dicrocoellose Chez les Bovins Abattus dans la Métropole de Lagos (Nigéria)

Résumé

1,052 vésicules biliaires de bovins abattus dans la métropole de Lagos ont été examinées entre septembre 1980 et août 1982. Le taux d’infection de *D. houps* était de 18.5%. Les infections étaient plus fréquentes entre juin et août qui sont la période des pluies dans les zones pastorales du Nord qui approvisionnent en grande partie Lagos en bovins. Il est probable que les infections soient contractées pendant la saison sèche entre janvier et mai, lorsque les animaux sont concentrés dans les micro-habitats autour des abreuvoirs.

Summary

1,052 gall bladders of cattle slaughtered in Lagos metropolis were examined between September 1980 and August 1982. *D. houps* infection rate was 18.5%. Infections were highest between June and August which were the rainy months in the cattle-rearing areas of the North from where most of Lagos cattle supplies come. Infections are likely to be acquired in the dry season between January and May when animals are concentrated in microhabitats around water holes.

Introduction

*Dicrocoelium houps* (Loss 1907) has been reported from many parts of West Africa in the bile ducts of cattle. It has been reported in the Republic of Chad (1); Ghana (2); Sierra Leone (3) and Nigeria (4). Cases of spurious human infections have also been reported in Ghana by (5).

Although it is known that dicrocoeliasis occurs in Nigeria, there has been no report of its incidence in Lagos metropolis, one of the largest cattle markets in Nigeria. Since nearly all cattle slaughtered daily in Lagos are brought directly from the drier northern parts of Nigeria and neighbouring countries, by rail or trucks and rarely on the hoof, it is hoped that the present study will give further information on the life cycle of the trematode in the areas where infections are acquired.

Materials and Methods

Gall bladders of cattle were collected once a week from Apapa Road abattoir, Lagos within the period of September 1980 to August 1982. The bladders were cut open in the laboratory and the contents emptied into beakers. The trematodes were sieved out and counted while the bile content was allowed to settle for 2-3 hours and the sediments were examined under the microscope for trematode ova. A total of 1,052 gall bladders were examined during the period.

Results

The adult trematodes found were 954 *D. houps*, 78 *Fasciola giantica* and 3 unpaired male *Schistosoma bovis*. Average occurrence of infection based on the presence of eggs or flukes was 13.9% for *D. houps* only, 4.4% for double infections of *D. houps* and *F. giantica* while 0.2% had both *D. houps* and *S. bovis*. The grand total rate of *D. houps* infection was 18.5%.

The variations which occur in the average monthly rates of infection

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together with the rainfall pattern at Maiduguri, a major cattle rearing area from where a substantial proportion of Lagos supplies come are shown in Figure 1. Highest infections of *D. hospes* occurred between June and August which are the months of heavy rainfall. September, October and November had moderately high rates of infection ranging from 15 to 23%. Low rates however occurred in the very dry months between January and May with a range of 2.9-14%.

**Discussion**

The average *D. hospes* infection rate of 18.5% recorded in this study is much lower than 58.8% reported from Ghana (2), 35.75% from Tanzania (6) and 80.6% from Uganda (7). It is however higher than the 4% infection rate reported by (3) from Sierra Leone.

There is no properly documented work on all stages of the life cycle of *D. hospes*. Previous investigations have dealt more with locating the possible snail intermediate hosts which were reported as *Limicolaria* and *Achatina* in Ghana (2). (8) identified three different species of *Limicolaria* which were naturally infected in Togo, while attempts to experimentally infect both *Limicolaria* and *Achatina* by (9) failed. There has been no report of the development of the trematode from metacercariae to sexually matured adults which is known to take 6-7 weeks in *D. dendriticum* while eggs appear in faeces about 4 weeks later (10).

Although it is evident from Figure 1 that infection was most prevalent in the rainy season and lowest in the dry months of January to May, it is probable that *D. hospes* infections are acquired in the dry months when food and water are scarce.
in the cattle rearing zones. Thus, concentration of cattle around water holes and surrounding microhabitats is suitable for the intermediate hosts. Infections contracted during this period will mature in the rainy season when grass and water are abundant for wide range grazing.

References


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ABSTRACTS

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2. Salmonella Serotypes Isolated in Animals in Senegal.
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4. Urease Activity of Brucella Species.

II. VIRAL DISEASES
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25. Control of Paratuberculosis (Johnne's Disease) in Goats by Vaccination.
1 IBAR/1985 D.H. Burrell

Immunisation of Sheep Against Experimental Pseudomonas aeruginosa Dermatitis and Fleece-rot Associated Body Strike


AUTHOR’S SUMMARY: Sheep immunised with an experimental Pseudomonas aeruginosa vaccine and unvaccinated control sheep were challenged by induction of experimental dermatitis with the homologous strain. All of six control sheep developed ulcerative dermatitis, and two of the six challenge sites were struck by larvae of Lucilia cuprina. Neither severe dermatitis nor strike occurred in six vaccinated sheep. These results were confirmed in an experimental challenge using three different serotypes of P. aeruginosa on each of 3 vaccinated and control sheep, although fly-strikes did not occur. In a field trial of the same vaccine, none of 26 vaccinated sheep developed severe, exudative fleece-rot lesions nor were any fly-struck, whereas 61 of 115 control sheep developed severe, exudative fleece-rot lesions, 21 of these becoming struck by L. cuprina. The isolates of P. aeruginosa recovered from the field challenge experiment were a different serotype to that used in the vaccine.

One cow was given a commercial multicomponent clostridial sheep vaccine in two successive pregnancies and the second cow in one pregnancy. The first cow produced a low concentration of epsilon antitoxin (Clostridium perfringens type D) in its blood and colostrum after the first course of three injections of vaccine. A higher concentration was produced by cow 2 after a course of six injections and by cow 1 after a further course of four injections in its next pregnancy. Two hundred ml of colostrum from cow 1 (after the second course of vaccine) was given to 12 newborn colostrum-deprived lambs. All showed a high concentration of antitoxin 48 hours later. The lambs were actively immunised by injections of the same clostridial vaccine at 3 and 9 weeks or 6 and 12 weeks old and all produced sufficient antitoxin to protect up to slaughter at 24 weeks. It is concluded that colostrum from cows vaccinated with sheep clostridial antigens can be fed to protect lambs passively.

2 IBAR/1985 M.P. Doutre and Y. Buisson

Salmonella Serotypes Isolated in Animals in Senegal


AUTHORS’ SUMMARY: An exhaustive list of Salmonella serotypes isolated in animals in Senegal for the last 30 years is presented. 184 serotypes are enumerated, among them, 115 have been demonstrated at least once in man in local hospitals. 21 new serotypes have been discovered. The question of rare and slightly or non pathogenic serotypes is discussed.

3 IBAR/1985 M.J. Clarkson, W.B. Faull and J.B. Kerry

Vaccination of Cows with Clostridial Antigens and Passive Transfer of Clostridial Antibodies from Bovine Colostrum to Lambs

Vet. Rec. (1985), 116 (17), 467

AUTHORS’ SUMMARY: Two Friesian cows were used to attempt to produce colostrum containing a high concentration of clostridial antibodies which could be fed to newborn lambs in order to passively transfer immunity to diseases caused by clostridia.

4 IBAR/1985 M.J. Corbel and D. McI. F.D. Hendry

Urease Activity of Brucella Species


AUTHORS’ SUMMARY: Examination of the urease activity of 604 brucella strains showed a limited correlation with species. Most strains of B. canis, B. neotomae and B. suis gave a positive urease reaction within 15 minutes, although some exceptions were noted. A substantial proportion of strains of B. abortus and B. melitensis also hydrolysed urea as rapidly as most B. suis strains. Although most B. ovis strains were negative to the urease test, 28.9% of those examined gave positive reactions.

5 IBAR/1985 B.M. Freeman and A.C.C. Manning

Failure to Induce Stress Reactions Following Vaccination Against Marek’s Disease or Newcastle Disease.


AUTHORS’ SUMMARY: Chickens aged 1 or 21 days were given single injections of a vaccine against Marek’s disease or Newcastle disease respectively, and monitored over a 3 week period for any signs of a stressor response. No consistent evidence of such a response was found. Some data on normal ontogenetic changes in various adrenal and plasma variables are also presented.


Pathology of Natural Rotavirus Infection in Clinically Normal Calves
AUTHORS' SUMMARY: During a longitudinal study of the epidemiology of rotavirus infection in a calf rearing unit, excretion of virus in faeces was detected by enzyme-linked immunosorbent assay in 40 of 48 (83%) unweaned calves aged between 3 days and 5 weeks. Fifty percent of the infected calves had no clinical signs of disease. Enterocytes containing rotavirus antigen and intestinal lesions were found in all of 12 clinically normal calves selected for necropsy between days 1 and 4 of virus excretion. Stunting and fusion of villi, exfoliation, disappearance and vacuolation of enterocytes and the presence of cuboidal enterocytes were observed in infected calves but not in rotavirus-free control calves. Lesions predominated in the upper small intestine, where rotavirus was most abundant, especially on the first two days of virus excretion. The numbers of enterocytes infected with rotavirus diminished before the lesions resolved.

7
IBAR/1985 W.P. Taylor and L.W. Rowe

A Microneutralisation Test for the Detection of Rinderpest Antibodies


AUTHORS' SUMMARY: A microplate system is described for the measurement of rinderpest neutralising antibodies. The method is as sensitive as the conventional roller tube but is more adversely affected by non-specific viral inhibitors and by cytotoxic sera. These problems were overcome by diluting the sera 1:10 prior to testing.

8

Detection of Foot-and-Mouth Disease Virus Antibody Using Counterimmuno-electrophoresis and Serum Neutralisation Test


AUTHORS' SUMMARY: A comparative investigation was made on the applicability, sensitivity and specificity of counterimmuno-electrophoresis (CIEP) for the rapid detection of antibody to foot-and-mouth disease virus in cattle sera using as reference a standard serum neutralisation test. The CIEP test was sensitive and exhibited a reasonable specificity.

9
IBAR/1985 A.F. Oggunrinade

Fasciolose Bovine au Nigéria. VI. Caractéristiques Parasitologiques des Infections sur le Terrain


RÉSUMÉ DE L'AUTEUR: L'auteur a étudié la parasitémie de 6 329 bovins naturellement infestés par Fasciola gigantica.

Il n'y avait pas de différences significatives dans la fréquence, les taux d'infection ou les caractéristiques de la saisie du foie entre les taureaux et les vaches. La majorité des bovins atteints avait moins de 50 douves chacun. En général, il existait une corrélation significative entre le taux d'infestation par les douves et la numération d'oeufs de Fasciola dans les fèces (r = 0,36; P < 0,02).
Prevalence of *Eimeria* in Faeces of Cattle in Saudi Arabia

AUTHORS’ SUMMARY: The faeces of 205 domestic cattle (*Bos taurus*) from five regions of Saudi Arabia were examined for the presence of coccidial parasites. The following species of *Eimeria* were recovered: *Eimeria auburnensis*, *E. bovis*, *E. cylindrica*, *E. ellipsoidalis*, *E. subspherica*, *E. wyomingensis* and *E. zuerni*. A total of 34.1% of the individual faecal samples were positive for the presence of coccidial oocysts. Mixed infections of two to four species were found in 15.7% of the specimens. *E. zuerni* and *E. bovis* occurred most frequently and were generally the most predominant species. The incidence of coccidiosis-infected cattle was higher in the eastern region.

Reproductive Criteria of Beef Bulls During and After Exposure to Increased Ambient Temperature

AUTHORS’ SUMMARY: Sixteen yearling Angus bulls were randomly assigned to one of two temperature-controlled chambers to determine the effects of elevated ambient temperature on body functions and semen characteristics. After 8 week adjustment at 23°C, eight heat-stressed bulls were exposed to 35 ± 1°C for 8 hours and 31 ± 1°C for 16 hours during each 24 hour period, and eight control bulls were maintained at 23 ± 1°C for 8 weeks. Then all bulls were exposed to 23°C for 8 weeks. Bulls were fed so that both control and stressed bulls gained at similar rates (0.58 kg/day). Semen was collected with an artificial vagina twice weekly before, during and after heat stress. During treatment, the respiratory rate of stressed bulls was greater (P < .001) than that of control bulls (54.2 ± 1.5 and 29.9 ± 1.5 breaths/min, respectively). Rectal temperatures were increased (P < .01) from 38.2 ± 0.1 to 38.7 ± 0.1°C and water consumption was increased by 35% in stressed bulls when compared with controls. Semen volume was not altered by treatment, but percentage of motile sperm decreased (P < .01) in stressed bulls by 2 weeks after the start of heat treatment. Sperm motility of stressed bulls returned to normal values 8 weeks after the end of heat treatment. Similarly, the percentage of aged acrosomes on sperm from stressed bulls increased (P < .01) by the second week of treatment and remained greater than that of controls throughout the stress period. Heat stress also resulted in more abnormal cells from the second week of treatment until 7-8 weeks after heat stress. These data indicate that exposure of bulls to elevated ambient temperatures results in decreased semen quality as evidenced by a reduced percentage of motile sperm, reduced sperm output and an increased percentage of abnormal and aged sperm. Approximately 8 weeks is required before semen quality returns to normal after heat stress of bulls.

The Effect of Level of Feeding on the Response of Lactating Ewes to Dietary Supplements of Fish Meal

AUTHORS’ SUMMARY: Thirty-six individually-penned ewes (mean live weight 69 kg), each suckling two lambs, were given one of three diets containing either 128 (low), 155 (medium) or 186 (high) g crude protein (CP) per kg dry matter. All diets contained (g/kg), milled hay, 570; molasses, 95; and a barley/fish meal concentrate, 330. The three protein concentrations were achieved by adjusting the proportions of barley and fish meal in the concentrate. Each diet was given at daily metabolisable energy (ME) intakes of 19, 23 and 27 MJ. Mean daily yields of milk in weeks 3-8 of lactation for ewes given the diet with the low concentration of crude protein increased from 2.32 kg at 19 MJ ME to 2.53 kg at 27 MJ. Corresponding values for the medium concentration of CP were 2.49 and 2.67 kg and for the high concentration 2.52 and 3.09 kg (P < .05 for differences between the concentration of dietary protein and level concentrations). For milk composition, interactions between the concentration of dietary protein and level of ME intake were not statistically significant but the main treatment effects were significant, with the protein concentration in milk increasing from 49.6 k/kg for ewes given the low concentration of dietary protein to 54.1 k/kg for those given the high (P < .001). Corresponding values for protein concentration in milk for the lowest and highest energy intake were 51.2 and 53.4 g/kg (P < .05). Losses of tissue protein were variable but decreased from 26 g/day for ewes given the low protein diet to 8 g/day for those given the high. In discussing the responses in milk yield to dietary protein and ME intake attention is drawn to the modifying influence of the energy contributed from body tissue.

Etude de la Production Laitière de la Brebis Djallonké en Relation Avec la Croissance des Agneaux
par le Fenbendazole Chez le Ovins en Zone Sahélienne au Sénégal.

Résumé de l'auteur: Le fenbendazole a été expérimenté en milieu rural au Sénégal sur des ovins naturellement infestés par des strongles digestifs, des ténias anoplocéphalidés et des coccidies. Il a été administré par voie orale à la dose unique de 10 mg/kg, une fois en saison sèche et une deuxième fois en saison humide, sur un lot de 200 ovins dans la région de Louga. Dans cette même région, un autre lot de 200 ovins, qui reçoit au même moment un placebo, sert de lot témoin. Les résultats de l'expérimentation sont appréciés par un suivi parasitologique et un suivi zootechnique. Le suivi parasitologique révèle l'efficacité totale du fenbendazole contre les strongles digestifs et Strongyloïdes papillosus. Le faible taux d'infestation par Moniezia ne permet pas d'aboutir à un résultat significatif. Les effets du dépasaristage se traduisent sur le plan zootechnique par une croissance pondérale améliorée et un taux de mortalité plus faible chez les animaux traités. Le fenbendazole peut être recommandé en zone tropicale sahélienne en raison de son efficacité, de son absence de toxicité et de sa facilité d'utilisation.

16 IBAR/1985 O.O. Tewe

Protein Metabolism in Growing Pigs Fed Corn or Cassava Peel Based Diets Containing Graded Protein Levels

Res. vet. Sci. (1985), 38 (3), 259

Author's summary: Sixty-four Large White cross Landrace weanling pigs were randomly allotted to eight treatments in a two by four factorial arrangement. The two dietary variables were cassava peel (0 and 40%) and crude protein (20, 15, 10 and 5%). Total serum protein concentration was significantly (P < 0.01) reduced by protein deficiency and by its interaction with cassava peel. The multiple coefficient of determination (R²) showed that protein intake was the primary factor determining changes in serum protein. R² values for cyanide intake (independent variable) on serum urea concentration increased from day 30 to 90 of the trial. Serum urea was increased on the 5% protein diets on days 60 and 90 of the trial. The R² values for cyanide and protein intake on serum urea concentration increased from day 30 to day 90 of the trial. Serum creatinine increased (P < 0.05) on the 5% protein diet on day 90 of the trial. The R² value for the effects of protein intake on serum creatinine was higher than for cyanide intake on days 30 and 90. The results confirmed the progressive and pronounced effects of long term cyanide intake on serum nitrogenous metabolites in pigs consuming between 110 and 120 ppm hydrocyanic acid, especially in diets containing 10% or less protein.

17 IBAR/1985 G. Vassillades

Essais de Traitement Anthelminthique


Essai de Traitement de la Dermatophilose Bovine à Madagascar par Injections de Spiramycine


Résumé des auteurs: Les essais du suanovil (solution injectable à base de spiramycine) et du strepnovil (solution injectable de l'association spiramycine + streptomycine) en République Démocratique de Madagascar dans le traitement de la dermatophilose bovine ont révélé l'efficacité de ces produits.

Mais à la différence de l'association bipénicilline-streptomycine, le suanovil reste efficace sur des rechutes éventuelles après un traitement avec ce même produit ou avec la bipénicilline-streptomycine à forte dose. Son utilisation peut donc être recommandée pour le traitement de la dermatophilose.


Effets de Doses Excessives d'Ocytocine sur la Contractilité Utérine Chez la Brebis

RÉSUMÉ DES AUTEURS: Une étude a été menée sur des brebis ovariectomisées traitées aux oestrogènes, afin d'évaluer objectivement les effets de doses croissantes d'ocytocine sur la contractilité de l'utérus et du cerveau. Des critères électromyographiques et électromécaniques ont été retenus. Il est apparu que pour des posologies supérieures à 2 µl pour 50 kg de poids vif, le tractus génital répondait par une contracture prolongée dont la durée était fonction de la dose utilisée. En revanche, une stimulation physiologique, immédiate et durable, de la contractilité fut obtenue pour des doses inférieures. En pratique, il semblerait donc opportun d'abaisser considérablement les posologies actuellement recommandées pour l'ocytocine.

20
IBAR/1985 A.U. Kalu and E. Haruna

Effects of Vasopressor Drugs on Number of Trypanosoma congoense in Ruminant Blood

Vet. Parasitol. (1985), 17 (4), 287

AUTHORS' SUMMARY: The effect of intravenous administration of vasopressor drugs on parasite concentration was studied in cows and sheep infected with Trypanosoma congoense. Ephedrine (USP), adrenaline (BP) and insulin increased jugular concentration of the parasite. Maximum effects were reached between 1½ and 2 hours with ephedrine and at 2½ - 3 hours with adrenaline. Increases in parasitaemia resulting from epinephrine were sustained only in sheep and adrenaline gave similar effects in both ruminants. Only epinephrine significantly increased parasitaemia within 45 min of administration. Ephedrine, hydralazine and guanethidine had no significant effects on parasitaemia. It is suggested that one of the mechanisms by which trypanocides increase jugular concentration of T. congoense may be by vasoconstriction of the capillary endothelium.

21
IBAR/1985 Yuji Takahashi, Yasumasa Kido, Masayuki Naol and El-Ichi Kokue

The Serial Biopsy Technique for Estimation of Drug Residue in Calf Kidney Using Gentamicin as a Model Drug


AUTHORS' SUMMARY: Residual levels of orally administered gentamicin (GM) in the calf kidney were serially determined using the renal biopsy technique. Twelve male calves were given GM per os 1 mg (potency)/kg of bodyweight twice a day for a week. Three calves were used for serial biopsy which was carried out at 3 hours and 3, 10 and 15 days after the final drug administration. Two calves were sacrificed to collect the kidney samples at each time mentioned above. The renal GM concentration was determined with gentamicin RIA kit with which more than 95% of GM added was recovered from the renal tissue in vitro and the concentration less than 0.05 µg (potency)/g was detectable in 20 mg of the tissue sample. There was no difference between the renal concentration of GM in the biopsied samples and the samples from the animals sacrificed immediately after biopsy. The time courses of the decrease in renal drug concentrations were monoeponential in both the serially biopsied samples and the samples from the sacrificed animals. These results suggested that the biopsy technique might have a value for prediction of the withdrawal time of drugs in the food producing animals.

22
IBAR/1985 Mahmud Hagi Mohamed, Abdulhamid Hagi Mohamed et A. Locatelli

Effets de la Privation Hydrique Prolongée sur des Paramètres Hémato-physiologiques et Hématochimiques du Dromadaire


RÉSUMÉ DES AUTEURS: Les auteurs ont étudié les effets de la privation d'eau (25 jours) sur trois dromadaires (une femelle et deux mâles), alimentés avec du foin, près de Mogadiscio (Somalie) de mars à mai 1983.

La privation d'eau cause un faible amaigrissement; parmi les paramètres hémato-physiologiques et hématochimiques examinés, seuls l'hématocrite, le cholestérol, CPK, le Na et le Cu ont montré des modifications significatives.

23
IBAR/1985 A.M. Makumyaviri, G. Ottwicz and A. Verhulst

Transport Stress and Histophysiology of the Suprarenal and Thyroid Glands of Cattle in Shaba: A Case of Cattle Transfer from Kundelungu to the Abattoir at Lubumbashi, Zaire


AUTHORS' SUMMARY: Histological and histochemical studies were carried out on the suprarenal and thyroid glands of 46 oxen slaughtered at Shaba in the Republic of Zaire. The long exhausting walk had given rise to some significant structural and functional changes in the suprarenal cortex associated with a hypersensitivity of the suprarenal cortex and a prolonged adaptive response.
Serology of Ovine Chlamydiosis by the ELISA Test. Use of a Commercial Antigen Prepared From a *Chlamydia trachomatis* Strain

AUTHORS' SUMMARY: An immuno-enzymatic technique of the ELISA type was applied to the sero-diagnosis of ovine chlamydiosis, an infection due to *Chlamydia psittaci*. The technique, although using a commercial antigen prepared from a *Chlamydia trachomatis* strain, proved to be more sensitive and more specific than the complement fixation test and at least of the value of the indirect immunofluorescence technique.

The paper makes a comparative study of the different results obtained by the three techniques on 485 ovine sera.

Control of Paratuberculosis (Johne's Disease) in Goats by Vaccination

AUTHORS' SUMMARY: After several years of unsuccessful efforts to eradicate paratuberculosis in goats in Norway by conventional methods such as general hygienic precautions and the isolation and slaughtering of clinically affected and serologically positive animals, a vaccination programme was initiated in 1967. The vaccine used consists of two live attenuated strains of *Mycobacterium paratuberculosis* suspended in a mixture of liquid paraffin, olive oil and pumice powder. The vaccine may be stored at 4°C for 2 weeks, the dose is 1 ml and the goat kids are vaccinated at the age of 2-4 weeks. The efficacy of the vaccine has been judged mainly by post mortem examination of vaccinated and unvaccinated goats in the period 1967-1982. During this period about 131,000 goats were vaccinated and, based on the post mortem examination of 15,219 goats, the infection rate was reduced from 53 to 1%. Moreover, infection occurred almost exclusively in goats which for some reason or other had not been vaccinated or which had been too old when vaccinated. The results of these examinations showed that the adjuvated vaccine with live *M. paratuberculosis* bacteria offers a high degree of protection against paratuberculosis in goats.
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Objet
Le Bulletin de la Santé et de la Production Animales en Afrique publie des articles de recherches originales sur les activités de production et de santé animales visant à développer l'industrie de l'élevage en Afrique et à mieux utiliser les ressources animales du Continent. La revue est un périodique trimestriel.

Présentation des articles
Deux exemplaires des articles doivent être adressés 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.


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 éditoriaux
— 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 par les noms des auteurs.

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