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The Blue Cross Book

for the Veterinary Profession

Hoechst Roussel Vet

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PREFACE

Mr. W. J. Bader
Managing Director
Hoechst Roussel Vet Pvt. Ltd.

Dear Sirs,

It is my pleasure to write to you once again while releasing the 10th issue of ***"The Blue Cross Book"*** .

We in Hoechst Roussel Vet, give a lot of importance to clinical research and generate field data on requests of the practicing veterinarians from the field. By doing so, we try to get as close to the field experts of the veterinary profession as possible. We see this partnership as a key to success in better animal health care.

I thank all the scientists in the veterinary profession for their interest and valuable contributions which made the release of this 10th issue of ***"The Blue Cross Book"*** possible.

I also thank all our esteemed readers for their constant support and suggestions to the Editor.

Best regards,

J. Bader

GUIDELINES TO CONTRIBUTORS

The Blue Cross Book is published biannually. The contributions to the journal are accepted in the form of invited review articles, research articles (clinical / field studies), case reports, other information pertaining to animal health and production. The decision of the Editorial Board members will be final regarding acceptance of the article for publication. The manuscript should be typed on one side of the paper with double spacing except for abstracts, footnotes and references for which single spacing be used. The style of reference citing should be stickly followed as shown below. The words to be printed in Italics should be underlined. The manuscript should be arranged in the following order :

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Name/s of author/s	:	K. Kumari, P. C. Chowdhri and P. K. Das
Place of work	:	Department of Pharmacology Bombay Veterinary College, Parel, Mumbai
Materials and Methods	:	
Results and Discussions	:	
References	:	**For Periodicals :** Name and initials of author/s, year of publication in parenthesis, abbreviated title of journal, volume number, first and last page number/s. e.g. Chhabra, D., Moghe, M. N. and Tiwari, S. K. (1996). *Ind. Vet. J.*, **82** : 1-3.
	:	**For Books :** Name/s of author, year of publication in parenthesis, title of the book, edition, name of publishers and page number/s. Radostitis, O. M., Blood, D. C. and Gray, C. C. (1994). Vetarinary Medicine, **8th Edn.**, *ELBS*, London
Tables and Figures	:	Tables are to be numbered in Roman numerals (I & II so on). each table should have a clear title. Figures should be good quality and numbered in Arabic numbers (1, 2, 3 so on).

Abstracts and sub-headings are not necessary for clinical articles and short communications. These should not exceed three typed pages. For case reports, history, observation, tentative and confirmatory diagnosis, line of treatment and follow up on the case should be given.

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In-vitro Efficacy of Cefquinome (INN) and Other Anti-infective Drugs Against Bovine Bacterial Isolates from Belgium, France, Germany, The Netherlands and The United Kingdom

A. Böttner, P. Schmid* and R. Humke
Hoechst Roussel Vet GmbH, Rheingaustraβe 190, D-65203 Wiesbaden, Germany

Introduction :

The cephalosporins and penicillins are two well known and very useful classes of anibacterials which are also classified as β-lactam antibiotics based on their common structured feature, the β-lactam ring and their common mechanism of anti-bacterial action, the inhibition of essential cell wall synthetic enzymes.

This mechanism of anti-bacterial action is particularly efficacious because the target enzymes in bacteria do not exist in human or animal cells. Thus, a major benefit of the β-lactam antibiotics is their high degree of safety in the target animal (Preston, 1992).

The newer Aminothiazolyl Cephalosporins (i.e. third generation cephalosporins) are a major advance in anti-bacterial therapy due to their broad anti-bacterial spectrum, resistance to enzymatic hydrolysis by β-lactamases and improvement of pharmacokinetic properties (Quadri *et al.*, 1993). In addition, fourth generation cephalosporins show markedly reduced affinity to β-lactamases and increased outer membrane permeability when compared to third generation cephalosporins (Hancock and Bellido, 1992).

Cefquinome is the first fourth generation cephalosporin developed for the use in veterinary medicine.

The in-vitro and in-vivo efficacy against a wide range of Gram-negative and Gram-positive bacterial pathogens and experimental bacterial infections have been recently demonstrated (Limbert *et al.*, 1991).

Here in the present report, it will be discussed about the anti-bacterial activity of Cefquinome against a large number of bovine bacterial pathogens, isolated by veterinary laboratories from all over the Europe in comparison to ten other anti-microbial drugs, commonly used in veterinary medicine.

Materials and Methods :

Bacterial strains

714 bacterial strains, associated with clinical disease outbreaks in Belgium, France, Germany, the Netherlands and the United Kingdom, were obtained from necropsy tissue-samples or appropriate diagnostic samples from various investigating laboratories in the Europe.

All pathogens were isolated, mainly between 1990 and 1993, and were identified by standard biochemical and morphological procedures (Bawlows *et al.*, 1991 and Blobel and Schlieper, 1991).

Susceptibility testing

Anti-microbial susceptibility testing was performed as a multicenter study using the

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* Hoechst AG, 65926 Frankfurt, Germany

Sceptor Pette™ System for MIC testing (Becton & Dickinson GmbH, Heidelberg). The Sceptor Pette System consists of

- the Sceptor Panel, a 84 well polystyrene plate precision dosed with appropriate amounts of dried, stabilised anti-microbials arranged to give two-fold dilutions after the addition of the inoculum,
- the Sceptor Pette™, a programmable, electronic pipetting device and
- Sceptor Gram-positive and Sceptor Gram-negative broth.

The concentration range of the antimicrobials tested is given in Table I. Preparation of the inoculum and processing of MIC testing were done according to the manufacturer's recommendations and also as per the guide lines of NCCLS (National Committee for Clinical Laboratory Standards, 1990).

An anti-microbial-free well was included on each plate as a growth control. For testing *Pasteurella multocida,* the Sceptor Gram-negative broth was supplemented with inactivated horse serum and yeast extract. Results were determined after incubation of the plate at 35° C for 18-20 hrs. The MIC was defined as the lowest concentration at which there was no growth, i.e. turbidity, as determined by the unaided eye. Four standard reference strains were tested to monitor the performance of the technique. The quality control results were within the expected ranges during this investigation.

Resistance of isolates

Strains were classified susceptible or resistant by using respective breakpoints. Since no approved standards/breakpoints exist for veterinary anti-infective drugs, breakpoints were established by considering pharmacokinetic data of each substance (Barthel and Traeder, 1992; Blobel and SchlieBer, 1991; Loscher *et al.*, 1991; NCCLS, 1990; Specht, 1986 and Enders, 1983).

Bacteria were considered resistant when the MIC for the anti-microbial substance exceeded the respective breakpoint given in Table I.

Results :

A total of 714 bacterial pathogens isolated from bovine tissue samples were tested for their in-vitro susceptibility to Cefquinome and nine other anti-infective substances commonly used in veterinary medicine. The results expressed as the maximum and minimum values of the MIC, the concentration of each anti-microbial drug that inhibited 50 per cent (MIC 50) and 90 per cent (MIC 90) as well as the percentage of resistance are summarised in Table II.

Of the 466 isolates of *Pasteurella spp.* tested, 462 (99,1%) were inhibited by Cefquinome at MIC values ranging from ≤0.06 to 2.0µg/ml, only one strain of *P. haemolytica* and three *P. multocida* strains were considered resistant (MIC 4 µg/ml).

All *Escherichia coli* (n=88) and *Salmonella* strains (n=160) tested, were susceptible to Cefquinome with MIC values below 1 µg/ml.

The remaining anti-infective substances showed a varying degree of susceptibility towards the bacterial isolates, tested in this study. The percentage of *Pasteurella multocida*, *Pasteurella haemolytica*, *Escherichia coli* and *Salmonella spp.* strains showing resistance to the anti-microbial substances tested is shown in Figures 1-4 as per the country of origin.

Table II : In-vitro efficacy of Cefquinome and 9 other anti-microbials against *Pasteurella spp.*, *Escherichia coli* and *Salmonella spp.* field isolates.

Isolate	Anti-microbial agents	MIC (µg / ml) MIC 50%	MIC 90%	Range
Pasteurella spp.	Cefquinome	<0.06	0.12	≤0.06 - 4
n = 466	Ceftiofur	<0.06	≤0.06	≤0.06 - >4
	Cephapirin	<0.25	0.5	≤0.025 - >16
	Amoxicillin	0.5	>2	≤0.06 - >2
	Amox./Clav. Ac.*	0.25	0.5	≤0.06 - >4
	Enrofloxacin	≤0.06	0.5	≤0.06 - >4
	Gentamicin	1	2	≤0.12 - >8
	Kanamycin	4	16	≤0.25 - >16
	Oxytetracycline	1	>16	≤0.25 - >16
	Tylosin	>8	>8	≤0.12 - >8
P. haemolytica	Cefquinome	≤0.06	0.12	≤0.06 - 4
n = 202	Ceftiofur	≤0.06	≤0.06	0.06 - >4
	Cephapirin	≤0.25	1	0.025 - >16
	Amoxicillin	0.25	>2	≤0.06 - >2
	Amox./Clav. Ac.*	0.25	0.5	≤0.06 - >4
	Enrofloxacin	≤0.06	>4	≤0.06 - >4
	Gentamicin	1	2	≤0.12 - >8
	Kanamycin	4	16	≤0.25 - >16
	Oxytetracycline	2	>16	≤0.25 - >16
	Tylosin	>8	>8	≤0.12 - >8
P. multocida	Cefquinome	≤0.06	0.12	≤0.06 - 4
n = 246	Ceftiofur	≤0.06	≤0.06	0.06 - 4
	Cephapirin	≤0.25	≤0.25	≤0.25 - >16
	Amoxicillin	0.5	0.5	≤0.06 - >2
	Amox./Clav. Ac.*	0.25	0.5	≤0.06 - >2
	Enrofloxacin	<0.06	0.12	≤0.06 - >4
	Gentamicin	1	2	≤0.12 - >4
	Kanamycin	4	16	≤0.25 - >16
	Oxytetracycline	1	>16	≤0.25 - >16
	Tylosin	8	>8	≤0.12 - >8
Escherichia coli	Cefquinome	0.12	0.25	≤0.06 - 1
n = 88	Ceftiofur	0.25	0.5	0.12 - >4
	Cephapirin	16	>16	4 - >16
	Amoxicillin	>2	>2	2 - >2
	Amox./Clav. Ac.*	4	>4	2 - >4
	Enrofloxacin	≤0.06	1	≤0.06 - >4
	Gentamicin	0.5	>8	≤0.12 - >8
	Kanamycin	>16	>16	0.5 - >16
	Oxytetracycline	>16	>16	1 - >16
	Tylosin	>8	>8	>8
Salmonella spp.	Cefquinome	0.12	0.5	≤0.06 - 1
n = 160	Ceftiofur	0.5	2	0.12 - > 4
	Cephapirin	4	>16	1 - >16
	Amoxicillin	2	>2	≤0.06 - >2
	Amox./Clav. Ac.*	4	>4	2 - >4
	Enrofloxacin	0.12	2	≤0.06 - >4
	Gentamicin	0.25	>8	≤0.12 - >8
	Kanamycin	2	>16	0.5 - >16
	Oxytetracycline	>16	>16	≤0.25 - >16
	Tylosin	>4	>4	0.5 - >4

* = Amoxicillin values shown

Ceftiofur, a third generation Cephphralosporin, had a range of susceptibility comparable to Cefquinome. MIC values for Enrofloxacin were in general very low but a resistance rate of 10 per cent on average was already seen. Oxytetracycline and Tylosin had little inhibitory effect and were resisted by almost all strains tested. Within the group of Aminoglycosides, Kanamycin was less active than Gentamicin. The in-vitro efficacy of Amoxicillin was favourable against *Pasteurella multocida* only; for the remaining bacterial isolates tested the level of resistance to Amoxicillin ranged from 30-100 per cent. The wide range of resistance in comparison to the mean values for some of the substances tested are due to the considerable differences between countries. (Figures:1-4)

As a result the overall level of resistance against the different anti-infectives tested was lower for Cefquinome. In comparison all other anti-infectives tested showed less effectiveness to the different pathogens.

Discussion :

An ideal anti-microbial agent is highly active against pathogens without harming the host.

The group of drugs which comes closest to this definition are the ß-lactam antibiotics. The excellent safety characteristics and the relatively braod anti-bacterial spectrum of cephalosporins has led to the chemical exploitation of the parent compound discovered by Brotzu, (1948).

The introduction of a methoxyimino-aminothiazolyl moiety in the acyl side chain of cephalosporins brought about significant enhancement of activity, extension of the anti-bacterial spectrum, espectially against Gram-negative bacteria and high resistance to inactivation by ß-lactamases (Durckheimer *et al.*, 1988 and Neu, 1983).

The introduction of polar C-3' - substituents into a cephem nucleus, improves the activity of the aminothiazolyl cephalosporins, especially against staphylococci, without loss of activity against Gram-negative bacteria. The result of this development are the fourth generation cephalosporins and Cefquinome is the first member of this group of modern compounds for the use in veterinary medicine.

Cefquinome is highly resistant to hydrolysis by plasmid encoded ß-lactamases from *E. coli, K. pneumoniae* and *P. aeruginosa* as well as chromosomal encoded ß-lactamases form *Citrobacter spp.*, *Enterobacter cloacae* and *K. oxytoca* (Limbert *et al.*, 1991).

This stability and the polarity of the compound are responsible for the excellent in-vitro activity of this compound against many Gram-negative and Gram-positive bacteria. Limbert *et al.*, (1991) found that Cefquinome has high anti-bacterial activity in vitro. *Staphylococcys aureus*, *Streptococci sp.*, *Pseudomonas aeruginosa* and members of the family Enterobacteriaceae (*Escherichia coli*, *Salmonella spp.*, *Klebsiella spp.*, *Enterobacter spp.*, *Citrobacter spp.*, and *Serratia marcescens*) were inhibited at low concentrations. Cefquinome was also active against many strains of methicillin-resistant staphylococci and enterococci. Although Limbert *et al.*, (1991) examined clinical isolates of human sources, results presented here for bovine pathogens are almost identical.

The results reported here, clearly show that Cefquinome is highly active (in-vitro) most of the pathogens tested in this study. There

was no evidence of resistance to Cefquinome in any of the isolates. For the remaining anti-infectives, tested for in-vitro activity in this study showed considerable differences of the rate of resistant development.

Results for Ceftiofur are comparable to Cefquinome apart for *Salmonella* strains from the UK where resistance was > 10% (MIC value > 2 µg/ml). MIC data obtained in this study are otherwise consistent with the results reported for Ceftiofur previously (Brown *et al.*, 1991).

For Enrofloxacin, a potent fluoroquinolone which has already been used in veterinary medicine for several years, an increase in resistance can be seen when compared to results published by Scheer, (1986). Resistance varies also considerably from country to country. An incidence of resistance in *E. Coli* and *Salmonella spp.* of over 20% is found in Germany. This fact might be related to differences between countries in the use of anti-infectives. In India, this question may not arise at this moment, since Enrofloxacin was launched a few years back only. Nevertheless, a detailed comperative study should be undertaken to assertain the status of resistance development, atleast by in-vitro methods.

Gentamicin was little resisted by *Pasteurella spp.* but resistance levels were > 25% to Kanamycin. For *E. coli* and *Salmonella spp.* resistance is again much higher to Kanamycin (up to 60%) than to Gentamicin (up to 20%) A similar resistance pattern is described by Loscher *et al.*, (1991).

Oxytetracycline exhibited a moderate

Table I : Concentrations of anti-infective substances tested and breakpoints.

Anti-infective Substances	Range (µg/ml)	Breakpoint (µg/ml)
Amoxicillin	0.06-2	2
Cefquinom	0.06-4	2
Ceftiofur	0.06-4	2
Cephapririn	0.25-16	2
Enrofloxacin	0.06-4	1
Gentamicin	0.06-8	4
Kanamycin	0.06-8	4
Oxytetracycline	0.25-16	1
Tylosin	0.12-8	1
Amoxicillin/Clavulanic acid	0.06/0.03-4/2	2/1

Figure 1 : Percentage of *Pasteurella haemolytica* strains showing resistance to Amoxicillin (AMO), Cefquinome (CEQ), Ceftiofur (CTF), Cephapirin (CAP), Enrofloxacin (ENR), Gentamicin (GEN), Kanamycin (KAN) and Oxytetracycline (OTE) by countries and for Europe (mean)

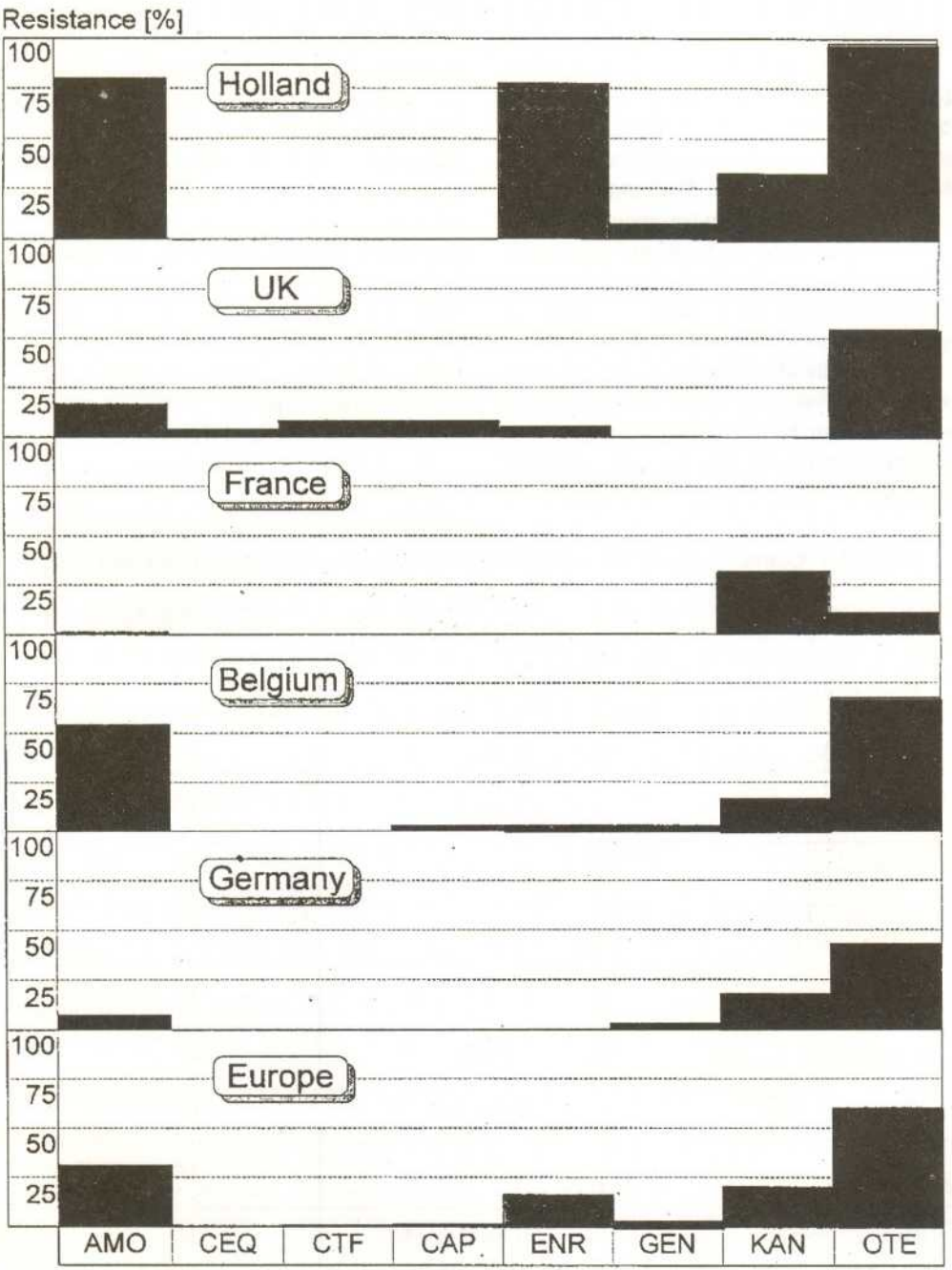


Figure 2 : Percentage of *Pasteurella multocida* strains showing resistance to Amoxicillin (AMO), Cefquinome (CEQ), Ceftiofur (CTF), Cephapirin (CAP), Enrofloxacin (ENR), Gentamicin (GEN), Kanamycin (KAN) and Oxytetracycline (OTE) by countries and for Europe (mean)

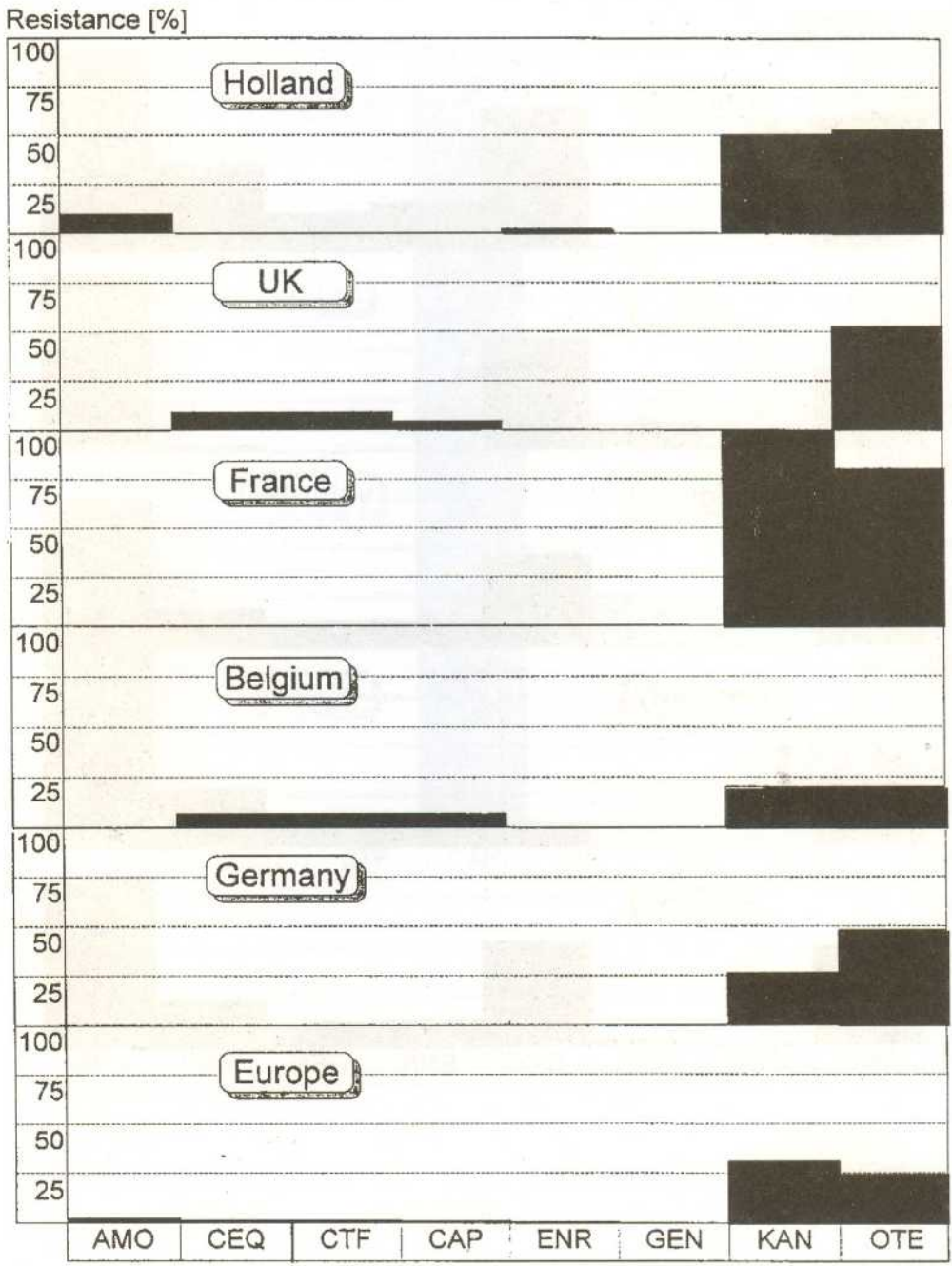


Figure 3 : Percentage of *Salmonella spp.* strains showing resistance to Amoxicillin (AMO), Cefquinome (CEQ), Ceftiofur (CTF), Cephapirin (CAP), Enrofloxacin (ENR), Gentamicin (GEN), Kanamycin (KAN) and Oxytetracycline (OTE) by countries and for Europe (mean)

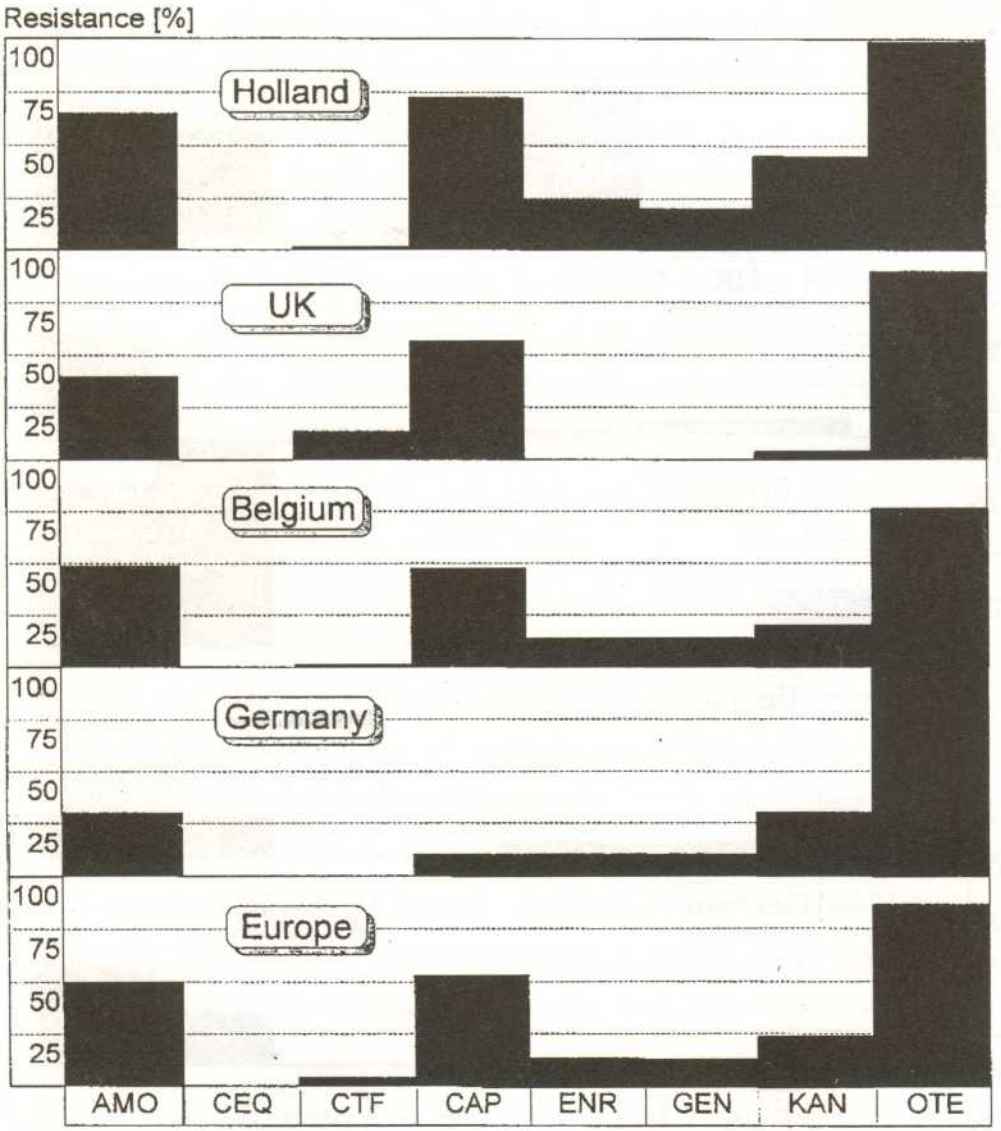
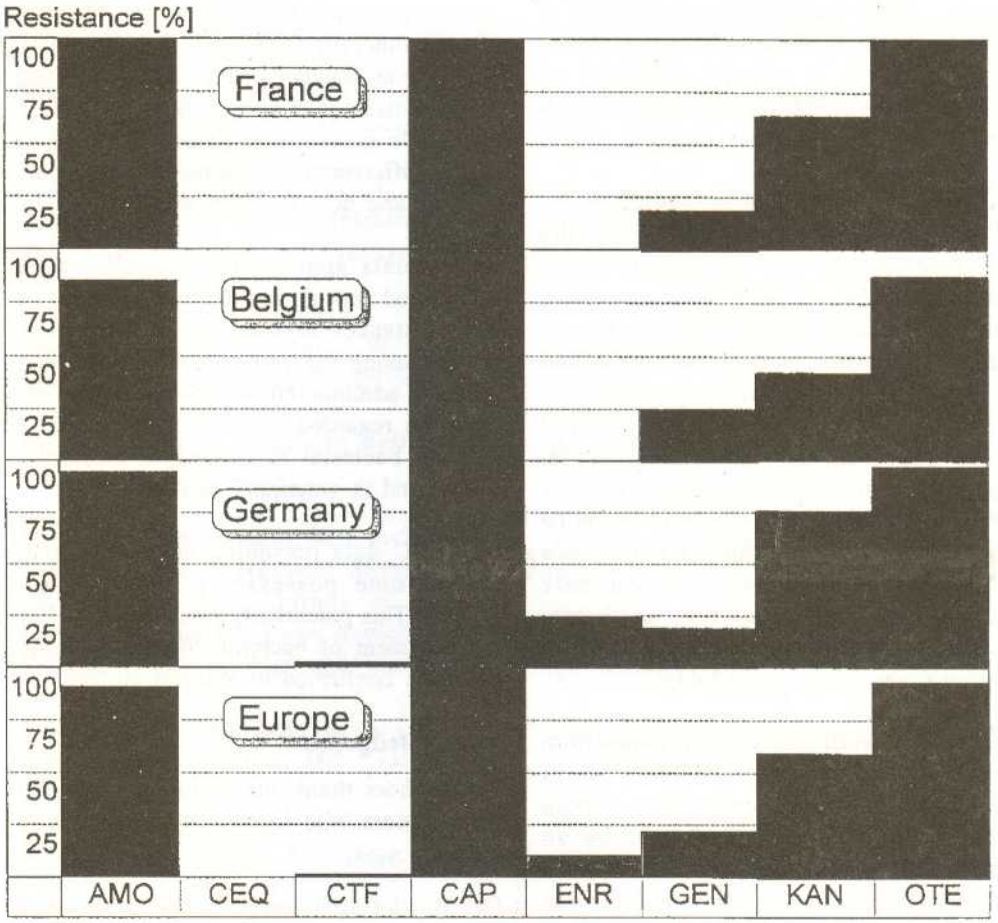


Figure 4 : Percentage of *Escherichia coli* strains showing resistance to Amoxicillin (AMO), Cefquinome (CEQ), Ceftiofur (CTF), Cephapirin (CAP), Enrofloxacin (ENR), Gentamicin (GEN), Kanamycin (KAN) and Oxytetracycline (OTE) by countries and for Europe (mean)



in-vitro profile only against *P. multocida* (level of resistance of nearly 25%). However, in all other species tested for MIC 90% data are > 16 µg/ml. Similar results were published by Grimshaw *et al.*, (1990), Specht, (1986) and Endres, (1983) and in comparison to MIC data from the earlier investigations, a steady increase in resistance to Oxytetracycline can be seen (Chang and Carter, 1976).

Tylosin was resisted almost completely by all strains tested in this study. Results published in 1986 showed slightly better MIC data for Tylosin against *P. haemolytica* and *P. multocida* (Specht, 1986), however, MIC values were already exceeding the breakpoint applied in this study (Table I).

Relatively high levels of resistance, especially with substances already used for a long time in veterinary medicine could be expected. However, a comparison with data published previously has to be carefully made since methods for antibiotic susceptibility testing and classifcation of resistance can vary considerably.

Only limited conclusions can be drawn from the country specific differences in resistance levels. Restricted use of certain anti-infective substances might be an explanation. Furthermore, it has also to be considered that country specific collection of isolates examined in this study might not be representative.

To draw conclusions from the in-vitro data to in-vivo efficacy is possible only to a limited degree (Greenwood, 1981 and Woolcock and Mutimer, 1983). MIC data have to be interpreted in the context of achievable serum concentrations, the concentration of the anti-infective drug at the site of infection and the clinical outcome after treatment with the respective drug.

As to the classification of susceptible and resistant isolates, breakpoints have been established in human medicine considering pharmacokinetics of the drug and the clinical outcome (NCCLS). For veterinary medicine no standards have been set yet and breakpoints used in human medicine are not applicable in veterinary medicine because of the differences in pharmacokinetics, the dosages and dose-regimen used.

Breakpoints applied in this study, were established considering pharmacokinetics of the substances investigated in the target species using the recommended dose and route of administration. These breakpoints could be regarded as a better reference to classify bacterial resistance to antibiotic drugs used in veterinary medicine.

The MIC data presented here show that Cefquinome possesses potent in-vitro activity. This potentiality of Cefquinome for the treatment of bacterial disases in cattle has been confirmed by clinical studies.

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Effects of Aflatoxin on Immune Response in Viral Diseases

S. M. Mohiuddin
College of Veterinary Science, Rajendranagar, Hyderabad

Introduction :

It is assumed that Aflatoxins have greater importance than other mycotoxins, since apart from their hepatotoxic effects, they are immuno-suppressive. It is well known that mouldy damaged feed ingredients which are unfit for human consumption, have been utilised in livestock and poultry feed.

Materials and Methords :

Humoral and cell-mediated immune response was studied in birds fed with Aflatoxin orally 100 µg / chick and 200 µg/ chick for 21 days from the 4th week onwards to group II and III respectively, while groups III and IV were fed with vit A 1000 IU / bird / day alongwith Aflatoxins of 100 µg and 200 µg/ birds / day respectively. Group I birds were maintained on toxin free diet. Blood samples were collected for haematological and serum samples for antibody titers and Alkaline Phosphatase activity after 14 days from groups I - V. All the chicks were vaccinated with Ranikhet disease (R2B strain) vaccine. Again on the 7th day post-vaccination booster dose of RD (R2B strain) vaccine was given. All the chickens were maintained on their respective diet for another 7 days, blood samples and serum were collected for analysis from all the groups.

Tissue phagocytic activity was studied in five cockrels, fed with Aflatoxin mixed feed at 20 ppm while the control (two cockrels) were maintained on toxin free diet for three months. Then 1 ml of saturated carbon solution was injected i/v through wing vein. All the birds were sacrificed after 48 hours, liver collected to study the cells phagocytic actively through histological parameters.

In the third experiment thirty broiller chicken of 5 weeks age were divided into three groups having ten birds in each group. Group II and III were fed with 1.0 ppm and 2.5 ppm of Aflatoxin prepared from *Aspergillus parasiticus* culture in rice, mixed in feed respectively, while group I maintained on maintained on toxin free diet for a period of three weeks. Alpha Naphthyl Estrase (ANAE) activity in blood smears and separated phagocytes was studied and Adenosine Deaminase Estrases (ADE) activity was studied in blood smears and serum.

Results and Discussion :

I. Studies on the effects of Aflatoxin on antibody synthesis against Ranikhet Disease (RD) vaccine in chicks.

In this study it has been concluded that feeding of aflatoxin @ 100 µg daily/chick for 14 days, prior to vaccination did not affect the haematological total count and differential count studies. The effect of feeding 100 µg and 200 µg of Aflatoxin daily for 14 days before R.D. vaccination was similar, but further feeding of Aflatoxin @ 200 kg for 21 days after R.D. vaccination resulted in increased serum-Alkaline phosphatase activity, increase RBCs fragility and decrease in antibodies without any change in other parameters studied. Administration of Vit. A gave protection against Aflatoxin, but failed to enhance the

immunity. Thus feeding of Aflatoxin 0.1 ppm to poultry during stress of vaccination did not interfere with any biochemical activities whereas feeding with 0.2 ppm interfered with biochemical changes and immunity production. Thus it is concluded that humoral immunity is affected at feeding 0.2 ppm to 4 weeks old chicks for a period of 21 days after vaccination.

II. Studies on phagocytic activity and heamatological changes in Aflatoxicosis in Poultry.

In this study it is concluded that the birds for Aflatoxin at 200 ppm over a period of three months showed a decrease in phagocytic activity of heterophils, kuffer cells and haemotological values. The total Basophilic and Thrombocytic counts were decreased , suggesting that Aflatoxins lowers the resistance of the body leading to increase the incidence of infectious diseases. The study suggest that cell mediated immune response is affected.

III. Enumaration of T. lymphocytes and Adenosine Deaminase Estrases activity in lymphocytes and serum in Aflatoxin fed birds.

In this study thirty broiler chicks were reared upto four weeks of age and they were divided into three groups. Group I served as control and was fed with Aflatoxin at 1.0 ppm and 2.5 ppm respective for three weeks. All these birds were sacrificed, blood smears, lymphocytes from peripheral blood and serum were collected for analysis.

Quantitative estimation of T. lymphocytes in the blood smear was carried out using Alpha Naphthyl Acetate Estrase (ANAE) staining method which give one or two reddish brown reaction productions in the cytoplasm.

The ANAE positive cell count (T.lymphocytes) in the blood in control group is 36.0 ± 4.34 while in group II is 26.0 ± 1.50 and in group III is 24.1 ± 1.22. There is a statistically significant decrease ($p<0.01$) in T. lymphocytes in group II and III.

Alpha Naphthyl Acetate Estrase (ANAE) staining has been shown to be a reliable cytochemical marker for identification of Monoeytes and T. lymphocytes in peripheral blood (Muller *et. al.*, 1979). Aflatoxin feeding in the two groups resulted in a significant decrease of T. lymphocytes thus affecting the cell mediated immune response. This confirms the lymphopenia noticed in aflatoxicosis by Campbell and Doerr, (1981).

Adenosine Deaminase Estrases (ADE) is separate lymphocytes and serum.

The DNA is one of the enzymes of purine salvage pathways. Of late the role of purine metabolism in the immune response has gained importance.

The need of proliferating cells for a balanced supply of both purine and pyramidine compounds for maintaining cell division and growth has long been known. But the importance of pathways of purine inter- conversion in maintaining the integrity of the immune system has been revealed by the striking impairment of its function found in association with deficiencies of specific enzyme of purine metabolism. The first enzyme defect to be found in association with severe immuno-deficiency disease is ADE (Giblet *et al.*, (1972). The lymphocytic ADE could severe as a marker of cell mediated immunity.

The lymphocyte ADE activity in group I was 50.28 ± 9.5, group II 49.25 ± 18.24 and group III 20.78 ± 10.5 nanomoles of ammonia liberated/million cells/ hour. There was a significant decrease ($P<0.01$) in the lymphocytic ADE in group II and III hence convincingly be concluded that the inhabitation of proliferation of T-cell is by Aflatoxins.

The serum ADE activity did not show any significant differences amongst the groups I, II and III. The activity in group I is 6.60 ± 2.04, group II 5.64 ± 2.05 and group III is 4.84 ± 1.65 1.U/ml. serum.

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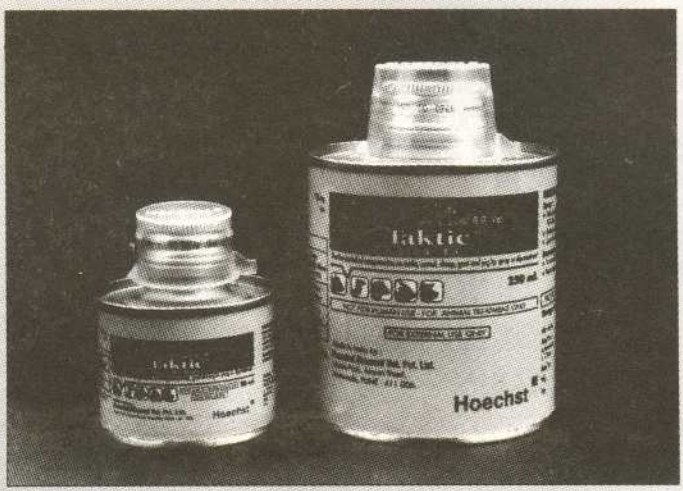

Application : Taktic is recommended by spray or dip treatment.

Animal		Spraying		Dipping	
		Taktic 12.5%	Lt. of water	Initial Fill	Replenishment
Cattle	Ticks	10 ml.	5	1 lt. Taktic per 500 lt. of water added.	1.5 lt. Taktic per 500 lt. of water added.
	Mange	10 ml.	5	1 lt. Taktic per 500 lt. of water added.	1.5 lt. Taktic per 500 lt. of water added.
Sheep Goat	Ticks	10 ml.	5	1 lt. Taktic per 500 lt. of water added.	1.5 lt. Taktic per 500 lt. of water added.
	Mange Lice/ Keds	20 ml.	5	2 lt. Taktic per 500 lt. of water added.	3 lt. Taktic per 500 lt. of water added.

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Observations on Clinical Ruminal Acidosis in Cattle and Buffaloes Around Patna

V. K. Sinha and B. Singh*
Department of Veterinary Epidemiology & Preventive Medicine, Bihar Veterinary College, Patna, Bihar

Introduction :

Ruminal acidosis is one of the major rumen dysfunctions seen in high yielding dairy cows and buffaloes. These dairy animals are much vulnerable to abrupt changes in the feed components particularly when excess carbohydrates rich grains are induced. This brings about upset in rumen digestion leading to acid indigestion. Genesis of ruminal acidosis is dependent on the lactic acid production by the gram positive bacteria and its subsequent absorption into the blood (Svendsen, 1979).

Materials and Methods :

In the present study twelve animals (eight cross bred cows and four buffaloes heifers) showing the symptoms of anorexia, dulness, salivation and history of accidental access to large quantities of whole wheat or rice grain, brought for the treatment at Bihar Veterinary College Hospital, Patna. They were examined as follows :

Temperature :
The rectal temperature were recorded immediately.

Ruminal Motility :
Ruminal movement were counted by placing hand on the left paralumber fossa and number of movement per five minutes were noted.

Rumen Liquor :
Rumen liquor was collected by a aspiration needle (6" long) and syringe before and post 6, 12 and 24 hours treatment and examined for the following.

pH Determination :
It was determined by wide range PH paper (BDH). Sedimentation Activity Time (SAT) was determined as per method of Nicholas and Penn (1958).

Protozoal Activities :
A drop of fresh ruminal fluid was taken on a clean glass slides and cover slip was fixed and examined under the microscope. Motality of protozoa was observed and graded as described by Misra *et al.*, (1972).

Lactic Acid Concentration :
Lactic acid concentration in the rumen liquor and blood was estimated before and post 6, 12 and 24 hours treatment as per the method of Barker and Summerson, (1941) in the protein free filtrate of rumen liquor and blood.

Treatment Schedule :

1. Neutralization of acidity war performed by using Magnesium hydroxide @ 100g followed by 60 g at 6 and 12 hour intervals.

2. Hostacycline® water soluble powder (Hoechst Roussel Vet Pvt. Ltd.) 50g

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* Institute of Animal Health and Production, Patna, Bihar

mixed with 250 ml of water and was given intra-ruminally.

3. Avil® (Hoechst Reussel Vet Pvt. Ltd.) 10ml intramuscular (I/m) was given, followed by 5ml daily for 2 days.

4. Berin was 10ml given I/m followed by 5ml daily for 2 days.

5. Half-a-litre of fresh rumen cud drawn from healthy animals and mixed with 50mg of Cobalt Sulphate, 50 tabs yeast and 100gm gur were given by mouth in the form of drench. This treatment was given at 24 hours.

Results and Discussions :

The decrease in pH of rumen liquor before treatment is believed to be due to increased fermentation activity of acid producing microbes (Koers *et al.*, 1976). Decrease in rumen liquor pH value in wheat induced acidosis in buffalo calves was reported by Sinha *et al.*, (1985). However, in the present study the decrease in pH of rumen liquor was not to the extent to cause appreciable clinical changes (Table I). This was clearly indicated by the symptoms observed in these animals. Symptoms of dehydration, opisthotonous and ataxia as reported by previous workers (Randhwa, 1979 and Sinha *et al.*, 1985) was not recorded in any of these animals because these cases were brought to the clinic immediately after ingestion of grains. However, white pasty diarrhoea seen in 4 of the 8 cows, was indicative of partially digested grain which might have escaped to lower alimentary tract associated with drainage of fluid. There was not much

Table I : Shows average values of different parameters in animals suffering from clinical ruminal acidosis.

Clinical Parameters of Observation	Values Before Treatment	Values at different hours (post-treatment)		
		6 hrs.	12 hrs.	24 hrs.
pH				
Rumen	5.00	5.50	6.00	6.50
Blood	7.35	7.40	7.45	
Lactic Acid (mg%)				
Rumen liquor	69	65	25	50
Blood	26	25	18	14
Protozoal Activity	-	-	+	++
Sedimentation activity time (in minutes)	65	55	45	30
Rectal temperature (°F)	102	101	101	100
Ruminal motility number/ 5 minutes	2	2	4	6

decrease in pH of blood before treatment. Although the corresponding lactic acid value was found raised than the normal ones. This might be due to strong buffering system of the blood. The protozoal activity decreased whereas sedimentation activity time increased as a result of acidosis. These early changes were due to the presence of lactic acidaemia and atony of rumen. On initiation of therapy, the values of different parameters started decreasing and by 24 hours these values returned to normal level. It clearly indicated that the present therapy proves efficacious in the treatment of clinical ruminal acidosis.

Conclusion :

Eight cross bred cows and four buffalo heifers with the history of accidental grain ingestion were diagnosed for acid indigestion and treated with oral dosing of Magnesium hydroxide, Hostacycline® water soluble powder and parenteral administration of Berin and Avil®. The aforesaid therapy alongwith drenching of fresh rumen cud from healthy animal, helped in combatting the acid indigestion.

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Influence of Nutriliv Forte on Broiler Performance

C. B. Singh, R. J. Sharma and Vir Singh
Hill Campus, Ranichauri, Tehri - Garhwal, U.P.

Introduction :

Many growth promoters and liver stimulants have been found to improve the weight gain in broilers and help in feed conversion. Liv 52 powder at 0.1% level in broiler ration has improved the weight gain significantly (Subramanian *et.al., 1982*). Protosel at the rate of 0.1% ml/chick in drinking water has been found to increase the body weight of broilers. (Singh *et.al., 1996*). Improved body weight in broilers was also observed by addition of Vetroliv to the feed. (Ramkrishna *et.al., 1987*). The present investigation was undertaken with a view to study the effectiveness of Nutriliv Forte (Vet-Care) as a nutritional supplement in drinking water at various level on broiler performance.

Materials and Methods :

Three hundred day old (initial body weight 38.85 gm.) commercial broiler chicks (Van- cob) of a single hatch were selected for this study. Chicks were kept in a deep litter system in PALI village poultry farm of Tehri District.

Three farms were selected for this purpose. During the study period (0-8 weeks) balanced ration and water was provided in all groups *adlibitum*. The Nutriliv Forte was given in drinking water from 0-8 weeks in two groups at the level of 10ml/ 100 chicks (group A) and 20 ml/ 100 chicks (group B) and the last group was served as control group (group C).

The data pertinent to feed consumption, body weight and mortality was observed for all three groups and the feed conversion efficiency, mortality percentage and body weight gain were calculated.

Result and Discussion :

The performance of broiler chicks of group

Table I : Performance of broiler chicks supplemented with Nutriliv Forte in drinking water

Sr. No.	Treatment Groups	8 weeks body weight (gm)	Feed efficiency	Mortality percentage (%)
1.	Group A (10 ml Nutriliv Forte /100 chicks)	1272	2.89	6
2.	Group B (20 ml Nutriliv Forte/ 100 chicks)	1289	2.88	5
3.	Group C (control group)	1242	3.08	7

A, B and C is presented in Table 1. The group B give 1.30% more body weight than group A and 3.64% than group C. The average body weight in group A, B and C were 1289 gm, 1272 gm and 1242 gm respectively.

Improved body weight by the supplementation of liver tonics has also been reported by (Subramanian *et.al., 1982*) and (Ramkrishna *et.al., 1987*).

However, the feed conversion efficiency has not established any significant difference but better conversion efficiency i.e. 2.89 and 2.88 were observed in group A and group B than the Nutriliv Forte help in better feed conversion.

Prasad and Singh (1983) could not observed any improvement either in weight gain and feed conversion.

Mortality percentage were 6,5, and 7 in groups A, B and C, respectively. Sundarasu *et.al., (1985)* recorded a 3% mortality in broilers in liver tonic trial on farm/station level.

Nutriliv Forte is a good nutritional supplement which helps in faster weight gain of broiler birds. The results of the experiment suggest that the poultry farmers in the hills can use the Nutriliv Forte as a nutritional supplement for broiler chicks at their farms.

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Let us preach where we all agree and leave the differences to remedy themselves.

As I have said to the Indian people again and again, if there is the darkness of centuries in a room and we go into the room and begin to cry, "Oh, its dark, its dark !", will the darkness go ? Bring in the light and the darkness will vanish atonce.

- Swami Vivekanand

Flavomycin-40 as a Feed Supplement in Dairy Cattle for High Milk Yield

V. S. Narsapur, S. R. Vaidya and A. K. Datta*
DOLCOS Veterinary Pathology Laboratory, Jogeshwari, Mumbai

Introduction :

Flavomycin-40 (Flavophospholipol) is a performance promoter and is widely used as feed supplement in livestock and poultry feed.

In cattle Flavomycin-40 has many modes of action. It selectively influences the rumen microflora so that amylolytic and cellulytic micro-organisms, which improve digestibility of cellulose are favoured. It also leads to increased formation of propionic and acetic acids, decrease in methane production, improvement in microbial protein synthesis and thereby improvement in energy and protein conversion from feed.

The trials of supplementing Flavomycin-40 in feed have been conducted in cows by Ruffo and Valerani, (1977), Baharecke *et al.*, (1984) Senft *et al.*, (1988) and Gunther, (1987) and all have reported that daily feeding of Flavomycin-40 increased the milk yield, milk fat and milk proteins in each and every animal. The positive effect is dose related. Thus at 50 mg dose of flavophospholipol/ animal/ day, the milk yield increased by 2.66%, fat by 2.16% and protein by 3.99, while at 130 mg dose flavophospholipol/ animal/ day, the milk yield increased by 10.2 - 11.9%, fat 30 - 40 kg and milk protein 22-27.3 kg/animal. Gunther, (1987) further stated that, Flavomycin-40 supplementation started at the beginning of lactation would prove more beneficial than at a later stage in lactation. In view of the above a trial was undertaken to find out the effect of Flavomycin-40 (Flavophospholipol) supplementation in feed in cows under the local climatic condition, of climate, management and nutritional status in dairy farms around Mumbai.

Materials and Methods :

The trial was undertaken in a cattle farm, managed by Girivanwasi Pragati Mandal, Nageshwarwadi, Dahanu from March to June, 1997.

This farm has a herd of about 50 cross bred cattle, out of which 37 were in milking and in different lactations. The animals are stall fed, are given concentrates, grass and hay. General management, hygiene, biosecurity of the farm was found satisfactory. The animals were all clinically healthy.

The milking animals were divided into two groups namely, 'T' (Treated) and 'C' (Control), consisted of 19 and 18 animals respectively of matching lactation and matching milk yield. Group 'T' animals were given 2.5 gms of Flavomycin-40 (equivalent to 100 mg of Flavophospholipol) per day per animal for conseculive 90 days. Group 'C' animals were not given Flavomycin-40 and they were represented as contol group. Both the groups were maintained under identical conditions of feed and environmental management. The following observations were recorded :

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* Hoechst Roussel Vet Pvt. Ltd., Neeta Park, Yerwada, Pune

1) Daily milk yield of each animal for 90 days prior to start of trial (8-12-96 to 7-3-97) noted from records and average milk yield per day per animal was calculated for this period (period A)

2) Daily milk yield of each animal for 90 days of trial period (8-3-97 to 6-6-97) (period B) was noted and average milk yield per day per animal was calculated for this period.

3) The percentage increase/decrease in milk yield per animal was calculated by comparing the value for periods A with B in both T and C groups and also comparing value of group T with that of group C.

4) Milk of 6 animals each from groups T and C were tested in Laboratory for fat and solids not fat (SNF) contents on 8-3-97 (start of trial) and again on 6-6-97 (at close of trial)

5) Blood of 6 animals each from Groups T and C were tested in Laboratory for Haemoglobin value (grams per 100 ml) on 8-3-97 (start of trial) and again on 6-6-97 (at close of trial)

Observations :

Out of 37 animals initially selected for trial, 11 animals had to be dropped out, since the milk yield in them dropped too steep owing to approaching dry period. Hence the trial results are based on 26 animals, 13 in each group.

Milk Yield (Reference Table I) :

In Flavomycin-40 treated Group (T), there was an average increase of 0.174 L of milk/animal/day during the period of 90 days Flavomycin-40 feeding. The treatment helped to increase of 3.8% in milk yield compared to pre-treatment levels. The increase was higher in younger animals(animals in II to IV lactation) than in older animals (V and above lactation).

The response was not uniform in all the animals and in 3 of 13 T group there was even marginal decline in the yield.

In controls there was decline in the milk yield in the corresponding period (average - 0.129 L of milk/animal/day) which was declined by 2.6% as compared to yield at pre-trial levels.

Comparing the T and C groups, the net increase of milk yield in Flavomycin-40 treated group works out to 6.4% over. The pre-trial averages of T and C groups taken together.

Milk Constituents (Reference Table II):

In the treated group, the net increase of fat in milk was by 13.4% and solid not fat (SNF) content was by 3.6%. In this aspect also there was individual variation.

Haemoglobin (Reference Table III) :

In the treated group, the net increase in Haemoglobin level was by 19.1% subject to again individual variation.

Discussions :

The results indicate that Flavophospholipol (Flavomycin-40) supplementation in feed @ 100 mg/animal/day (i.e. 2.5 gram of Flavomycin-40) decidely improves milk yield, milk fat, milk SNF contents and Haemoglobin levels.

The milk yield gradually declined in untreated animals which appears to be normal as lactation progresses and dry

Table I : Shows average daily milk production (litres) in treated and control group

Duration of Observation : A - 90 days before treatment (8-12-96 to 7-3-97)
B - 90 days during treatment (8-3-97 to 6-6-97)

Flavomycin-40 treated group					
No.of cows	Lactation	A	B	B-A	% Difference
7	II	4.033 (2.37-6.51)	4.218 (2.67-5.93)	+0.186 (-0.084 to + 0.495)	+4.6%
3	III & IV	4.811 (4.1-6.02)	4.982 (4.155 to 6.25)	+0.171 (-0.159 to 0.442)	+3.6%
3	V & above	5.625 (3.341-7.358)	5.773 (3.697-7.497)	+0.148 (0.05 to 0.3565)	+2.6%
13		4.58	4.753	+0.174	+3.8%

Control Group					
No.of Cows	Lactation	A	B	B-A	% Difference
7	II	5.66 (2.725-8.775)	5.61 (2.992-8.355)	-0.05 (-0.12 to +0.364)	0.9%
3	III & IV	4.187 (1.733-6.5)	4.021 (1.577 to 6.367)	-0.167 (-0.133 to +0.213)	-3.9%
3	V & above	3.588 (2.511-4.8)	3.353 (2.352-4.472)	-0.279 (-0.159 to 0.353)	-7.6%
13		4.851	4.722	-0.129	-2.6%

(Figures in ( ) brackets indicate range)

(Net Increase of milk yield in Flavomycin-40 treated group over pre-trial average of both groups = 6.4%)

period approaches, Flavomycin-40 supplementation nullifies this decline and further maintains the yield at higher level during and after mid lactation. The findings in this trial in respect to increase of milk yield, and as well as higher constituents of milk are comparable to the observations of Senft *et al.* 1988.

If haemoglobin level can be presumed as an indicator of status for good health it can be concluded that, the health of the animals is improved by 19.1% with Flavomycin-40 treatment. This would give undoubtedly a boost for immune competence, stress tolerance capacity of animals thereby contributing to the improvement in production.

Conclusion :

Flavomycin-40 supplementation @ 2.5g (equivalent to 100 mg of Flavophospholipol) /animal/day for 90 days, improves milk yield by 6.4%, milk fat by 13.4% and milk SNF contents by 3.6% Flavomycin-40

Table II : Shows effect of Flavomycin-40 treatment (90 days) on composition of milk (A : Fat percent and B : Solids Not Fat, SNF) in dairy Cattle

Group	No. of Cows	Values on 8/3/97 I	Values on 6/6/97 II	II-I	%age Difference
A : FAT PERCENT MILK					
Treated	6	4.07 (37-4.9)	4.72 (4.1-5.5)	+0.65 (0.1-1)	15.9%
Control	6	4.208	4.32	0.11	2.6%
Net increase (average) of milk fat % in treated compared with the controls					13.4%
B : SOLIDS NOT FAT CONTENTS IN MILK					
Treated	6	9.263 (9.0-9.59)	9.61 (9.22-10.12)	+0.345 (0.02-0.96)	3.7%
Control	6	8.795 (8.26-9.54)	8.81 (8.2-9.4)	+0.01	0.1%
Net increase (average) of milk fat % in treated compared with the controls					3.6%

Figures in ( ) brackets indicate range

Table III : Shows effect of Flavomycin-40 treatment (90 days) on blood haemoglobin levels in Dairy Cattle

Group	No. of Cows	Values on 8/3/97 I	Values on 6/6/97 II	II-I	%age Difference
Treated	6	8.6 (7.8-9.6)	10.66 (9.8-12)	+2.07 (1.4-2.8)	24.0%
Control	6	8.86 (7.6-9.8)	9.1 (7.8-10.2)	+0.433 (-0.6 to +0.4)	4.9%
Net increase (average) of milk fat % in treated compared with the controls					19.1%

Figures in column I, II, and II-I, are values of Haemoglobin Grams per 100 ml. blood.

Figures in ( ) brackets indicate range.

supplementation at the said level also boosts up Haemoglobin level by 19.1%.

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Enrofloxacin (Floxidin® Vet) in the Treatment of Sub-clinical Mastitis in Cows

L. Vijata Reddy, P. C. Choudhuri and P. A. Hamza*
Department of Medicine, College of Veterinary Science, Tirupati, Andgra Pradesh

Mastitis is one of the most problematic diseases affecting dairy cows and causes substantial economic loss to the dairy farmers. The disease may occur in the clinical form or in the sub-clinical form without any apparent clinical symptoms. Normally, the sub-clinical form remains cryptic at the time of lactation and dry period. If it is untreated, it may manifest clinical mastitis during the post-parturient period. For an effective prevention and control of mastitis in a dairy herd diagnosis and treatment of mastitis in the sub-clinical from is very essential. However, effective treatment of sub-clinical mastitis in bovine is eluding due to development of drug resistance as a result of prolonged indiscriminate use of antibiotics. To overcome this problem newer antibiotics and chemotherapeutic agents are to be evaluated from time to time (Pal *et al.*, 1989). In the present study an attempt was made to assess the efficacy of the newly introduced bactericidal quinolone derivative 'Enrofloxacin' (Floxidin® Vet, Hoechst Roussel Vet Pvt. Ltd.) as compared to the conventional antibiotic combination of Ampicillin and Cloxacillin in the treatment of sub-clinical mastitis in cows.

Materials & Methods :

Milk samples were collected aseptically from 20 crossbred cows of 4 to 6 years of age in their early lactation (i.e. from 15-90 days of calving) into sterile tubes. A portion of the sample was introduced into nutrient broth and incubated aerobically for 12 hrs at 37°C. Subsequently an inoculum of 0.1 ml of the incubated sample was placed into blood agar and other selected media in petri dishes. Depending upon the preliminary characters the pure cultures were identified upto generic level using appropriate cultural and biochemical tests as per Bergey manual of determinative Bacteriology (Buchanan and Gibbons, 1974 and Collee *et al.*, 1989). Based on the cultural, studies, 12 cows which were found positive for sub-clinical mastitis were selected for conducting the therapeutic trial. These animals were randomly devided into 2 groups of 6 each. Cows of group I were given intra-mammary injection of Enrofloxacin 150 mg (1.5 ml of Floxidin® Vet 10% injectible solution, Hoechst Roussel Vet Pvt. Ltd.) diluted in 6 ml of sterile distrilled water through the teat canal in to each of the 17 affected quarters daily for 3 consecultive days.

Cows of group II were injected with Cloxacillin (200 mg) + Ampicillin (75 mg) suspended in 6 ml distilled water into each of the 13 affected quarters daily for 3 days.

Each quarter treated, was reexamined three days after completion of the course of treatment. The post-treatment milk samples were subjected to CMT (California Mastitis Test), EC (Electrical Conductivity) and bacteriological studies.

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* College of Veterinary Science, Rajendranagar, Hyderabad.

CMT (California Mastitis Test) was performed for the milk samples as per the procedure of Schalm and Noorlander, (1957) employing a modified reagent (Sharma and Rajani, 1969). Electrical conductivity (EC) of milk samples was tested by using milk checker (manufactured by Oriental Instruments Limited, Tokyo, Japan). The sampling cup of the milk checker was filled with milk nearly to full and the switch was pressed "ON". Holding this switch in "ON" position, Electrical Conductivity (EC) was read on digital display (3 digit display with an accuracy of 3% ± 1 digit).

Results and Discussions :

Out of 17 sub-clinically effected quarters from 6 cows treated milk became culturally negative giving a recovery rate of 88.23% quarter wise and 66.66% cow-wise (Table I). Milk samples of all the recovered animals became CMT (California Mastitis Test) negative. In the recovered animals the Electrical Conductivity (EC) became 4.4 ± 0.19 ms/cm from the pretreatment value of 5.16 ± 0.22 ms/cm. The treatment was effective 100% against *Micrococcus spp.* and *Streptococcus spp.* infections. Enrofloxacin is a quinolone derivative and information regarding the treatment of sub-clinical mastitis with this drug is not available. Radostits *et al.*, (1994) have also reported that the new quinolones have an excellent activity and it is worth trying for the treatment of mastitis.

The combination of Ampicillin (200mg) and Cloxacillin (75mg) cured 9 out of 13 quarters and 3 out of 6 animals accounting for 69.23% and 50.00% recovery rate quarter wise and animal wise respectively. After treatment of 9 milk samples from the recovered animals became CMT (California Mastitis Test) negative. The Electrical Conductivity (EC) of milk which was 5.12±.12 ms / cm causes down to 4.49±0.18

Table I : Shows the effect of Floxidin® Vet. (Enrofloxacin) treatment cow-wise (A) and total quarter-wise (B) in comparison to Ampicillin and Cloxacillin treatment is sub-clinical mastitis.

Groups	A: Number of Cows			B: Number of quarters		
	Total No. Tested	Total No. Cured	% Cure	Total No. Tested	Total No. Cured	% Cure
Group I	6	4	66.66	17	15	88.23
				Species-wise		
Enrofloxacin (150 mg in 6 ml D.W./ quarter)				*Staphylococcus Spp.*(7)	6	85.71
				Micrococcus Spp.(7)	7	100.00
				E. coli (2)	1	50.00
				Streptococcus Sp. (1)	1	100.00
Group II Ampicillin+Cloxacillin	6	3	50.00	13	9	69.23
				Species-wise		
(200mg + 75 mg in 6 ml D.W./ quater)				*Staphylococcus Spp.*(10)	7	70.00
				Micrococcus Spp.(2)	1	50.00
				E.coli (1)	1	100.00

Figures in Parenthesis, indicate the total numbers of affected quateres treated.
D. W. : Distilled Water

ms/cm in the recovered animals. It was effective against 70% of *Staphylococcus* affected cases, 50% of *Micrococcus spp.* and 100% of *E. coli* infection. Pal *et al.*, (1989) found that the combination of Ampicillin plus Coxacillin sodium could cure 66.67% of staphylococcal infections. While Dhakal, (1984) reported a cure rate of 62.50% (quarter infections), by treating with Campidex (containing Ampicillin 75mg and Cloxacillin 200mg). Rama Rao, (1987) and Pendnekar and Swarup, (1991) observed a higher cure rate of 80.00% and 83.33% with Floclox D and Ampiclox respectively.

Conclusion :

Therapeutic trials were conducted in 18 sub-clinical mastitis cows of three groups in early lactation. Enrofloxacin (150 mg) infused into 17 quarters of 6 cows belonging to group I brought about 88.23% cure rate, while 13 affected quaters of 6 cows of group II were infused with a combination of Ampicillin plus Cloxacillin, and brought only 69.23% recovery rate, indicating to be less effective than the former treatment (Floxidin® Vet).

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Post-bite Efficacy of Candur®R (PCEC Rabies Vaccine) in Dogs

R. Jayakumar, N. Premalatha, K. Kumanan and K. Nachimuthu
Department of Animal Biotechnology, TANUVAS, Madras Veterinary College, Chennai

Introduction :

Rabies is an important zoonotic disease, that kills thousands of human beings and millions of animals each year (Baer, 1988). The problem is most severe in developing countries. In India, about 700,000 people undergo rabies post-exposure treatment each year following rabid animal bite (Petricciani, 1993). In India, stray dogs play a primary role in maintaining and spreading rabies among animals and man (Lakhanpal and Sharma, 1985., Bhatia *et al.*, 1988 and Madhusudanan and Agarwal, 1990). In view of high cost of post-exposure treatment in man, the elimination of dog rabies has become even more important. Immunization of domestic animals represents the single most effective means in the control of rabies in animals and subsequent human exposure (Kappus, 1976).

Post-exposure treatment in man and pre and post-exposure treatments in animals is practiced throughout India. More than twenty four centres are producing anti-rabies vaccine both for human and animal use. Majority of these centres are engaged in the production of five per cent sheep brain vaccine. Only three centres engaged in the production of BHK 21, Vero and Purifed Chick Embryo Cell (PCEC) culture inactivated vaccines for animal use in our country. Research in various parts of the world resulted in the production of a safe and potent tissue culture rabies vaccine using primary chick embryo cells as the cell substrate (Barth *et al.*, 1985).

This fibroblast vaccine is now marketed in India by Hoechst Roussel Vet Pvt. Ltd. under the trade name Candur®R and currently used for prophylactic as well as post-exposure treatments in animals.

The present study was designed to test the protective capacity of Candur®R (PCEC Rabies Vaccine) in dogs as per the requirement, prescribed by the licensing authority of India.

Materials and Methods :

Vaccine :

Rabies vaccine, Candur®R were used from Hoechst Roussel Vet Pvt. Ltd., a purified Chick Embryo Cell (PCEC) cultured inactivated vaccine (Batch No. 011), having a potency value of 1.0 iu/ml.

Rabies Challenge Virus :

The street rabies virus was collected from the brain and salivary gland of a proven rabid dog. The material was homogenized in Minimum Essential Medium (MEM) containing 20% bovine foetal calf serum and adequate antibiotics. The homogenized material was titrated in suckling mice and the titre was found to be $10^{4.8}$ $MICLD_{50}$ per 0.03 ml.

Experimental Animals and Design :

Thirty, nondescript male dogs of 2-3 years of age were procured locally and housed in individual cages. (Fig. 1 and Fig. 2) Dogs were dewormed with Panacur® 150 mg tablets (Hoechst Roussel Vet Pvt. Ltd.) @ 50 mg/ kg B. W. for conseculive three days and tested for the presence of rabies

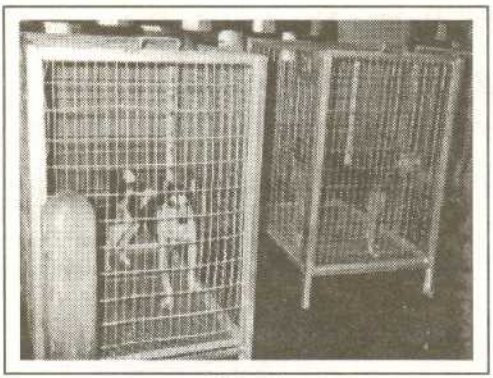

Fig. 1

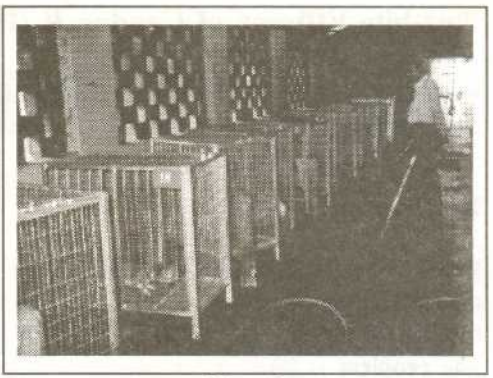

Fig. 2

Figs. 1&2 : Show experimental dogs, housed in individual cages.

antibodies by Dipstick dot ELISA (Jayakumar *et al.*, 1995). Only those animals found negative for rabies, were included in the study. They were fed with rice and beef *ad-libitum* and supplemented with minerals and vitamins. The dogs were divided into three groups, each consisting of 10 animals. All these animals were inoculated with 1 ml of street rabies virus ($10^{4.8}$ $MICLD_{50}$ per 0.03 ml.) by deep intramuscular route in the hind legs. After exposing all the animals with rabies virus, these animals were vaccinated (groupwise) as scheduled below.

Group I
10 dogs received 1 ml of test vaccine by intramuscular route on days 0, 3, 7, 14, 28th post-exposure to virulent rabies street virus inoculation.

Group II
10 dogs were inoculated with test vaccine by intramuscular route on days 0, 3, 7, 14, 28 and 90th post-exposure to the rabies street virus.

Group III
10 dogs served as control without any vaccination after the rabies street virus

Table I : Shows the efficacy of Candur®R in post-challenged experimental Dogs

Challenge Virus	Vaccination Schedule		Number of dogs Died / Number of dogs Challenged	
	Group 1	Group 2	Group 1	Group 2
Rabies street virus $10^{4.8}$ $MICLD_{50}$ per 0.03 ml, dose 1 ml I/M	0 day 3 day 7 day 14 day 28 day	0 day 3 day 7 day 14 day 28 day 90 day	0/10	0/10

challenge and reared away from the treatment group in individual cages. All the animals were observed daily for the development of any clinical signs till the completion of the experiment period of six months. The animals that survived the study period were sacrificed and their brain and salivary glands were collected and tested for the presence of specific rabies antigen.

Table II : Shows the results of control Dogs, inoculated with Street Rabies Virus

No. of Dogs Inoculated / No. of Dogs Died	Challenge Virus	Average Incubation Period	Form of the Disease		Confirmatory Test	
			Furious	Paralytic	FAT	DOT ELISA
10/10	Rabies Street Virus $10^{4.8}$ $MICLD_{50}$ / 0.03 ml, 1 ml I/M	55 days	88%	20%	100%	100%

Results and Discussions :

All the 20 animals in group I and II which received 5 doses and 6 doses respectively of the test vaccine Candur®R, survived the challenge with rabies street virus. These animals in group I and II were killed at the end of the study period and their brain and salivary gland samples did not reveal the presence of rabies antigen (Table I), where as the control group (group III) animals succumbed to the disease. The brain and salivary gland samples were found to be positive for rabies antigen (Table II).

Number of studies have been conducted in human post-exposure treatment with purified chick embryo cell (PCEC) culture rabies vaccine in India (Seghal *et al.*, 1988., Selvakumar and John, 1989., Madhusudan and Tripathi, 1989, Sheth *et al.*, 1992, Natarajan *et al.*, 1992., Seghal *et al.*, 1994 and Seghal *et al.*, 1995). But in case of animal vaccination with purified chick embryo cell (PCEC) culture vaccine in domestic animals, the present study could well be the first of its kind in India.

The experiment described and discussed in this report is designed to demonstrate the importance of post-exposure treatment with the purified chick embryo cell (PCEC) culture vaccine in dogs as per the schedule of vaccination recommended by the manufacturer namely on days 0, 3, 7, 14, 28 and 90. However, the curative efficacy of rabies vaccines in animals has been found to be very poor in the experiments conducted by a number of earlier workers (Garg, 1989., Blancou *et al.*, 1991., Ramanna *et al.*, 1991., Baltazar and Blancou, 1995 and Clark and Wilson, 1996).

The present experiment was conducted according to the guidelines of World Health Organization (WHO) and it is evident from the results that all the vaccinated animals survived and unvaccinated animals succumbed to the rabies virulent street virus challenge. Since all the unvaccinated controls died due to rabies infection, the challenge experiment could be regarded as fool proof. Most of the developed countries sacrifice their animals exposed to rabid animal bites. However, such policy could not be possible to advocate in India due to the socio-economic conditions and religious sentiments. The results of the

present study showed that the Candur®R rabies vaccine is protective irrespective of the doses (5 or 6) used in post-bite situation in dogs.

Acknowledgments :
The authors are grateful to Dr. A. K. Datta, Head, R & D and Regulatory Affairs, Hoechst Roussel Vet Pvt.Ltd for the financial assistance for this study. The authors are also very much grateful to Dr. A. T. Venugopalan, Director, Centre for Animal Health Studies, TANUVAS, Chennai and the Dean, Madras Veterinary College, Chennai for the facilities extended and constant encouragement.

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Case Report :
Note on Outbreak of Pox in Sheep

K. Nalini Kumari, P. C. Choudhuri and P. Annapoorna
College of Veterinary Science, Tirupati, Andhra Pradesh

Of all the pox diseases in animals, sheep pox is most serious and causing death rate of 5% in the benign form and 50% in the malignant form, often seen in lambs (Radostits *et al.*, 1994). Goat pox can also cause more serious lesions in sheep than sheep pox virus. Sheep pox is endemic in various parts of the country. Several outbreaks have been reported by Murty and Singh, (1971); Kataria and Sharma, (1992); Reddy Krishna Mohan, (1993); Bhaskar Rao *et al.*, (1994) and Prasad *et al.*, (1995). The present communication describes an outbreak of sheep pox in sheep at College of Veterinary Sciences, Tirupati.

History :

Thirty six ewes of Nellore breed were procured locally from the neighbouring district, Cudapah. The animals were kept in quarantine for 1 month in a separate compound at about a quarter kilometer away from the existing sheep shed.

Observations :

From the old flock consisting of 75 sheep of Nellore breed, symptoms of sheep pox were noticed in 6 ewes (3-4 years) on 9th day and in another ewe (4 years) and an ewe lamb (4 months) on 13th day of introduction of new animals into the campus though kept seperately. The lesions observed were papules and nodules under the tail, on the vulval lips and nostrils (all animals) and all over body i.e., on the ears, face, back, groin, axilla, belly, legs, perineum and udder (3 animals). The ewes showed no other abnormalities, except the skin lesions. But the lamb was dull, depressed, anorectic and showed a febrile reaction (106.8F), congested conjunctival mucous membranes, with mucopurulent discharges and mucoid nasal discharges and pneumonia. The affected animals were immediately segregated and given treatment. The lesions were washed with potassium permanganate lotion and boric acid ointment was applied. The affected animals were given Oxytetracycline (5 mg/1 kg b. wt, I/M) and Chloril (1-2 ml I/M) for 5 days. The body temperature became normal by 6th day but the lamb became recumbant by 9th day. The lamb also received Liver Extract (2 ml, I/M) on alternate days from the initiation of therapy and 15 ml of Dextrose Saline intravenously from 9th onwards. But the lamb died on 13th day.

The skin lesions passed through the stages of papules, nodules and scab formation by 15 days. Vesicular and pustular stages were not very much appreciated. The scabs persisted for another 15-20 days before they finally dropped off leaving a white patch. The remaining flock was immediately vaccinated after which no fresh cases were recorded.

Discussions :

After the outbreak, on further investigation it came to light that the new flock was said to have been protected by Ovination by the source of the owners as proper vaccination facilities were not available in such an interior endemic area. Subsequently a thorough examination of the new flock revealed, infective discharges from the site of Ovination in three animals. Though the new

flock was kept away, the same personnel handling both the flocks might have spread the disease. The animals affected were all females which supports that females are more susceptible (Murty and Singh, 1971). The morbidity, mortality and case fatality rates noticed in the present case, were 10.7%, 1.3% and 12.5% respectively. A morbidity rate of 60.75% (Bhaskara Rao *et al.*, 1994) and 20% (Reddy Krishna Mohan, 1993) were reported in earlier outbreaks.

In the present outbreak, presentation of cases even with nodules on 9th and 13th day of introduction of new animals may suggest the onset of disease 3 or 4 days earlier which inturn indicate the incubation period ranging from 5 to 8 days which is agreeable with Radostits *et al.*, (1994). An observation of absence of vesicles and pustules coincides with the findings of Manohar Rao, (1978). The course of the disease was 30-35 days which is similar to the observations of Murty and Singh, (1971). In addition to the presence of skin lesions absence of system reaction in ewes and the presence of it in the lamb indicate that the ewes suffered from a benign while the solitary lamb had malignant form of the disease. Bhaskar Rao *et al.*, (1994) and Reddy Krishna Mohan, (1993) reported systemic involvement with pneumonia.

Inspite of absence of systemic reaction, antibiotics were given to prevent secondary bacterial infection. Absence of fresh cases after vaccination with a live attenuated vaccine indicate its efficacy. Two goats kept along with sheep remained unaffected suggesting that the infection could be sheep pox.

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Case Report :
A Case of Cholelithiasis in a Cattle

Neelu Gupta
Department of Pathology, College of Veterinary Science and Animal Husbandary, Anjora, Durg, (M.P.)

Cholelithiasis is commonly known as gall stones in gall bladder. The gall stones are composed of a mixture of cholesterol, bile pigment, salts of bile acid, calcium salt. Domestic animals and are not clinically significant until the gall stores obstruct the billiary system (Thomas, 1989 and Sastry, 1983). Present paper records a cause of death in cattle due to cholelithiasis.

A carcase of an adult female Sahiwal Cattle, 10 year of age from Research Dairy Unit, Department of Physiology, College of Veterinary Science & Animal Husbandary, Anjora, Durg (M.P.), was received for necropsy. Cattle died after prolonged illness, showing clinical signs of dullness, depression, gradual loss of body weight and recumbancy. On necropsy, lungs were found congested, liver showed congestion, inflammation with some yellowish spots. Gallbladder showed yellow, green and blackish gall stones, one large and two small. All were friable in consistency. Wall of gall bladder was thickened and firm (Fig. 1).

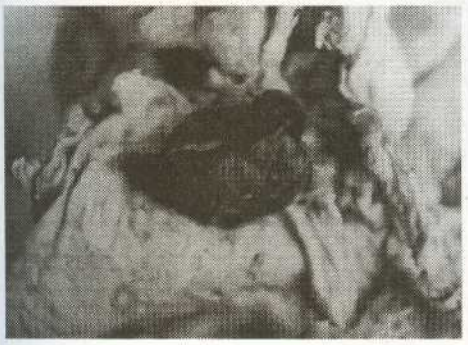

Fig. 1 : Cholelithiasis in Sahiwal Cattle showing Gall Stone in gall bladder.

On microscopic examination of H & E stained liver sections, showed coagulative necrosis and infiltration of macrophages, lymphocytes, plasma cells and few neutrophils. Bile duct was dialated, Gall bladder showed atrophy of columnar epithelium, hypertrophy of muscle bundles and infiltration of lymophocytes, plasma cells, macrophages and few eosinophils.

The necropsy lesions were suggestive of gall stones as reported by Jubb (1992) and Sastry (1983). The result of histopathology indicate that atrophy of columnar epithelium of gall bladder was due to pressure of stones.

In the present case, the cause of death may be due to cholelithiasis. Jubb (1992) reported gall stones formation in cattle. He suggested that the origin of such stones is uncertain, but their development is probably secondary to chronic disease and related to disturbances of the resorbing activity of the gall bladder, where by the bile salts are removed faster than the stone forming compounds.

Sastry (1983) suggested that the gall stones occur as a result of cholecystitis. The dead cells of bacterial or mucous membrane origin may form the nuclei, around which deposition of cholestrol bile pigments, and bile salts are occured. Sand particles and food materials that may reach the gall bladder through the bile duct from the duodenum during violent peristalsis which may also form nuclei of stone formations.

Acknowledgements :
The authors is greatful to the Dean, College of Veterinary Science & Animal Husbandary, Anjora, Durg (M.P.) for the facilities provided.

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Case Report :
Schistosomus reflexus in a Mehshana Buffalo

R. K. Tanwar
Department of Animal Breeding and Genetics, College of Veterinary and Animal Science, Bikaner, Rajasthan

Schistosomus reflexus is a fetal monster characterized by a marked ventral curvature of the spine so that the occiput of the head lies near the sacrum. The body and chest walls are bent laterally and thoracic and abdominal viscera are exposed (Roberts, 1971). It is a non-inherited teratogic defect observed to occur very commonly in cattle, very rarely in sheep and goats. Two cases of schistosomus reflexus have been observed in buffaloes in a conjoined female twin and in a co-twin with normal dead, female calf (Sastry and Murthy, 1984). This report describes a case of single schistosomus reflexus monster in a Mehshana buffalo, hither to unreported (Fig.1).

An eight year old Mehshana buffalo was examined per vaginum in its second calving as she was unable expel out the fetus even after 2-3 hours of rupture of water bag. On examination, it was found that four limbs were present in vagina and head was lying in between the limbs. No other region was palpable. Rectal examination proved futile in ascertaining the foetus presentation, position and posture. It was suspected to be monster and decided to apply traction using chains on all the four legs. By applying traction in this manner, resulted in expulsion of monster pumped for two minutes.

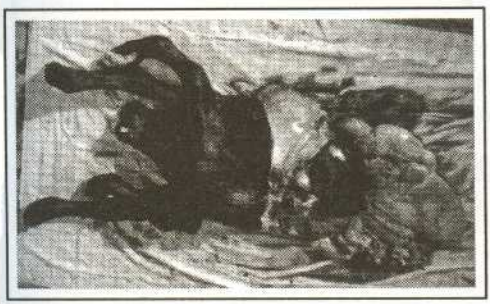

Fig. 1 : Shows Schistosomus reffexus monster in a Mehshana Buffalo.

There was marked curvature on the back. Occiput of the head and sacrum were lying together. Hoofs of all the four legs were resembles like equine. There was no ankylosis of joints and limbs were flexible. Upper eye lid and lower eye lid of left eye were united together and no eye lashes were present on that eye. One kidney was absent. Five pairs of ribs were present only. There was complete absence of thoracic and abdominal walls and viscera were exposed. Liver was enlarged and swollen. The rumen and reticulum was distended with fluid. Sex organs of neither male nor female were present except one large and one small scrotal sac. Anus was absent. Both lungs were small in size as compare to lungs of fully developed calf.

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"Hongkong's bird flu of 1997"

In Hongkong, from a 3-year old child who died of respiratory failure in May 1997, a strain of influenza virus has been isolated and identified by scientists at the WHO collaborating centre (at USA, Netherlands & UK) as influenza A (H_5N_1). This virus strain is known to infect ducks, chickens and other birds but never been previously isolated from humans.

Till end of December 1997, a total of 13 confirmed & six suspected cases of H_5N_1 influenza have been identified in people of all age groups and four reported died (Three confirmed & one of suspected cases)

Blood samples tested of 502 persons from Hongkong, nine were found positive for antibodies against influenza A H_5N_1 strain. All these were among poultry workers and none from the family members or in contact of the ill child died in May 97. Hence David Heymann, Director, Division of emerging & other communicable Diseases surveillance WHO /HQ Geneva stated that from person to person transmissions, if occurring, is relatively inefficient at this time and that none of the available evidences suggest the possibility of a wide spread epidemic.

Following precautionary measures have been undertaken :

1. Hongkong authorities have eliminated nearly 103 millions chickens in two days (30 and 31 Dec.97) and have disinfected the farms and processing plants.
2. Centre in Disease control and prevention, Atlanta, USA, has prepared a kit for diagnosis of H_5N_1 Virus Strain which is being sent to all National Influenza Centres.
3. WHO collaboration centres are working on various H5 strains to develop high yield strain which could be made available for vaccine production in case of need.
4. Work with H_5N_1 Virus is being restricted to laboratories having high emtainment level P3+.

However, no travel restrictions have been suggested so far.

Circular No. IC-97-35, Dated : 31.12.97.
WHO Regional Office for South East Asia

More about 'flu'

Dr. Kennedy Stuartidge of university of Hongkong who has done pioneering work (studied more than 1 million Virus Samples) on influenza postulates that China was a major source of world's viruses and that and their ancestors are few original reservoirs.

After 1640s, ducks in China found their way from viruses to paddy fields and they began their intimate association with freeness and farm animals. Influenza viruses which thrive in duck intestine, passed through faces to water and respiratory tract to pigs where they became adapted to survival in man. The spanish flu of 1918 that killed 20 million people was earned by virus the Hongkong few of 1968 which killed 700,000 people all over the world could have been originated from ducks from China.

Time, vol. 150 : 25 Dt. 22.12.97.

Avian leukosis Virus Sub group J. : ALV-J

A new subgroup of exogenous ALV viruses has been isolated from broiler breeders and named ALV-J Virus. It causes myelocytomas particularly in meat type lines of chickens and form tumours on inner-sternum vertebrae, and infiltration of liver, spleen and

kidneys. In most cases tumours are normally seen from sexual maturity onwards but cases have been observed before lower of age(4-9 weeks). Mortality may vary from either 1-2% and even 20% and immuno- suppression is marked.

ALV-J spreads horizontally more than other sub-groups of ALV and is getting more prevalent in commercial meat strains. Layer hens may be reservoirs of ALV-J. No treatment or vaccine being available. Control methods include biosecurity & prevention of other immuno-suppressive factors. Protocols for detecting & eradication, based on breaking verticle transmission being followed to control ALV. A & B subgroups are not equally efficient in this case.

R. L. Witter, (1997). *Aeta Veterinary* **45(3)** pp : 251-256

Special report by Primary Breeders Veterinary Round Table USA, *Poultry Times of India,* (1997). Vol. **2(14)** : 14-17.

"Headless mice born in genetic trials"

Scientists at the university of Texas USA have created mice without head, but with fully developed body by altering a gene. Out of 1000 such mice embryos tried, four grew to be born as head less mice but died soon after.

This research combined with cloning technology may lead to growing human organs in vitro (in head-less human) for transplantation. This would also by pass legal definition of what constitutes human embryo.

Times of India, Dt. 24.12.97.

FDA approves Rabipur in USA

Rabipur, a purified chick cell culture Rabies vaccine, manufactured by Hoechst Marrion Roussel's plant at Ankleshwar India, has been granted marketing clearance by U.S. Food & Administration.

Times of India, Dt.25.12.97.

New test for Bovine Spongy Form Encepalopathy (Mad cow Disease)

Dr. Stanley Prusiner (Noble Prize winner for Medicine 1997, for discovery of Prion Proteins causing BSE and related diseases) along with Fred Cohen and colleagues at university of California USA have developed a test for BSE (Mad cow disease) which uses genetically engineered mice having a bovine gene. The intra-cranial inoculation of BSE Suspected Material in such mice leads to development of BSE in 200 days, (if the test material is positive) which is much more shorter period than other similar tests using ordinary mice. However, the new test being still too slow can be used to (i)Assay infectivity of different parts of cow body (ii)To determine the age above which cows are dangerous as human food (iii)To determine whether blood of cows is infective to man(a hotly debate point at present)

Times of India, Dt. 24.12.97.

Starving Mosquitoes to death

Dov. Borovshy of university of Florida USA has genetically altered Chlorella algae to produce a substance that switches off Digestive Process in Mosquitoes. The mosquito which eat such algae further cannot take any food and they starve to death in three days. This method can be used as biological control of mosquitoes although the chlorella doesn't remain indefinitely in the environment and has to be propagated repeatedly.

Times of India, Dt.22.12.97.

Buffalo embryo cloned on

Scientist at National Dairy Research Institute

Karnal India, have fully cloned buffalo embryo by nucleus transfer technique and have grown it to 32 cell stage, suitable for implantation in a serrogate buffalo. Theoretically, each of the 64 cells (practically about 12-16) can be subjected to same process to create as many number of coloned embryos Dr. Alan Trounson & coworkers of Monarch university, Clayton, Australia have earlier cloned a line of 470 cattle embryos by similar process.

New Scientist, Dt.13.3.97 and *Statesman,* Calcutta 30.12.97

Anticoccidial effects of Linseed oil and Turmeric

Scientists at Parasitic Biology and epidermiology Lab Beltsville USA, have found that inclusion of 10% Linseed oil in corn soya feed of chicken reduced the scores of *Eimeria tenella* by 64%. Similarly inclusion of 1% turmeric in feed reduced *E maxima lesion* by 58%.

Poultry Times of India, 2(14) : 1 Nov. 1997

"Garlic as treatment for lead poisoning"

Dr. S. K. Dwivedi and his team from Indian Veterinary Research Institute (IVRI), Izatnagar, India have found that garlic effectively reduces lead toxicity. Simultaneous administration of garlic and lead in laboratory animals reduced severity of lead toxicity lesion prevented accumulation of lead in liver and enhanced its excretion through urine. Dr. O. S.Tomar Director of the institute has opined that this discovery would help in protecting persons like traffic police, industrial workers etc. besides animals who are exposed to lead poisoning.

Dr. Tomar further stated, other Medicinal plants viz Arjuna (*Terminatia arjuna*) Ashwagandharwa(*Withania somnifera*) and Amla are effective against toxicity by heavy metals. Use of these in food animals will reduce levels of heavy metals in beef, mutton & milk etc. rendering these items safer for human consumption.

Times of India, Dt. 16.1.98

Efficacy of Cefqinome for Treatment of Cows with Mastitis Experimentally Induced using *Escherichia coil*

Dr. Shpigel and his associates from Israel and Dr. Böttner from Hoechst Veterinar, Germany informed that Cefquinome the 4th generation antibiotic of Cephaloporin origin has got excellent effect both in Intra-muscular and Intra-mammary use in *Escherichia coil* mastitis in Dairy cattle.

A brief note of their findings are as under :

The efficacy of intra-mammary administered cefquinome was evaluated in experimental *Escherichia coil* mastitis in dairy cows. Forty-seven multiparous, Israeli Holstein cows in early lactation that produced at least 25 litre/ day of milk were used, and 400 to 750 cfu of *E. coil* were infused into two healthy quarters of each cow. Cows were randomly assigned to one of the following treatment groups (1) 75 mg of Cefquinome administred intra-mammary route, three times at 12-hrs. intervals, (2) 75 mg of cefquinome administred intra-mammarily route, three times at 12-hrs. intervals and 1 mg/kg of cefquinome administred intra-muscularly two times at a 24-hrs. interval, (3) 1 mg/kg of Cefquinome administred intra-muscularly two times at a 24-hrs. intervals and (4) 75 mg of Ampicillin and 200 mg of Cloxacillin Administred intra-mammary roule. Cefquinome (groups 2 and 3), significantly improved clinical recovery and return to milk production.

The bacteriological cure rates were

considerably and significantly higher for cows in the groups treated with Cefquinome than for cows in the group treated with Ampicillin. This study supported the efficacy of Cefquinome in the treatment of clinical coliform mastitis in dairy cows.

N. Y. Shpigel *et al.*, (1997).*J. Dairy Sci.*, **80** : 318 - 323

READERS' COLUMN

1. **Dr. R. K. Rajangam**
Professor & Head, University Training & Research Centre, Nagorcoil

Apart from the research articles, all of the trials using products of "Hoechst Roussel Vet", "A note on Animal Cloning by V. S. Narsapur and A. K. Datta was very informative, simple and illustrative.

2. **Dr. R. Kaliyappan**
Coimbatore, Tamil Nadu

It contains practical oriented and up-to-date knowledge sources and I am very impressed by this issue (9th issue) of ***"The Blue Cross Book"***. But sometimes it is not regularly supplied to institutions.

3. **Dr. D. K. Chetia**
District Animal Health & Veterinary Officer, Arunachal Pradesh

The article "Efficacy of a combined formulation of Fenbendazole and Oxyclozanide suspension in cattle" is liked by me. The article "Studies on Babesiosis in calves and its treatment with Berenil" is of great value to field staff. Such study reports if incorporated, adds great benefit.

4. **Dr. Kamal Kanti Bose**
Assistant Director of A.R.D., Institute of A.H. & Veterinary Biologicals (Research & Training), Calcutta

The article "A note on Animal Cloning" and "Transmissible Spongiform Encephalopatties in Animals (TSE's)" in ***"The Blue Cross Book"*** are the hot topics of discussions in the present day amongst all. Superb, please keep it up.

Suggestion : Articles on molecular biology and biotechnology should be invited.

I am very much interested to have your BCB (***The Blue Cross Book***) in a regular way so as to enable me to send articles for publication in future.

5. **Dr. R. Karim**
District A. H. & Veterinary Officer, Haiborgaon, Naigaon, Assam

I do not like this because there are spelling mistakes in some of the articles.

6. **Dr. D. N. Pandey**
Professor, Varanasi

It provides articles of applied nature in the days of information technology keeping view the increasing trend of pet keeping in India, clinical articles are needed in relation to canine practice.

7. **Dr. C. Venkata Subbarao**
Professor & Head, CCL, Kilpauk, Chennai

I suggest that every issue should come with a photograph of a canine breed with a brief history of its origin and other information as an additional news. If this gets a place in this valuable book it can be useful to veterinarians all over.

8. **Dr. Fakhruddin**
Senior Assistant Professor, (Vety. Medicine), Veterinary College, Bikaner

All the articles are broadly informative and also of good technical quality. Articles particularly "A note on Animal cloning", "Transmissible Spongiform Encephalopathies in Animals (TSE's) and "Cryptsoporidium and Cryptosporidiosis" are of the particular interest. Other articles, on drug trials are also useful. In all, *"The Blue Cross Book"* is a good, informative and useful publication for teachers, researchers and field veterinarians.

9. **Dr. R. Jayachandran**
Veterinary Surgeon, Govt. Veterinary Dispensary, Kottayam, Kerala

The articles on "Efficacy of Enrofloxacin against *Pasteurella multocida*" and "Chemotherapy of Myiasis in dogs with Ivermectin" were interesting. I would like to inform that I have already tried both these drugs in the above mentioned clinical cases and got successful results.

10. **Dr. Vivek Hinduras Ksheersagar**
Manager Inputs, Pune Dist. Co-op Milk Producers' Union Ltd., Katraj Dairy, Pune

It consists of interesting and variety of articles for knowledge updates. In research articles, practical field trials are to be taken for which the Dairy Milk Union can be made available.

READERS' COLUMN

11. Dr.Mahesh Parikh
Assistant Director of Animal Husbandry, Mahemdavad, Gujarat

It gives very practical research and useful information on Animal Husbandary and Veterinary practices. I want to send my articles entitled "Animals are our best friends"

12. Dr. Vijay Kumar Kapil
Veterinary Officer, Amritsar

In this book detailed information for veterinary products have been submitted. ***"The Blue Cross Book"*** for the month July 1997 is informative and solutions are provided for the problems faced by veterinarians in the field. This is an excellent book to update knowledge especially for practicing veterinarians.

I want to send articles entitled "Treatment of Calves and Cattle with Fenbendazole (Panacur®) shortly.

13. Dr. S. P. Deshpande
University Librarian, Dr. P. D. Krishi Vidyapeeth, Akola.

"The Blue Cross Book" which you have so kindly sent to me will be of very useful to the staff and students of this Agricaltural University and hence the same has been preserved in the Reading Hall of the University Library.

14. Dr. Ram Kishan Tanwar
Ph.D. (Vety. Medicine), Puran Ginnani, Bikaner, Rajasthan

It contains good clinical articles. I shall send articles entitled, 'Esophageal diverticulam on a cause of pulmonary emphysome in Buffalo'. Please invite articles on Pharmacokinetics of latest drug which will be of very useful.

The Blue Cross Book
for the Veterinary Profession

Hoechst Roussel Vet Pvt. Ltd.
Neeta Park, Building 1,
Airport Road, Yerwada,
Pune : 411 006.

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