

COMPARATIVE EFFICACY OF DIFFERENT TREATMENT PROTOCOLS IN THERAPEUTIC MANAGEMENT OF LEUKODERMA IN BUFFALOES

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ABSTRACT

The present study was conducted on 24 buffaloes having leukoderma. Three treatment protocols were evaluated. The cases were diagnosed on the basis of history and clinical signs and later confirmed by micro mineral status. Hematology and serum micro minerals status were evaluated before and after the treatment. Animals in Group 1, treated with injection copper glycinate of 1 ml/ 100 kg bwt S/C at interval of 10 days, showed 66.67% recovery in 60 days. Recovery in second group administered multi mineral injection 10 ml S/C at weekly interval was 83.33% in 60 days. In group third mineral mixture powder was administered 50 gram orally daily till 60 days with 16.67% recovery. Recovery was adjudged by the disappearance of the clinical signs. After end of treatment the clinical observations and analysis of all the findings of hematological and serum mineral values revealed that Group 2 had best therapeutic response.

Keywords: Leukoderma, buffaloes, *Bubalus bubalis*, treatment, treatment and serum micro minerals

INTRODUCTION

Leukoderma is the most common chronic depigmentation disorder affecting 1 to 2% of world population (Szczurko and Boon 2008). It includes loss of functioning melanocytes which causes the appearance of white patches on skin (Hong *et al.*, 2005). Leukoderma or vitiligo is best defined as an acquired, progressive disorder that selectively destroys some or all melanocytes residing in the interfollicular epidermis and occasionally in the hair follicle as well. Im *et al.* (1994) defined leukoderma as a pigmentry disorder of unknown cause characterized by depigmented patches due to destruction of melanocyte. The mechanism by which melanocytes are lost may be multiple but is not yet identified. Leukoderma has been recorded in dairy animals, mainly buffaloes (Gill and Gill, 1975). It is usually not harmful and causes no physiological pain to the animal. The present study was conducted to compare the therapeutic efficacy of different treatment protocols in leukodermic buffaloes.

MATERIALS AND METHODS

The research work was conducted in and around the Narendra Deva University of Agriculture and Technology, Kumarganj, Faizabad (U.P.). The buffaloes showing white patches on various part of body were selected for the study. All buffaloes were clinically examined. Respiration, pulse rate, heart rate, rectal temperature, feed intake, rumen motility was in normal range. Urination and defecation was normal. Fecal examination and skin scrapping were performed for the detection of any parasitic eggs/larvae in the feces and detection of demodex, mange and fungal infection respectively.

Blood samples were collected aseptically from jugular vein of each animal using sterilized disposable syringe and placed in two sets of sterile glass tubes, first set with anticoagulant, EDTA (1 mg/ml of blood) for hematological purposes and second set without anticoagulant for serum separation. The blood samples were analyzed for Hb, PCV, TLC, TEC by using standard methods (Benjamin, 1985). The serum micro minerals such as copper, iron, zinc and manganese were estimated on atomic absorption spectrophotometer using standard diagnostic kits at Department of Soil Science, College of Agriculture, G.B.P.U.A. and

T., Pantnagar U.S. Nagar Uttarakhand. Diagnosis of leucoderma in buffaloes were based on the basis of history, clinical signs, fecal examinations, and skin scrapping, blood and serum micro minerals analysis. The data generated was subjected to statically analysis as per method described by Snedecor and Cochran (1994).

TREATMENT GROUPS

The study was conducted on 24 buffaloes divided in 3 equal groups having 8 buffaloes each. Before the start of therapy deworming of all animals was done with Bolus Fenbendazole 7.5 mg/kg body weight. Animals having leukoderma were assigned different groups according to the following treatment plan. Recovery status of different treatment groups assessed as seen visibly and on the basis of hematological and serum mineral status pre and post therapy. After 10 days of deworming observation pertaining to change in skin colour, hair colour, milk yield and fertility status were recorded at 10 days interval during course of treatment. Blood sample was collected before and after treatment (day 0 and day 60). The blood sample was analyzed for Hb, PCV, TLC and TEC by using standard protocol. The serum

Group	No. of animals	Treatment protocol	Therapeutic regimen
1	8	Copper Glycinate Injection (copper 75 mg/ml)	1 ml/100 kg body weight s/c repeated at 10 days interval
2	8	Multi - mineral Injection (Zinc 60 mg/ml, manganese 10 mg/ ml, selenium 5 mg/ml, copper 15 mg/ml)	10 ml s/c repeated at 7 days interval
3	8	Oral Mineral Mixture (Zinc 960 mg/100 gm, manganese 150 mg/100 gm, selenium 1 mg/100 gm, copper 120 mg/100 gm)	50 gm orally SID

sample was used for analysis of micro minerals viz. Copper, iron, Zinc and Manganese. Buffaloes were divided in following groups in Table below.

RESULTS AND DISCUSSION

Animals in Group 1 treated with injection copper glycinate showed 66.67% recovery in 60 days. Remarkable improvement was observed in remaining 2 animals (33.33%) with drastic reduction in leukodermic patches. Buffaloes in Group 2 (administered with multi mineral injection 10 ml (total dose) subcutaneously at weekly interval) showed 83.33% recovery in 60 days (Figure 1 and 2). Recovery was noticed in the remaining one animal with only few leukodermic patches left at brisket region extending up to axilla. Recovery in animals supplemented with mineral mixture 50 gram orally daily till 60 days was

16.67%. In remaining 83.33% animals significant improvement was observed but complete recovery was absent. In the single recovered animal, reoccurrence was reported after one month of cessation of therapy.

After end of treatment the clinical observations and analysis of all the findings of hematological and serum mineral values revealed that Group 2 had best therapeutic response.

Hemoglobin (Hb) value increased significantly in Group 2 (Table 1). The increase in Hb level in all groups is suggestive of increased absorption and utilization of iron for RBC synthesis owing to improvement in serum copper level. In order for hemoglobin synthesis to occur, iron must be converted to ferric form before being incorporated in to the hemoglobin molecule. This process is accomplished by ceruloplasmin, which is a copper containing enzyme synthesized in liver for this purpose (Saenko *et al.*, 1994). In all the



Figure 1. Leukoderma in buffalo before treatment.



Figure 2. Buffalo recovered from Leukoderma after 60 days treatment.

three groups, non significant increase in the PCV value was noticed (Table 1). This increase in the PCV value might be due to increase in the total RBC count and increase in Hb value. Increased TEC value in all the groups was attributed to supplementation of iron, copper and zinc those play vital role in the synthesis of RBC. In all the three treatment groups these trace element were supplemented to the animals.

The serum copper level ($\mu\text{g}/\text{dl}$) increased from 49.85 ± 2.34 to 83.01 ± 1.03 , 48.99 ± 1.29 to 93.83 ± 1.00 and 50.62 ± 1.00 to 81.31 ± 1.31 respectively in Group 1, Group 2 and Group 3 (Table 2). The increase was significantly ($P<0.05$) higher. Maximum increase was seen in Group 2. The percentage increase in copper value in Group 1, 2 and 3 was 66.5%, 91.5% and 60% respectively.

The higher percentage increase in serum copper value in Group 2 resulted better response of the protocol followed by Group 1 and Group 3 respectively. Production of melanin depends upon tyrosinase, which is a copper containing enzyme. Described that copper ions are needed for activating enzyme tyrosinase, which is responsible for melanin synthesis. Higher serum copper level in Group 2 after treatment is responsible for best response.

Serum iron level increased non significantly in all groups (Table 2). The highest

percent increase in Group 2 was 5.8% followed by 5% and 3% respectively in Group 1 and Group 3. This is suggestive of better increase Hb, PCV, TEC concentration in Group 2 and 1 due to haemopoietic effect of iron. Melanin is an iron and sulphur containing brownish pigment so increased serum iron level resulted in best treatment protocol of leukoderma in Group 2. After treatment there was a non significant ($P<0.05$) increase in serum manganese level in all groups (Table 2). However the percentage increase was 20.7%, 27.5% and 22.8% in Group 1, 2 and 3. Maximum increase was seen in Group 2 because of subcutaneous administration of mineral have better bioavailability. Animals of Group 2 and Group 3 showed increased milk yield and better conception. Post therapy zinc level showed a non significant ($P<0.05$) increase with percentage increase of 19.6%, 26.8% and 21.2% in Group 1, 2 and 3 respectively (Table 2). A non significant change in serum zinc level was recorded by Singh and Randhawa (2008); Gapat *et al.* (2013). Zinc is involved in keratinization of epithelial tissue (Underwood, 1977) and appears to modify cell membrane by stabilizing the membrane structure thus reducing the peroxidation damage to the cell (Nockles and Blair, 1996).

Selenium, Zinc and Copper function to maintain low tissue concentration of reactive oxygen species and lipid hydroperoxide (Bettger

Table 1. Hematology of leukodermic buffaloes pre and after the treatment.

Parameters	Group 1		Group 2		Group 3	
	Pre	Post	Pre	Post	Pre	Post
Hb (gm/dl)	10.91 \pm 0.26	12.83 \pm 0.29	10.67 \pm 0.24	13.37* \pm 0.28	10.95 \pm 0.35	11.73 \pm 0.27
PCV (%)	32.30 \pm 0.60	39.12 \pm 0.42	32.40 \pm 1.03	40.90 \pm 1.25	33.1 \pm 1.01	35.15 \pm 1.10
TEC ($\times 10^6$ /mm ³)	6.79 \pm 0.170	7.37 \pm 0.21	6.70 \pm 0.16	7.74 \pm 0.28	6.89 \pm 0.25	7.29 \pm 0.19
TLC ($\times 10^3$ /mm ³)	10.52 \pm 0.28	10.49 \pm 0.29	10.98 \pm 0.44	11.8 \pm 0.19	10.43 \pm 0.32	10.96 \pm 0.28

*Significant difference ($P<0.05$).

Table 2. Serum mineral status of leukodermic buffaloes pre and after the treatment.

Parameters	Group 1		Group2		Group3	
	Pre	Post	Pre	Post	Pre	Post
Copper ($\mu\text{g}/\text{dl}$)	49.85 \pm 2.34	83.01* \pm 1.03	48.99 \pm 1.29	93.83* \pm 1.00	50.62 \pm 1.00	81.31* \pm 1.31
Iron ($\mu\text{g}/\text{dl}$)	100.11 \pm 1.88	105.14 \pm 0.96	100.50 \pm 0.96	106.32 \pm 1.44	99.82 \pm 0.62	103.2 \pm 1.0
Manganese ($\mu\text{g}/\text{dl}$)	51.68 \pm 1.17	62.41 \pm 1.07	51.41 \pm 1.16	65.56 \pm 1.44	52.68 \pm 1.15	64.65 \pm 1.41
Zinc ($\mu\text{g}/\text{dl}$)	74.10 \pm 2.08	88.64* \pm 1.68	72.92 \pm 2.44	92.45* \pm 1.35	75.55 \pm 1.80	91.60* \pm 2.31

*Significant difference ($P < 0.05$).

et al., 1979). These protective benefits result in improved immune response. Selenium is important for disease resistance because it is an integral component of the enzyme glutathione peroxidase (Arthur, 1985) which functions in the cytosol of the cell (Rice and Kennedy, 1986). This enzyme catalyzes the conversion of hydrogen peroxide to water and converts lipid peroxides to lipid alcohol and thus maintains low tissue concentrations of peroxides which if allowed to a mass in cell can severely damage the cells and tissues (Smith, 1988).

CONCLUSION

In the present study best therapeutic response was observed in Group 2 animals. This may be due to better absorption and direct utilization of mineral from subcutaneous depot. Oral mineral mixture have poor therapeutic response because in ruminant stomach there are many antagonists like Mo, S, Cd, and Pb etc which interfere with absorption of copper and other micronutrient from rumen.

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